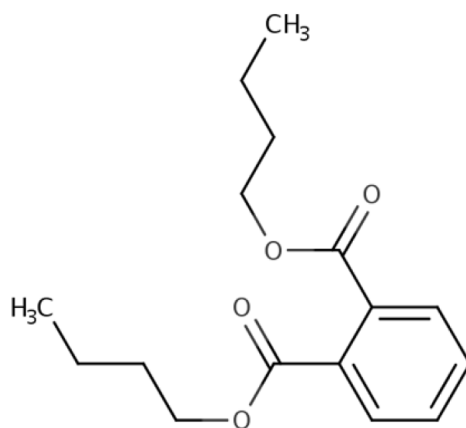

**Data Quality Evaluation Information for
Environmental Hazard for
Dibutyl Phthalate (DBP)
(1,2-Benzenedicarboxylic acid, 1,2-dibutyl ester)**

Systematic Review Support Document for the Risk Evaluation

CASRN: 84-74-2



December 2025

This supplemental file contains information regarding the data quality evaluation results relevant to the analysis of environmental hazard for the *Environmental Hazard Assessment for Dibutyl Phthalate (DBP)*. EPA conducted data quality evaluation based on author-reported descriptions and results; additional analyses (e.g., statistical analyses performed during data integration into the risk evaluation) potentially conducted by EPA are not contained in this supplemental file. EPA used the TSCA systematic review process described in the *Draft Systematic Review Protocol Supporting TSCA Risk Evaluations for Chemical Substances* (referred to hereafter as the '2021 Draft Systematic Review Protocol'). Any updated steps in the systematic review process since the publication of the 2021 Draft Systematic Review Protocol are described in the *Risk Evaluation for Dibutyl Phthalate (DBP) – Systematic Review Protocol*.

Separate data quality evaluation forms were used for different organisms as described in the PECO statement in Appendix H.5.11 of the 2021 Draft Systematic Review Protocol. Each health outcome was evaluated independently within a given reference; therefore, each reference may have more than one overall quality determination (OQD) to more appropriately reflect the quality of each health outcome and the respective hazard endpoints as described by the study authors. Some data evaluation forms have general additional comments presented adjacent to the OQD to add further context. No OQD is determined for each reference as a whole, if it contains data from more than one evidence stream. The study details and respective endpoints are organized by first the relevant habitat (i.e., aquatic vs. terrestrial), then taxa categories (e.g., vertebrates, invertebrates, vegetation) followed by taxonomic groups (e.g., fish, amphibian, mammalian, avian, worms, vascular plants), individual species, and finally exposure duration and health outcome (e.g., mortality) categories relevant to the endpoint being evaluated.

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HERO ID	Reference	Page
Dibutyl Phthalate		
Habitat: Aquatic (freshwater)		
Taxa: Vertebrates		
<i>Carassius auratus</i>		
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	<i>Hyalella azteca</i>	
679311	Call, D. J., Cox, D. A., Geiger, D. L., Genisot, K. I., Markee, T. P., Brooke, L. T., Polkinghorne, C. N., Vandeventer, F. A., Gorsuch, J. W., Robillard, K. A., Parkerton, T. F., Reiley, M. C., Ankley, G. T., Mount, D. R. (2001). An assessment of the toxicity of phthalate esters to freshwater benthos. 2. Sediment exposures. <i>Environmental Toxicology and Chemistry</i> 20(8):1805-1815.	444
679312	Call, D. J., Markee, T. P., Geiger, D. L., Brooke, L. T., Vandeventer, F. A., Cox, D. A., Genisot, K. I., Robillard, K. A., Gorsuch, J. W., Parkerton, T. F., Reiley, M. C., Ankley, G. T., Mount, D. R. (2001). An assessment of the toxicity of phthalate esters to freshwater benthos. 1. Aqueous exposures. <i>Environmental Toxicology and Chemistry</i> 20(8):1798-1804.	448
7325945	Lake Superior Research Institute, (1997). Sediment toxicity testing program for phthalate esters.	450
	<i>Ischnura verticalis</i>	
1334646	Jr, Mayer, F., Sanders, H. O., Walsh, D. F. (1973). Toxicity, residue dynamics, and reproductive effects of phthalate esters in aquatic invertebrates. <i>Environmental Research</i> 6(1):84-90.	464
	<i>Lumbriculus variegatus</i>	
679312	Call, D. J., Markee, T. P., Geiger, D. L., Brooke, L. T., Vandeventer, F. A., Cox, D. A., Genisot, K. I., Robillard, K. A., Gorsuch, J. W., Parkerton, T. F., Reiley, M. C., Ankley, G. T., Mount, D. R. (2001). An assessment of the toxicity of phthalate esters to freshwater benthos. 1. Aqueous exposures. <i>Environmental Toxicology and Chemistry</i> 20(8):1798-1804.	466
7325945	Lake Superior Research Institute, (1997). Sediment toxicity testing program for phthalate esters.	468
	<i>Macrobrachium rosenbergii</i>	
789598	Sung, H. H., Kao, W. Y., Su, Y. J. (2003). Effects and toxicity of phthalate esters to hemocytes of giant freshwater prawn, <i>Macrobrachium rosenbergii</i> . <i>Aquatic Toxicology</i> 64(1):25-37.	470
	<i>Orconectus nais</i>	

1334646	Jr, Mayer, F., Sanders, H. O., Walsh, D. F. (1973). Toxicity, residue dynamics, and reproductive effects of phthalate esters in aquatic invertebrates. <i>Environmental Research</i> 6(1):84-90.	476
	<i>Paleomonetes kadiakensis</i>	
1334646	Jr, Mayer, F., Sanders, H. O., Walsh, D. F. (1973). Toxicity, residue dynamics, and reproductive effects of phthalate esters in aquatic invertebrates. <i>Environmental Research</i> 6(1):84-90.	478
	<i>Paratanytarsus parthenogenetica</i>	
1321996	Adams, W. J., Biddinger, G. R., Robillard, K. A., Gorsuch, J. W. (1995). A summary of the acute toxicity of 14 phthalate esters to representative aquatic organisms. <i>Environmental Toxicology and Chemistry</i> 14(9):1569-1574.	480
	<i>Paratanytarsus parthenogenica</i>	
1316219	Bionomics,, EG&G (1984). Acute toxicity of twelve phthalate esters to <i>Paratanytarsus parthenogenica</i> (final report) report no BW-83-6-1424.	482
Taxa: Plants (Non-vascular)		
	<i>Algae</i>	
1332820	Kuang, Q. J., Zhao, W. Y., Cheng, S. P. (2003). Toxicity of dibutyl phthalate to algae. <i>Bulletin of Environmental Contamination and Toxicology</i> 71(3):602-608.	484
	<i>Chlorella emersonii</i>	
1333016	Melin, C., Egneus, H. (1983). Effects of di-n-butyl phthalate on growth and photosynthesis in algae and on isolated organelles from higher plants. <i>Physiologia Plantarum</i> 59(3):461-466.	486
1333016	Melin, C., Egneus, H. (1983). Effects of di-n-butyl phthalate on growth and photosynthesis in algae and on isolated organelles from higher plants. <i>Physiologia Plantarum</i> 59(3):461-466.	488
	<i>Chlorella pyrenoidosa</i>	
5433509	Gu, S., Zheng, H., Xu, Q., Sun, C., Shi, M., Wang, Z., Li, F. (2017). Comparative toxicity of the plasticizer dibutyl phthalate to two freshwater algae. <i>Aquatic Toxicology</i> 191:122-130.	490
6966450	Li, Z., Yi, X., Zhou, H., Chi, T., Li, W., Yang, K. (2020). Combined effect of polystyrene microplastics and dibutyl phthalate on the microalgae <i>Chlorella pyrenoidosa</i> . <i>Environmental Pollution</i> 257:113604.	494
	<i>Chlorella vulgaris</i>	
679344	Chi, J., Li, B., Wang, Q. Y., Liu, H. (2007). Influence of nutrient level on biodegradation and bioconcentration of phthalate acid esters in <i>Chlorella vulgaris</i> . <i>Journal of Environmental Science and Health, Part A: Toxic/Hazardous Substances & Environmental Engineering</i> 42(2):179-183.	496
	<i>Pseudokirchneriella subcapitata</i>	
789536	Jonsson, S., Baun, A. (2003). Toxicity of mono- and diesters of o-phthalic esters to a crustacean, a green alga, and a bacterium. <i>Environmental Toxicology and Chemistry</i> 22(12):3037-3043.	498
	<i>Scenedesmus obliquus</i>	

5433509	Gu, S., Zheng, H., Xu, Q., Sun, C., Shi, M., Wang, Z., Li, F. (2017). Comparative toxicity of the plasticizer dibutyl phthalate to two freshwater algae. <i>Aquatic Toxicology</i> 191:122-130.	500
5551982	Huang, G. L., Sun, H. W., Song, Z. H. (1999). Interactions between dibutyl phthalate and aquatic organisms. <i>Bulletin of Environmental Contamination and Toxicology</i> 63(6):759-765.	504
1332820	Kuang, Q. J., Zhao, W. Y., Cheng, S. P. (2003). Toxicity of dibutyl phthalate to algae. <i>Bulletin of Environmental Contamination and Toxicology</i> 71(3):602-608.	506
5551982	Huang, G. L., Sun, H. W., Song, Z. H. (1999). Interactions between dibutyl phthalate and aquatic organisms. <i>Bulletin of Environmental Contamination and Toxicology</i> 63(6):759-765.	508
1332820	Kuang, Q. J., Zhao, W. Y., Cheng, S. P. (2003). Toxicity of dibutyl phthalate to algae. <i>Bulletin of Environmental Contamination and Toxicology</i> 71(3):602-608.	510
<i>Scenedesmus sp.</i>		
6967432	Cunha, C., Paulo, J., Faria, M., Kaufmann, M., Cordeiro, N. (2019). Ecotoxicological and biochemical effects of environmental concentrations of the plastic-bond pollutant dibutyl phthalate on <i>Scenedesmus sp.</i> <i>Aquatic Toxicology</i> 215:105281.	514
<i>Selenastrum capricornutum</i>		
1323217	Adachi, A., Asa, K., Okano, T. (2006). Efficiency of rice bran for removal of di-n-butyl phthalate and its effect on the growth inhibition of <i>Selenastrum capricornutum</i> by di-n-butyl phthalate. <i>Bulletin of Environmental Contamination and Toxicology</i> 76(5):877-882.	521
1321996	Adams, W. J., Biddinger, G. R., Robillard, K. A., Gorsuch, J. W. (1995). A summary of the acute toxicity of 14 phthalate esters to representative aquatic organisms. <i>Environmental Toxicology and Chemistry</i> 14(9):1569-1574.	525
1316196	Bionomics,, Springborn (1984). FYI Submission: Toxicity of fourteen phthalate esters to the freshwater green alga <i>Selenastrum capricornutum</i> .	527
1333016	Melin, C., Egneus, H. (1983). Effects of di-n-butyl phthalate on growth and photosynthesis in algae and on isolated organelles from higher plants. <i>Physiologia Plantarum</i> 59(3):461-466.	529
Taxa: Plants (Vascular)		
<i>Hordeum vulgare</i>		
1333016	Melin, C., Egneus, H. (1983). Effects of di-n-butyl phthalate on growth and photosynthesis in algae and on isolated organelles from higher plants. <i>Physiologia Plantarum</i> 59(3):461-466.	531
<i>Lemna minor</i>		
1323213	Huang, Q., Wang, Q., Tan, W., Song, G., Lu, G., Li, F. (2006). Biochemical responses of two typical duckweeds exposed to dibutyl phthalate. <i>Journal of Environmental Science and Health, Part A: Toxic/Hazardous Substances & Environmental Engineering</i> 41(8):1615-1626.	533
<i>Spinacea oleraceae</i>		
1333016	Melin, C., Egneus, H. (1983). Effects of di-n-butyl phthalate on growth and photosynthesis in algae and on isolated organelles from higher plants. <i>Physiologia Plantarum</i> 59(3):461-466.	537
<i>Spirodela polyrhiza</i>		

1323213	Huang, Q., Wang, Q., Tan, W., Song, G., Lu, G., Li, F. (2006). Biochemical responses of two typical duckweeds exposed to dibutyl phthalate. Journal of Environmental Science and Health, Part A: Toxic/Hazardous Substances & Environmental Engineering 41(8):1615-1626.	539
<i>Triticum sp.</i>		
3515118	Gao, M., Dong, Y., Zhang, Z., Song, W., Qi, Y. (2017). Growth and antioxidant defense responses of wheat seedlings to di-n-butyl phthalate and di (2-ethylhexyl) phthalate stress. Chemosphere 172(Elsevier):418-428.	543
3515118	Gao, M., Dong, Y., Zhang, Z., Song, W., Qi, Y. (2017). Growth and antioxidant defense responses of wheat seedlings to di-n-butyl phthalate and di (2-ethylhexyl) phthalate stress. Chemosphere 172(Elsevier):418-428.	545
Taxa: Other		
<i>fungus</i>		
1323196	Liang, W., Deng, J. Q., Zhan, F. C., Wu, Z. B. (2009). Effects of constructed wetland system on the removal of dibutyl phthalate (DBP). Microbiological Research 164(2):206-211.	551
Habitat: Aquatic (marine)		
Taxa: Vertebrates		
<i>sheepshead minnow (Cyprinodon variegatus)</i>		
1316224	Bionomics,, Springborn (1984). Acute toxicity of thirteen phthalate esters to the sheepshead minnow (Cyprinodon variegatus) (final report).	553
Taxa: Invertebrates		
<i>Animalia</i>		
5495608	Tagatz, M. E., Deans, C. H., Moore, J. C., Plaia, G. R. (1983). Alterations in composition of field-developed and laboratory-developed estuarine benthic communities exposed to di-normal-butyl phthalate. Aquatic Toxicology 3(3):239-248.	555
<i>Annelida</i>		
5495608	Tagatz, M. E., Deans, C. H., Moore, J. C., Plaia, G. R. (1983). Alterations in composition of field-developed and laboratory-developed estuarine benthic communities exposed to di-normal-butyl phthalate. Aquatic Toxicology 3(3):239-248.	559
<i>Artemia salina</i>		
5569571	Sugawara, N. (1974). Effect of phthalate esters on shrimp. Bulletin of Environmental Contamination and Toxicology 12(4):421-424.	563
<i>Arthropoda</i>		
5495608	Tagatz, M. E., Deans, C. H., Moore, J. C., Plaia, G. R. (1983). Alterations in composition of field-developed and laboratory-developed estuarine benthic communities exposed to di-normal-butyl phthalate. Aquatic Toxicology 3(3):239-248.	567
<i>Chordata</i>		

5495608	Tagatz, M. E., Deans, C. H., Moore, J. C., Plaia, G. R. (1983). Alterations in composition of field-developed and laboratory-developed estuarine benthic communities exposed to di-normal-butyl phthalate. <i>Aquatic Toxicology</i> 3(3):239-248.	571
<i>Coelenterata</i>		
5495608	Tagatz, M. E., Deans, C. H., Moore, J. C., Plaia, G. R. (1983). Alterations in composition of field-developed and laboratory-developed estuarine benthic communities exposed to di-normal-butyl phthalate. <i>Aquatic Toxicology</i> 3(3):239-248.	575
<i>Echinodermata</i>		
5495608	Tagatz, M. E., Deans, C. H., Moore, J. C., Plaia, G. R. (1983). Alterations in composition of field-developed and laboratory-developed estuarine benthic communities exposed to di-normal-butyl phthalate. <i>Aquatic Toxicology</i> 3(3):239-248.	579
<i>Haliotis diversicolor supertexta</i>		
697762	Liu, Y., Guan, Y., Yang, Z., Cai, Z., Mizuno, T., Tsuno, H., Zhu, W., Zhang, X. (2009). Toxicity of seven phthalate esters to embryonic development of the abalone <i>Haliotis diversicolor supertexta</i> . <i>Ecotoxicology</i> 18(3):293-303.	583
1322103	Yang, Z. H., Zhang, X. J., Cai, Z. H. (2009). Toxic effects of several phthalate esters on the embryos and larvae of abalone <i>Haliotis diversicolor supertexta</i> . <i>Chinese Journal of Oceanology and Limnology</i> 27(2):395-399.	585
1249532	Zhou, J., Cai, Z. H., Xing, K. Z. (2011). Potential mechanisms of phthalate ester embryotoxicity in the abalone <i>Haliotis diversicolor supertexta</i> . <i>Environmental Pollution</i> 159(5):1114-1122.	587
1322103	Yang, Z. H., Zhang, X. J., Cai, Z. H. (2009). Toxic effects of several phthalate esters on the embryos and larvae of abalone <i>Haliotis diversicolor supertexta</i> . <i>Chinese Journal of Oceanology and Limnology</i> 27(2):395-399.	593
<i>Mollusca</i>		
5495608	Tagatz, M. E., Deans, C. H., Moore, J. C., Plaia, G. R. (1983). Alterations in composition of field-developed and laboratory-developed estuarine benthic communities exposed to di-normal-butyl phthalate. <i>Aquatic Toxicology</i> 3(3):239-248.	595
<i>Mysidopsis bahia</i>		
1321996	Adams, W. J., Biddinger, G. R., Robillard, K. A., Gorsuch, J. W. (1995). A summary of the acute toxicity of 14 phthalate esters to representative aquatic organisms. <i>Environmental Toxicology and Chemistry</i> 14(9):1569-1574.	599
1316220	Bionomics,, EG&G (1984). Acute toxicity of twelve phthalate esters to mysid shrimp (<i>Mysidopsis bahia</i>).	601
<i>PALAEMONETES PUGIO</i>		
1333217	RB, Laughlin, J. R., Neff, J. M., Hung, Y. C., Goodwin, T. C., Giam, C. S. (1978). The effects of three phthalate esters on the larval development of the grass shrimp <i>Palaemonetes pugio</i> (Holthuis). <i>Water, Air, and Soil Pollution</i> 9(3):323-336.	603
<i>Palaemonetes pugio</i>		
5557723	Clark, J. R., JR, Patrick, J. M., Jr, More, J. C., Lores, E. M. (1987). Waterborne and sediment-source toxicities of six organic chemicals to grass shrimp (<i>Palaemonetes pugio</i>) and amphioxus (<i>Branchiostoma caribaeum</i>). <i>Archives of Environmental Contamination and Toxicology</i> 16(4):401-408.	609

5557723	Clark, J. R., JR, Patrick, J. M., Jr, More, J. C., Lores, E. M. (1987). Waterborne and sediment-source toxicities of six organic chemicals to grass shrimp (<i>Palaemonetes pugio</i>) and amphioxus (<i>Branchiostoma caribaeum</i>). Archives of Environmental Contamination and Toxicology 16(4):401-408.	611
	<i>Rhynchocoela</i>	
5495608	Tagatz, M. E., Deans, C. H., Moore, J. C., Plaia, G. R. (1983). Alterations in composition of field-developed and laboratory-developed estuarine benthic communities exposed to di-normal-butyl phthalate. Aquatic Toxicology 3(3):239-248.	613
Taxa: Plants (Non-vascular)		
	<i>Dunaliella parva</i>	
790153	Acey, R., Healy, P., Unger, T. F., Ford, C. E., Hudson, R. A. (1987). Growth and aggregation behavior of representative phytoplankton as affected by the environmental contaminant di-n-butyl phthalate. Bulletin of Environmental Contamination and Toxicology 39(1):1-6.	615
	<i>Karenia brevis</i>	
3230225	Liu, N., Wen, F., Li, F., Zheng, X., Liang, Z., Zheng, H. (2016). Inhibitory mechanism of phthalate esters on <i>Karenia brevis</i> . Chemosphere 155:498-508.	617
	<i>Skeletonema costatum</i>	
789981	Medlin, L. K. (1980). Effects of di-n-butyl phthalate and salinity on the growth of the diatom <i>Skeletonema costatum</i> . Bulletin of Environmental Contamination and Toxicology 25(1):75-78.	623
	<i>Synechococcus lividus</i>	
790153	Acey, R., Healy, P., Unger, T. F., Ford, C. E., Hudson, R. A. (1987). Growth and aggregation behavior of representative phytoplankton as affected by the environmental contaminant di-n-butyl phthalate. Bulletin of Environmental Contamination and Toxicology 39(1):1-6.	631
	<i>Thalassioria pseudomona</i>	
790153	Acey, R., Healy, P., Unger, T. F., Ford, C. E., Hudson, R. A. (1987). Growth and aggregation behavior of representative phytoplankton as affected by the environmental contaminant di-n-butyl phthalate. Bulletin of Environmental Contamination and Toxicology 39(1):1-6.	633
Habitat: Aquatic (brackish)		
Taxa: Vertebrates		
	<i>Cyprinodon variegatus</i>	
789995	Wofford, H. W., Wilsey, C. D., Neff, G. S., Giam, C. S., Neff, J. M. (1981). Bioaccumulation and metabolism of phthalate esters by oysters, brown shrimp, and sheepshead minnows. Ecotoxicology and Environmental Safety 5(2):202-210.	635
	<i>sheepshead minnow (Cyprinodon variegatus)</i>	
1316224	Bionomics,, Springborn (1984). Acute toxicity of thirteen phthalate esters to the sheepshead minnow (<i>Cyprinodon variegatus</i>) (final report).	637
	<i>Xenopus laevis</i>	

128004	Lee, S. K., Veeramachaneni, R., D.N. (2005). Subchronic exposure to low concentrations of di-n-butyl phthalate disrupts spermatogenesis in <i>Xenopus laevis</i> frogs. <i>Toxicological Sciences</i> 84(2):394-407.	639
Taxa: Invertebrates		
	<i>Artemia salina</i>	
1315792	Sugawara, N. (1974). Toxic effect of a normal series of phthalate esters on the hatching of shrimp eggs. <i>Toxicology and Applied Pharmacology</i> 30(1):87-89.	647
	<i>Crassostrea virginica</i>	
789995	Wofford, H. W., Wilsey, C. D., Neff, G. S., Giam, C. S., Neff, J. M. (1981). Bioaccumulation and metabolism of phthalate esters by oysters, brown shrimp, and sheepshead minnows. <i>Ecotoxicology and Environmental Safety</i> 5(2):202-210.	649
	<i>Mysidopsis bahia</i>	
1316220	Bionomics,, EG&G (1984). Acute toxicity of twelve phthalate esters to mysid shrimp (<i>Mysidopsis bahia</i>).	651
	<i>Nitocra spinipes</i>	
51937	Linden, E., Bengtsson, B. E., Svanberg, O., Sundstrom, G. (1979). The acute toxicity of 78 chemicals and pesticide formulations against two brackish water organisms, the bleak (<i>Alburnus alburnus</i>) and the harpacticoid <i>Nitocra spinipes</i> . <i>Chemosphere</i> 8(11-12):843-851.	653
	<i>Penaecus aztecus</i>	
789995	Wofford, H. W., Wilsey, C. D., Neff, G. S., Giam, C. S., Neff, J. M. (1981). Bioaccumulation and metabolism of phthalate esters by oysters, brown shrimp, and sheepshead minnows. <i>Ecotoxicology and Environmental Safety</i> 5(2):202-210.	655
Habitat: Terrestrial		
Taxa: Vertebrates		
	<i>Capra hircus</i>	
1332948	Cornell University, (1931). Report upon the toxicity of plasticizers.	657
	<i>Coturnix coturnix</i>	
2346127	Bello, U. M., Madekurozwa, M.,-C, Groenewald, H. B., Aire, T. A., Arukwe, A. (2014). The effects on steroidogenesis and histopathology of adult male Japanese quails (<i>Coturnix coturnix japonica</i>) testis following pre-pubertal exposure to di(n-butyl) phthalate (DBP). <i>Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology</i> 166:24-33.	671
	<i>Gallus domesticus</i>	
1332945	DuPont, (1949). Toxicity of dibutyl phthalate.	677

94541	Korhonen, A., Hemminki, K., Vainio, H. (1983). Embryotoxic effects of phthalic acid derivatives, phosphates and aromatic oils used in the manufacturing of rubber on three day chicken embryos. <i>Drug and Chemical Toxicology</i> 6(2):191-207.	681
	<i>Gallus gallus</i>	
1332948	Cornell University, (1931). Report upon the toxicity of plasticizers.	685
1332948	Cornell University, (1931). Report upon the toxicity of plasticizers.	695
	<i>Gallus gallus domesticus</i>	
1249807	Abdul-Ghani, S., Yanai, J., Abdul-Ghani, R., Pinkas, A., Abdeen, Z. (2012). The teratogenicity and behavioral teratogenicity of di(2-ethylhexyl) phthalate (DEHP) and di-butyl Phthalate (DBP) in a chick model. <i>Neurotoxicology and Teratology</i> 34(1):56-62.	711
	<i>Streptopelia risoria</i>	
681729	Peakall, D. B. (1974). Effects of di-n-butyl and di-2-ethylhexyl phthalate on the eggs of ring doves. <i>Bulletin of Environmental Contamination and Toxicology</i> 12(6):698-702.	717
Taxa: Invertebrates		
	<i>Caenorhabditis elegans</i>	
5043459	Shin, N., Cuenca, L., Karthikraj, R., Kannan, K., Colaiácovo, M. P. (2019). Assessing effects of germline exposure to environmental toxicants by high-throughput screening in <i>C. elegans</i> . <i>PLoS Genetics</i> 15(2):e1007975.	719
2215375	Tseng, I. L., Yang, Y. F., Yu, C. W., Li, W. H., Liao, C., V.H. (2013). Phthalates induce neurotoxicity affecting locomotor and thermotactic behaviors and AFD neurons through oxidative stress in <i>Caenorhabditis elegans</i> . <i>PLoS ONE</i> 8(12):e82657.	727
	<i>Dermatophagoides farinae</i>	
485854	Kang, S. W., Kim, H. K., Lee, W. J., Ahn, Y. J. (2006). Toxicity of bisabolangelone from <i>Ostericum koreanum</i> roots to <i>Dermatophagoides farinae</i> and <i>Dermatophagoides pteronyssinus</i> (Acari : Pyroglyphidae). <i>Journal of Agricultural and Food Chemistry</i> 54(10):3547-3550.	735
1332803	Kim, H. K., Tak, J. H., Ahn, Y. J. (2004). Acaricidal activity of <i>Paeonia suffruticosa</i> root bark-derived compounds against <i>Dermatophagoides farinae</i> and <i>Dermatophagoides pteronyssinus</i> (Acari: Pyroglyphidae). <i>Journal of Agricultural and Food Chemistry</i> 52(26):7857-7861.	739
1323180	Kim, H. K., Yun, Y. K., Ahn, Y. J. (2008). Fumigant toxicity of cassia bark and cassia and cinnamon oil compounds to <i>Dermatophagoides farinae</i> and <i>Dermatophagoides pteronyssinus</i> (Acari: Pyroglyphidae). <i>Experimental and Applied Acarology</i> 44(1):1-9.	743
1341977	Kim, H. K., Yun, Y. K., Ahn, Y. J. (2007). Toxicity of atractylon and atractylenolide III identified in <i>Atractylodes ovata</i> rhizome to <i>Dermatophagoides farinae</i> and <i>Dermatophagoides pteronyssinus</i> . <i>Journal of Agricultural and Food Chemistry</i> 55(15):6027-6031.	745
788260	Wang, Z., Kim, H. K., Tao, W., Wang, M., Ahn, Y. J. (2011). Contact and fumigant toxicity of cinnamaldehyde and cinnamic acid and related compounds to <i>Dermatophagoides farinae</i> and <i>Dermatophagoides pteronyssinus</i> (Acari: Pyroglyphidae). <i>Journal of Medical Entomology</i> 48(2):366-371.	747
	<i>Dermatophagoides pteronyssinus</i>	
485854	Kang, S. W., Kim, H. K., Lee, W. J., Ahn, Y. J. (2006). Toxicity of bisabolangelone from <i>Ostericum koreanum</i> roots to <i>Dermatophagoides farinae</i> and <i>Dermatophagoides pteronyssinus</i> (Acari : Pyroglyphidae). <i>Journal of Agricultural and Food Chemistry</i> 54(10):3547-3550.	749

1332803	Kim, H. K., Tak, J. H., Ahn, Y. J. (2004). Acaricidal activity of Paeonia suffruticosa root bark-derived compounds against Dermatophagoides farinae and Dermatophagoides pteronyssinus (Acari: Pyroglyphidae). Journal of Agricultural and Food Chemistry 52(26):7857-7861.	753
1323180	Kim, H. K., Yun, Y. K., Ahn, Y. J. (2008). Fumigant toxicity of cassia bark and cassia and cinnamon oil compounds to Dermatophagoides farinae and Dermatophagoides pteronyssinus (Acari: Pyroglyphidae). Experimental and Applied Acarology 44(1):1-9.	757
1341977	Kim, H. K., Yun, Y. K., Ahn, Y. J. (2007). Toxicity of atractylon and atractylenolide III identified in Atractylodes ovata rhizome to Dermatophagoides farinae and Dermatophagoides pteronyssinus. Journal of Agricultural and Food Chemistry 55(15):6027-6031.	759
788260	Wang, Z., Kim, H. K., Tao, W., Wang, M., Ahn, Y. J. (2011). Contact and fumigant toxicity of cinnamaldehyde and cinnamic acid and related compounds to Dermatophagoides farinae and Dermatophagoides pteronyssinus (Acari: Pyroglyphidae). Journal of Medical Entomology 48(2):366-371.	761
<i>Drosophila melanogaster</i>		
2510760	Misra, S., Singh, A., Ch, R., Sharma, V., Mudiam, R., M.K., Ram, K. R. (2014). Identification of Drosophila-based endpoints for the assessment and understanding of xenobiotic-mediated male reproductive adversities. Toxicological Sciences 141(1):278-291.	763
2510760	Misra, S., Singh, A., Ch, R., Sharma, V., Mudiam, R., M.K., Ram, K. R. (2014). Identification of Drosophila-based endpoints for the assessment and understanding of xenobiotic-mediated male reproductive adversities. Toxicological Sciences 141(1):278-291.	767
3350270	Williams, M. J., Wiemerslage, L., Gohel, P., Kheder, S., Kothegala, L. V., Schioth, H. B. (2016). Dibutyl Phthalate Exposure Disrupts Evolutionarily Conserved Insulin and Glucagon-Like Signaling in Drosophila Males. Endocrinology 157(6):2309-2321.	769
2510760	Misra, S., Singh, A., Ch, R., Sharma, V., Mudiam, R., M.K., Ram, K. R. (2014). Identification of Drosophila-based endpoints for the assessment and understanding of xenobiotic-mediated male reproductive adversities. Toxicological Sciences 141(1):278-291.	779
2510760	Misra, S., Singh, A., Ch, R., Sharma, V., Mudiam, R., M.K., Ram, K. R. (2014). Identification of Drosophila-based endpoints for the assessment and understanding of xenobiotic-mediated male reproductive adversities. Toxicological Sciences 141(1):278-291.	787
<i>Eisenia fetida</i>		
2816887	Du, L., Li, G., Liu, M., Li, Y., Yin, S., Zhao, J. (2015). Biomarker responses in earthworms (Eisenia fetida) to soils contaminated with di-n-butyl phthalates. Environmental Science and Pollution Research 22(6):4660-4669.	791
3625226	Neuhauser, E. F., Loehr, R. C., Malecki, M. R., Milligan, D. L., Durkin, P. R. (1985). The toxicity of selected organic chemicals to the earthworm Eisenia fetida. Journal of Environmental Quality 14(3):383-388.	793
2816887	Du, L., Li, G., Liu, M., Li, Y., Yin, S., Zhao, J. (2015). Biomarker responses in earthworms (Eisenia fetida) to soils contaminated with di-n-butyl phthalates. Environmental Science and Pollution Research 22(6):4660-4669.	795
2816887	Du, L., Li, G., Liu, M., Li, Y., Yin, S., Zhao, J. (2015). Biomarker responses in earthworms (Eisenia fetida) to soils contaminated with di-n-butyl phthalates. Environmental Science and Pollution Research 22(6):4660-4669.	797
<i>Eutrombicula hirsti</i>		
1341925	Frances, S. P. (1994). Response of a chigger, eutrombicula-hirsti (acari, trombiculidae) to repellent and toxicant compounds in the laboratory. Journal of Medical Entomology 31(4):628-630.	799
<i>Folsomia fimetaria</i>		
789786	Jensen, J., Langevelde, van, J., Pritzl, G., Krogh, P. H. (2001). Effects of di(2-ethylhexyl) phthalate and dibutyl phthalate on the collembolan Folsomia fimetaria. Environmental Toxicology and Chemistry 20(5):1085-1091.	803

789786	Jensen, J., Langevelde, van, J., Pritzl, G., Krogh, P. H. (2001). Effects of di(2-ethylhexyl) phthalate and dibutyl phthalate on the collembolan <i>Folsomia fimetaria</i> . <i>Environmental Toxicology and Chemistry</i> 20(5):1085-1091.	805
789786	Jensen, J., Langevelde, van, J., Pritzl, G., Krogh, P. H. (2001). Effects of di(2-ethylhexyl) phthalate and dibutyl phthalate on the collembolan <i>Folsomia fimetaria</i> . <i>Environmental Toxicology and Chemistry</i> 20(5):1085-1091.	809
	<i>Lasius niger</i>	
2347468	Lenoir, A., Touchard, A., Devers, S., Christidès, J. P., Boulay, R., Cuvillier-Hot, V. (2014). Ant cuticular response to phthalate pollution. <i>Environmental Science and Pollution Research</i> 21(23):13446-13451.	813
	<i>Meloidogyne incognita</i>	
3350275	Yang, G., Zhou, B., Zhang, X., Zhang, Z., Wu, Y., Zhang, Y., Lü, S., Zou, Q., Gao, Y., Teng, L. (2016). Effects of Tomato Root Exudates on <i>Meloidogyne incognita</i> . <i>PLoS ONE</i> 11(4):e0154675.	815
3350275	Yang, G., Zhou, B., Zhang, X., Zhang, Z., Wu, Y., Zhang, Y., Lü, S., Zou, Q., Gao, Y., Teng, L. (2016). Effects of Tomato Root Exudates on <i>Meloidogyne incognita</i> . <i>PLoS ONE</i> 11(4):e0154675.	819
3350275	Yang, G., Zhou, B., Zhang, X., Zhang, Z., Wu, Y., Zhang, Y., Lü, S., Zou, Q., Gao, Y., Teng, L. (2016). Effects of Tomato Root Exudates on <i>Meloidogyne incognita</i> . <i>PLoS ONE</i> 11(4):e0154675.	821
	<i>Spodoptera frugiperda</i>	
2219889	Filho, D.N., I., Vieceli, N. C., Cardoso, E. M., Lovatel, E. R., Gonzatti, C. F., Marzotto, J. A., Montezano, D. G., Specht, A. (2013). Two generations of fall armyworm (Lepidoptera: Noctuidae) contamination by di-n-butylphthalate. <i>Journal of Toxicology and Environmental Health, Part A: Current Issues</i> 76(16):973-977.	823
	<i>Tyrophagus putrescentiae</i>	
1323221	Tak, J. H., Kim, H. K., Lee, S. H., Ahn, Y. J. (2006). Acaricidal activities of paeonol and benzoic acid from <i>Paeonia suffruticosa</i> root bark and monoterpenoids against <i>Tyrophagus putrescentiae</i> (Acari: Acaridae). <i>Pest Management Science</i> 62(6):551-557.	833
Taxa: Plants (Vascular)		
	<i>Achilla millefolium</i>	
9430481	Lã,Kke, H., Rasmussen, L. (1983). Phytotoxicological effects of Di-(2-ethyl hexyl)-phthalate and Di-n-butyl-phthalate on higher plants in laboratory and field experiments. <i>Environmental Pollution Series A: Ecological and Biological</i> 32(3):179-199.	841
	<i>Avena sativa</i>	
5551990	Isogai, Y., Komoda, Y., Okamoto, T. (1972). Biological activities of n-butyl phthalate and its analogous compounds on various bioassays of plant growth regulators. <i>Scientific papers of the College of General Education, University of Tokyo</i> 22(2):129-135.	849
2915866	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. <i>Frontiers of Environmental Science & Engineering</i> 9(2):259-268.	851
	<i>Brassica campestris</i>	
1302103	Dueck, T. A., Dijk, Van, C. J., David, F., Scholz, N., Vanwalleghe, F. (2003). Chronic effects of vapour phase di-n-butyl phthalate (DBP) on six plant species. <i>Chemosphere</i> 53(8):911-920.	857
	<i>Brassica napus</i>	

4829418	Kong, X., Jin, D., Jin, S., Wang, Z., Yin, H., Xu, M., Deng, Y. (2018). Responses of bacterial community to dibutyl phthalate pollution in a soil-vegetable ecosystem. <i>Journal of Hazardous Materials</i> 353(Elsevier):142-150.	861
	<i>Brassica oleracea</i>	
5678863	Hardwick, R. C., Cole, R. A., Fyfield, T. P. (1984). Injury to and death of cabbage (<i>brassica-oleracea</i>) seedlings caused by vapors of di butyl phthalate emitted from certain plastics. <i>Annals of Applied Biology</i> 105(1):97-105.	867
	<i>Brassica parachinensis</i>	
5043543	Zhao, H. M., Huang, H. B., Luo, Y. M., Huang, C. Q., Du, H., Xiang, L., Cai, Q. Y., Li, Y. W., Li, H., Mo, C. H., He, Z. (2018). Differences in root physiological and proteomic responses to dibutyl phthalate (DBP) exposure between low- and high-DBP accumulation cultivars of <i>Brassica parachinensis</i> . <i>Journal of Agricultural and Food Chemistry</i> 66(51):13541-13551.	879
	<i>Brassica parachinensis L.</i>	
3070947	Zhao, H. M., Du, H., Xiang, L., Li, Y. W., Li, H., Cai, Q. Y., Mo, C. H., Cao, G., Wong, M. H. (2016). Physiological differences in response to di-n-butyl phthalate (DBP) exposure between low- and high-DBP accumulating cultivars of Chinese flowering cabbage (<i>Brassica parachinensis L.</i>). <i>Environmental Pollution</i> 208(Pt B):840-849.	887
	<i>Brassica rapa</i>	
1296241	Liao, C. S., Yen, J. H., Wang, Y. S. (2009). Growth inhibition in Chinese cabbage (<i>Brassica rapa</i> var. <i>chinensis</i>) growth exposed to di-n-butyl phthalate. <i>Journal of Hazardous Materials</i> 163(2-3):625-631.	899
1298079	Liao, C. S., Yen, J. H., Wang, Y. S. (2006). Effects of endocrine disruptor di-n-butyl phthalate on the growth of Bok choy (<i>Brassica rapa</i> subsp. <i>chinensis</i>). <i>Chemosphere</i> 65(10):1715-1722.	905
5605728	Zhu, F., Zhu, C., Chen, N., Zhou, D., Gao, J. (2018). Will spent mushroom substrate application affect the dissipation and plant uptake of phthalate esters?. <i>Journal of Soils and Sediments</i> 18(4):1579-1589.	911
	<i>Browallia speciosa</i>	
1333234	Virgin, H. I., A-M, Holst, Morner, J. (1981). Effect of di-n-butylphthalate on the carotenoid synthesis in green plants. <i>Physiologia Plantarum</i> 53(2):158-163.	913
	<i>Carica papaya</i>	
5433168	Kannan, S., Ramani, S. (1987). Mechanisms of fe-deficiency tolerance in crop cultivars - effects of dibutyl phthalate and caffeic acid on fe-chlorosis recovery. <i>Journal of Plant Nutrition</i> 10(9-16):1051-1058.	919
	<i>Cucumis sativus</i>	
5551990	Isogai, Y., Komoda, Y., Okamoto, T. (1972). Biological activities of n-butyl phthalate and its analogous compounds on various bioassays of plant growth regulators. <i>Scientific papers of the College of General Education, University of Tokyo</i> 22(2):129-135.	925
2915866	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. <i>Frontiers of Environmental Science & Engineering</i> 9(2):259-268.	927
3502464	Wang, L., Sun, X., Chang, Q., Tao, Y., Wang, L., Dong, J., Lin, Y., Zhang, Y. (2016). Effect of di-n-butyl phthalate (DBP) on the fruit quality of cucumber and the health risk. <i>Environmental Science and Pollution Research</i> 23(23):24298-24304.	933
	<i>Gossypium</i>	

1639289	Wu, Y., Yuan, S. L. (2012). Dibutyl phthalate pollution on cotton growth and physiological characteristics of cotton. <i>Advanced Materials Research</i> 518-523:5436-5441.	937
	<i>Holcus lanatus</i>	
1302103	Dueck, T. A., Dijk, Van, C. J., David, F., Scholz, N., Vanwalleghem, F. (2003). Chronic effects of vapour phase di-n-butyl phthalate (DBP) on six plant species. <i>Chemosphere</i> 53(8):911-920.	941
	<i>Leptochloa chinensis</i>	
5432995	Chuah, T. S., Oh, H. Y., Habsah, M., Norhafizah, M. Z., Ismail, B. S. (2014). Potential of crude extract and isolated compounds from golden beard grass (<i>Chrysopogon serrulatus</i>) for control of sprangletop (<i>Leptochloa chinensis</i>) in aerobic rice systems. <i>Crop and Pasture Science</i> 65(5):461-469.	945
5432995	Chuah, T. S., Oh, H. Y., Habsah, M., Norhafizah, M. Z., Ismail, B. S. (2014). Potential of crude extract and isolated compounds from golden beard grass (<i>Chrysopogon serrulatus</i>) for control of sprangletop (<i>Leptochloa chinensis</i>) in aerobic rice systems. <i>Crop and Pasture Science</i> 65(5):461-469.	947
	<i>Lolium perenne</i>	
2915866	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. <i>Frontiers of Environmental Science & Engineering</i> 9(2):259-268.	949
	<i>Medicago sativa</i>	
2915866	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. <i>Frontiers of Environmental Science & Engineering</i> 9(2):259-268.	955
	<i>Nicotiana tabacum</i>	
5627041	Deng, J., Zhang, Y., Hu, J., Jiao, J., Hu, F., Li, H., Zhang, S. (2017). Autotoxicity of phthalate esters in tobacco root exudates: Effects on seed germination and seedling growth. <i>Pedosphere</i> 27(6):1073-1082.	961
	<i>Nicotinana tobacum</i>	
792357	Jia, Z. H., Yi, J. H., Su, Y. R., Shen, H. (2011). Autotoxic substances in the root exudates from continuous tobacco cropping. <i>Allelopathy Journal</i> 27(1):87-96.	965
	<i>Oryza sativa</i>	
5551990	Isogai, Y., Komoda, Y., Okamoto, T. (1972). Biological activities of n-butyl phthalate and its analogous compounds on various bioassays of plant growth regulators. <i>Scientific papers of the College of General Education, University of Tokyo</i> 22(2):129-135.	973
5432995	Chuah, T. S., Oh, H. Y., Habsah, M., Norhafizah, M. Z., Ismail, B. S. (2014). Potential of crude extract and isolated compounds from golden beard grass (<i>Chrysopogon serrulatus</i>) for control of sprangletop (<i>Leptochloa chinensis</i>) in aerobic rice systems. <i>Crop and Pasture Science</i> 65(5):461-469.	983
	<i>Phaseolus vulgaris</i>	
1302103	Dueck, T. A., Dijk, Van, C. J., David, F., Scholz, N., Vanwalleghem, F. (2003). Chronic effects of vapour phase di-n-butyl phthalate (DBP) on six plant species. <i>Chemosphere</i> 53(8):911-920.	985
	<i>Picea abies</i>	

1302103	Dueck, T. A., Dijk, Van, C. J., David, F., Scholz, N., Vanwalleghe, F. (2003). Chronic effects of vapour phase di-n-butyl phthalate (DBP) on six plant species. <i>Chemosphere</i> 53(8):911-920.	987
	<i>Plantago major</i>	
1302103	Dueck, T. A., Dijk, Van, C. J., David, F., Scholz, N., Vanwalleghe, F. (2003). Chronic effects of vapour phase di-n-butyl phthalate (DBP) on six plant species. <i>Chemosphere</i> 53(8):911-920.	991
	<i>Raphanus sativus</i>	
2915866	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. <i>Frontiers of Environmental Science & Engineering</i> 9(2):259-268.	993
1333234	Virgin, H. I., A-M, Holst, Morner, J. (1981). Effect of di-n-butylphthalate on the carotenoid synthesis in green plants. <i>Physiologia Plantarum</i> 53(2):158-163.	999
	<i>Sinapis Alba</i>	
9430481	LäKke, H., Rasmussen, L. (1983). Phytotoxicological effects of Di-(2-ethyl hexyl)-phthalate and Di-n-butyl-phthalate on higher plants in laboratory and field experiments. <i>Environmental Pollution Series A: Ecological and Biological</i> 32(3):179-199.	1005
	<i>Sinapsis alba</i>	
680337	Løkke, H., Bro-Rasmussen, F. (1981). Studies of mobility of di-iso-butyl phthalate (DiBP), di-N-butyl phthalate (DBP), and di-(2-ethyl hexyl) phthalate (DEHP) by plant foliage treatment in a closed terrestrial simulation chamber. <i>Chemosphere</i> 10(11-12):1223-1236.	1007
	<i>Sorghum bicolor</i>	
5433174	Kannan, S. (1986). Effects of dibutyl phthalate and phthalic-acid on chlorosis recovery in iron-deficiency stressed sorghum cultivars. <i>Journal of Plant Nutrition</i> 9(12):1543-1551.	1011
	<i>Trifolium repens</i>	
1302103	Dueck, T. A., Dijk, Van, C. J., David, F., Scholz, N., Vanwalleghe, F. (2003). Chronic effects of vapour phase di-n-butyl phthalate (DBP) on six plant species. <i>Chemosphere</i> 53(8):911-920.	1019
	<i>Triticum aestivum</i>	
2915866	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. <i>Frontiers of Environmental Science & Engineering</i> 9(2):259-268.	1023
	<i>Triticum aestivum L.</i>	
5495646	Gao, M., Guo, Z., Dong, Y., Song, Z. (2019). Effects of di-n-butyl phthalate on photosynthetic performance and oxidative damage in different growth stages of wheat in cinnamon soils. <i>Environmental Pollution</i> 250:357-365.	1029
	<i>Triticum sp</i>	
3350318	Gao, M., Qi, Y., Song, W., Xu, H. (2016). Effects of di-n-butyl phthalate and di (2-ethylhexyl) phthalate on the growth, photosynthesis, and chlorophyll fluorescence of wheat seedlings. <i>Chemosphere</i> 151:76-83.	1033
	<i>Vigna radiata</i>	

2510954	Ma, T. T., Christie, P., Luo, Y. M., Teng, Y. (2014). Physiological and antioxidant responses of germinating mung bean seedlings to phthalate esters in soil. <i>Pedosphere</i> 24(1):107-115.	1037
	<i>Vigna sinensis</i>	
5495799	Wang, S. G., Lin, X. G., Yin, R., Hou, Y. L. (2003). Effects of di-n-butyl phthalate on mycorrhizal and non-mycorrhizal cowpea plants. <i>Biologia Plantarum</i> 47(4):637-639.	1043

Study Citation:	Ren, Z., Ren, B., Ren, B., Chen, B., Pan, H., Li, S., Xu, S., Tae-Soo, C., Wang, W. (2019). Is circadian rhythm a good indicator in the environmental assessment? The toxic effects of contaminants in trace level on the behavior responses of goldfish (<i>Carassius auratus</i>). <i>Ecological Indicators</i> 405:700-708.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Carassius auratus</i> ; Adult			
Health Outcome:	Behavioral			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5673506			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The test substance was identified by name and molecular formula only.
	Metric 2:	Test Substance Source	Low	The test substance was reported to be from Chinese Standard Sample Center, but it was not reported to be analytically verified.
	Metric 3:	Test Substance Purity	High	The test substance was technical grade with >95% purity.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of a concurrent solvent control in which 0.5% DMSO was used, which is equal to or greater than the concentrations of DMSO used in exposure groups.
	Metric 5:	Negative Control Response	High	The solvent control response was reported in Figures 1-6 and was adequate.
	Metric 6:	Randomized Allocation	Medium	Researchers reported one male and one female goldfish were randomly allocated to each test chamber.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	Little details were provided on the preparation of the test stock solution and test concentration. Analytical verification of exposure concentrations was not reported. Vessel material was not reported.
	Metric 8:	Consistency of Exposure Administration	High	All exposures were for six days in a flow through system with test chambers that were 15cm long and 10cm in diameter at 22 +/- 2 C.
	Metric 9:	Measurement of Test Substance Concentration	Low	It was not reported if the test substances were measured at any point in the study.
	Metric 10:	Exposure Duration and Frequency	High	The duration was reported to be six days. This was adequate to see a response.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	There was only one exposure level as the goal was not to have a dose response, but to compare the responses of different chemicals.
	Metric 12:	Testing at or Below Solubility Limit	High	The DBP concentration was below the water solubility limit, and a vehicle solvent was used with an appropriate solvent control response.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The adult goldfish were from the Institute of Environment and Ecology at Shandong Normal University in China. Goldfish are a relevant species for this test.
	Metric 14:	Acclimatization and Pretreatment Conditions	Medium	Pretreatment environmental conditions were the same as test conditions. Acclimation to the flow-through cylindrical chambers prior to treatment was not reported.
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Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Carassius auratus</i> ; Adult			
Health Outcome:	Behavioral			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5673506			
Domain		Metric	Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Medium	There were two goldfish per test chamber and four replicates per test chemical.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	The goldfish were kept at 22C with a 16L:8D photoperiod (4000 lux) in a test chamber containing two goldfish. The flow rate was maintained at 2L/h. The fish were fed <i>Limnodrilus hoffmeisteri</i> and flake fish food. pH and DO were not reported.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—behavioral changes due to DBP exposure.
	Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	The statistical methods were described in detail in the "Data analysis" section of the paper.
	Metric 22:	Reporting of Data	High	The data for the exposure and control responses were reported in Figures 1-6.
	Metric 23:	Explanation of Unexpected Outcomes	High	The study authors did not report any unexpected outcomes.
Additional Comments:	This evaluation was on the effect of DBP on <i>C. auratus</i> circadian rhythms. The behavioral outcome was selected as the outcome of interest.			

Overall Quality Determination**Medium**

Study Citation:	Pfuderer, P., Janzen, S., T, R.W., , J. R. (1975). The identification of phthalic acid esters in the tissues of cyprinodont fish and their activity as heart rate depressors. Environmental Research 9(3):215-223.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Carassius auratus</i> ; Juvenile			
Health Outcome:	Cardiovascular			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1333101			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only.
	Metric 2:	Test Substance Source	Low	The source was not reported.
	Metric 3:	Test Substance Purity	High	The chemical purity was reported as 99.94%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Uninformative	A concurrent negative control group was not included.
	Metric 5:	Negative Control Response	Low	The biological response of the negative control groups was not reported.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations. The cited reference for methods (Francis et al., 1975) was unavailable, therefore this metric score reflects the amount of details provided in the study being reviewed.
	Metric 8:	Consistency of Exposure Administration	Uninformative	A sonicated emulsion was used for exposure, presumably for all concentrations. Exposure administration cannot be expected to be consistent across test organisms/study groups using emulsions.
	Metric 9:	Measurement of Test Substance Concentration	Uninformative	Exposure concentrations were not measured and nominal values are highly uncertain due to the nature of the test substance.
	Metric 10:	Exposure Duration and Frequency	Uninformative	The duration of exposure and/or exposure frequency were not reported.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were justified for a dose response.
	Metric 12:	Testing at or Below Solubility Limit	Low	An emulsion was used, so there is high probability at least one concentration exceeds solubility.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test animals was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.
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Study Citation:	Pfuderer, P., Janzen, S., T, R.W., , J. R. (1975). The identification of phthalic acid esters in the tissues of cyprinodont fish and their activity as heart rate depressors. Environmental Research 9(3):215-223.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Carassius auratus</i> ; Juvenile			
Health Outcome:	Cardiovascular			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1333101			
Domain	Metric	Rating	Comments	
	Metric 15: Number of Organisms and Replicates per Group	Low	The number of test organisms and/or replicates was not reported. The figure on heart rate activity (Fig 6) showed only the response of an individual fish but mean heart rate was given in Table 1. The cited reference for methods (Francis et al., 1975) was unavailable, therefore this metric score reflects the amount of details provided in the study being reviewed.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the amount of details provided in the study being reviewed.	
	Metric 17: Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported. The cited reference for methods (Francis et al., 1975) was unavailable, therefore this metric score reflects the amount of details provided in the study being reviewed.	
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported. The cited reference for methods (Francis et al., 1975) was unavailable, therefore this metric score reflects the amount of details provided in the study being reviewed.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	Uninformative	Statistical analysis was not conducted.	
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment group (Table 1).	
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes. Heart rate data was provided with some measure of variability in Table 1.	
Additional Comments:	Overall, a poorly designed and poorly written study. A negative control group was not reported. Details of experimental system, test media preparation, exposure concentrations, duration of exposure were not provided. It was reported that a sonicated emulsion was used for exposure. Mean heart rate data was provided for DBP for the concentrations tested in Table 1. The cited reference for methods was not available at the time of review.			

Overall Quality Determination**Uninformative**

Study Citation:	Adams, W. J., Biddinger, G. R., Robillard, K. A., Gorsuch, J. W. (1995). A summary of the acute toxicity of 14 phthalate esters to representative aquatic organisms. Environmental Toxicology and Chemistry 14(9):1569-1574.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Cyprinodon variegatus</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1321996; Linked HERO ID(s): 1321996, 1316224			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Test substance nomenclature was reported without a CASRN.
	Metric 2:	Test Substance Source	Low	The test substance was reported as provided by the manufacturer from commercially available batches. The manufacture name and batch number were not provided. No analytical data was reported.
	Metric 3:	Test Substance Purity	High	
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	The negative control response was reported.
	Metric 5:	Negative Control Response	High	The control response was acceptable.
	Metric 6:	Randomized Allocation	Low	The allocation method was not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The experimental system was well described. However, headspace or measures to prevent volatilization were not reported.
	Metric 8:	Consistency of Exposure Administration	High	Exposure administration was consistent across groups.
	Metric 9:	Measurement of Test Substance Concentration	Medium	Sample extracts were analyzed by gas chromatography at the start and the end of the test. In static studies, final test concentrations frequently were 50% of the initial concentrations. Loss of the phthalate esters was thought to be principally due to adsorption to the test vessels.
	Metric 10:	Exposure Duration and Frequency	High	The duration and frequency of the exposure were appropriate for the test.
	Metric 11:	Number of Exposure Groups/Spacing of Exposure Levels	High	Exposure levels were appropriate. A range finding test was performed.
	Metric 12:	Testing at or Below Solubility Limit	High	The test was performed at or below the water solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	An appropriate acclimation period for the test was reported.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Each test was performed using duplicate test concentrations with 10 organisms per test vessel.
Domain 5: Outcome Assessment				
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Study Citation:	Adams, W. J., Biddinger, G. R., Robillard, K. A., Gorsuch, J. W. (1995). A summary of the acute toxicity of 14 phthalate esters to representative aquatic organisms. <i>Environmental Toxicology and Chemistry</i> 14(9):1569-1574.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Cyprinodon variegatus</i> ; Juvenile
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	1321996; Linked HERO ID(s): 1321996, 1316224

Domain	Metric	Rating	Comments
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions were appropriate for the test.
	Metric 17: Outcome Assessment Methodology	High	The intended outcomes were reported.
	Metric 18: Consistency of Outcome Assessment	High	Outcome assessment was consistent across groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	Environmental conditions were consistent across groups.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There were no reported differences between groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were performed and described.
	Metric 22: Reporting of Data	Medium	Only treatment endpoints were reported.
	Metric 23: Explanation of Unexpected Outcomes	High	No unexpected outcomes were reported.
Additional Comments: None			

Overall Quality Determination**High**

Study Citation:	Agus, H. H., Erkmen, B., Sümer, S., Sepici-Dinçel, A., Erkoç, F. (2015). Impact of DBP on histology and expression of HSP 70 in gill and liver tissue of <i>Cyprinus carpio</i> . Molecular Biology Reports 42(9):1409-1417.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Cyprinus carpio</i> ; Juvenile			
Health Outcome:	Hepatic/Liver			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3071043			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The DBP was identified by CASRN.
	Metric 2:	Test Substance Source	Low	The DBP was reported to be from Merck, but it was not reported to be analytically verified.
	Metric 3:	Test Substance Purity	High	The DBP was reported to be 100% pure.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using a solvent control.
	Metric 5:	Negative Control Response	High	The negative control response was reported in Table 1 and Figure 3 and was adequate for the outcomes of interest.
	Metric 6:	Randomized Allocation	Low	It was not reported how the fish were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	Few details were provided on the preparation of test substance. Vessel material was not reported.
	Metric 8:	Consistency of Exposure Administration	Low	All tests were conducted in 120L tanks, but little other information was provided on test consistency.
	Metric 9:	Measurement of Test Substance Concentration	Low	It was not reported if test concentrations were measured at any point.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration was reported to be 96h with samples taken at 4, 24, and 96h. This was adequate to observe a response.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	3 exposure groups (1, 5, and 10 mg/L) were tested in a range-finder to determine an appropriate sublethal dose. These were appropriate concentrations for the range-finder, as a sublethal dose was identified as 1 mg/L. This is marked as N/A, because the authors' intention for the study was to assess effects from a sublethal concentration over a time course.
	Metric 12:	Testing at or Below Solubility Limit	High	The exposure concentration was below the solubility limit, and acetone was used as a vehicle solvent.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The carp are an appropriate test species and were reported to be juveniles. They were not obtained from a reliable source, they were obtained from the State Hydraulic Works General Directorate, DSI, Yedikir Dam Lake, Amasya, Turkey, indicating they were wild-caught.

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Study Citation:	Agus, H. H., Erkmen, B., Sümer, S., Sepici-Dinçel, A., Erkoç, F. (2015). Impact of DBP on histology and expression of HSP 70 in gill and liver tissue of <i>Cyprinus carpio</i> . Molecular Biology Reports 42(9):1409-1417.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Cyprinus carpio</i> ; Juvenile			
Health Outcome:	Hepatic/Liver			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3071043			
Domain	Metric	Rating	Comments	
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The carp were acclimated for 15d prior to the start of the study.
	Metric 15:	Number of Organisms and Replicates per Group	Low	5 to 7 organisms were sampled per time point in the treatment group and 16 organisms were in the control group, but the number of replicates (fish tanks) was not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Organisms were kept in 120L tanks with a 12L:12D photoperiod in dechlorinated tap water. It was reported the fish were fed commercial fish food and were then starved prior to the study. The feeding regimen was not reported. More information on temperature is needed.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest–Histopathological effects on the liver.
	Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. At each sample period, fish tissue was fixed and later examined using light microscopy.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions. Study authors needed to provide more information on the environmental conditions of the fish. Water quality measurements are reported, but only one number for each parameter is given, making it unclear if these are representative of control or exposure group conditions, or an average of both, or whether conditions differed between control and exposure groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were described in the "Data Analysis" section of the paper and were appropriate.
	Metric 22:	Reporting of Data	High	Results were reported in Table 1 and Figures 3 and 4 for the control response and the exposure response. The data was appropriate for the outcomes of interest.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.
Additional Comments:	This portion of the study was on the histopathological effects of DBP on <i>C. carpio</i> livers. The liver/hepatic outcome was selected as the outcome of interest.			

Overall Quality Determination**Medium**

Study Citation:	Agus, H. H., Erkmen, B., Sümer, S., Sepici-Dinçel, A., Erkoç, F. (2015). Impact of DBP on histology and expression of HSP 70 in gill and liver tissue of <i>Cyprinus carpio</i> . <i>Molecular Biology Reports</i> 42(9):1409-1417.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Cyprinus carpio</i> ; Juvenile			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3071043			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The DBP was identified by CASRN.	
	Metric 2: Test Substance Source	Low	The DBP was reported to be from Merck, but it was not reported to be analytically verified.	
	Metric 3: Test Substance Purity	High	The DBP was reported to be 100% pure.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a solvent control.	
	Metric 5: Negative Control Response	High	The negative control response was reported in Figures 5 and 6 and was adequate for the outcomes of interest.	
	Metric 6: Randomized Allocation	Low	It was not reported how the fish were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	Few details were provided on the preparation of the test substance. Vessel material was not reported.	
	Metric 8: Consistency of Exposure Administration	Low	All tests were conducted in 120L tanks, but little other information was provided on test consistency.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if test concentrations were measured at any point.	
	Metric 10: Exposure Duration and Frequency	High	The exposure duration was reported to be 96h with samples taken at 4, 24, and 96h. This was adequate to observe a response.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	N/A	Three exposure groups (1, 5, and 10 mg/L) were tested in a range-finder to determine an appropriate sublethal dose. These were appropriate concentrations for the range-finder, as a sublethal dose was identified as 1 mg/L. This metric is marked as N/A, because the authors' intention for the study was to assess effects from a sublethal concentration over a time course.	
	Metric 12: Testing at or Below Solubility Limit	High	The exposure concentration was below the solubility limit, and acetone was used as a vehicle solvent.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Medium	The carp are an appropriate test species and were reported to be juveniles. They were not obtained from a reliable source, they were obtained from the State Hydraulic Works General Directorate, DSI, Yedikir Dam Lake, Amasya, Turkey, indicating they were wild-caught.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	The carp were acclimated for 15d prior to the start of the study.	
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Study Citation:	Agus, H. H., Erkmen, B., Sümer, S., Sepici-Dinçel, A., Erkoç, F. (2015). Impact of DBP on histology and expression of HSP 70 in gill and liver tissue of <i>Cyprinus carpio</i> . Molecular Biology Reports 42(9):1409-1417.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Cyprinus carpio</i> ; Juvenile			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3071043			
Domain	Metric	Rating	Comments	
	Metric 15: Number of Organisms and Replicates per Group	Low	Five to seven organisms were sampled per time point in the treatment group and 16 organisms were in the control group, but the number of replicates (fish tanks) was not reported.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Organisms were kept in 120L tanks with a 12L:12D photoperiod in dechlorinated tap water. It was reported the fish were fed commercial fish food and were then starved prior to the study. The feeding regimen was not reported. More information is needed on temperature as well.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—changes in HSP70 expression over time.	
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. Real time PCR was used to measure HSP70 in the gills and the liver, and Western Blot was used to measure HSP70 protein levels.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions. Study authors needed to provide more information on the environmental conditions of the fish. Water quality measurements are reported, but only one number for each parameter is given, making it unclear if these are representative of control or exposure group conditions, or an average of both, or whether conditions differed between control and exposure groups.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical methods were described in the "Data Analysis" section of the paper and were appropriate.	
	Metric 22: Reporting of Data	High	Results were reported in Figures 5 and 6 for the exposure response and the control response. Data presented was appropriate for the outcomes of interest.	
	Metric 23: Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in the figures.	
Additional Comments:	This portion of the study was on the effect of DBP on HSP70 expression and protein levels. The mechanistic biomarker outcome was selected as the outcome of interest.			
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Study Citation:	Agus, H. H., Erkmen, B., Sümer, S., Sepici-Dinçel, A., Erkoç, F. (2015). Impact of DBP on histology and expression of HSP 70 in gill and liver tissue of <i>Cyprinus carpio</i> . Molecular Biology Reports 42(9):1409-1417.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Cyprinus carpio</i> ; Juvenile
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	3071043

Domain	Metric	Rating	Comments
Overall Quality Determination		Medium	

Study Citation:	Agus, H. H., Erkmen, B., Sümer, S., Sepici-Dinçel, A., Erkoç, F. (2015). Impact of DBP on histology and expression of HSP 70 in gill and liver tissue of <i>Cyprinus carpio</i> . Molecular Biology Reports 42(9):1409-1417.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Cyprinus carpio</i> ; Juvenile			
Health Outcome:	Respiratory			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3071043			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The DBP was identified by CASRN.	
	Metric 2: Test Substance Source	Low	The DBP was reported to be from Merck, but it was not reported to be analytically verified.	
	Metric 3: Test Substance Purity	High	The DBP was reported to be 100% pure.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a solvent control.	
	Metric 5: Negative Control Response	High	The negative control response was reported in Table 1 and Figure 1 and was adequate for the outcomes of interest.	
	Metric 6: Randomized Allocation	Low	It was not reported how the fish were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	Few details were provided on the preparation of test substance. Vessel material was not reported.	
	Metric 8: Consistency of Exposure Administration	Low	All tests were conducted in 120L tanks, but little other information was provided on test consistency.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if test concentrations were measured at any point.	
	Metric 10: Exposure Duration and Frequency	High	The exposure duration was reported to be 96h with samples taken at 4, 24, and 96h. This was adequate to observe a response.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	N/A	3 exposure groups (1, 5, and 10 mg/L) were tested in a range-finder to determine an appropriate sublethal dose. These were appropriate concentrations for the range-finder, as a sublethal dose was identified as 1 mg/L. This is marked as N/A, because the authors' intention for the study was to assess effects from a sublethal concentration over a time course.	
	Metric 12: Testing at or Below Solubility Limit	High	The exposure concentration was below the solubility limit, and acetone was used as a vehicle solvent.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Medium	The carp are an appropriate test species and were reported to be juveniles. They were not obtained from a reliable source, they were obtained from the State Hydraulic Works General Directorate, DSI, Yedikir Dam Lake, Amasya, Turkey, indicating they were wild-caught.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	The carp were acclimated for 15d prior to the start of the study.	
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Study Citation:	Agus, H. H., Erkmen, B., Sümer, S., Sepici-Dinçel, A., Erkoç, F. (2015). Impact of DBP on histology and expression of HSP 70 in gill and liver tissue of <i>Cyprinus carpio</i> . Molecular Biology Reports 42(9):1409-1417.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Cyprinus carpio</i> ; Juvenile			
Health Outcome:	Respiratory			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3071043			
Domain	Metric	Rating	Comments	
	Metric 15: Number of Organisms and Replicates per Group	Low	5 to 7 organisms were sampled per time point in the treatment group and 16 organisms were in the control group, but the number of replicates (fish tanks) was not reported.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Organisms were kept in 120L tanks with a 12L:12D photoperiod in dechlorinated tap water. It was reported the fish were fed commercial fish food and were then starved prior to the study. The feeding regimen was not reported. More information is needed on temperature as well.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest–Histopathological effects on the gills.	
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. At each sample period, fish tissue was fixed and later examined using light microscopy.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions. Study authors needed to provide more information on the environmental conditions of the fish. Water quality measurements are reported, but only one number for each parameter is given, making it unclear if these are representative of control or exposure group conditions, or an average of both, or whether conditions differed between control and exposure groups.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical methods were described in the "Data Analysis" section of the paper and were appropriate.	
	Metric 22: Reporting of Data	High	Results were reported in Table 1 and Figures 1 and 2 for the control response and the exposure response. The data was appropriate for the outcomes of interest.	
	Metric 23: Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.	
Additional Comments:	This portion of the study was on the histopathological effects of DBP on <i>C. carpio</i> gills. The respiratory outcome was selected as the outcome of interest.			

Overall Quality Determination**Medium**

Study Citation:	Poopal, R. K., Ramesh, M., Maruthappan, V., Rajendran, R. B. (2017). Potential effects of low molecular weight phthalate esters (C16H22O4 and C12H14O4) on the freshwater fish <i>Cyprinus carpio</i> . <i>Toxicology Research</i> 6(4):505-520.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Cyprinus carpio</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3974208			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The DBP was identified by name only.	
	Metric 2: Test Substance Source	Low	The source of the DBP was reported to be from Loba Chemie Pvt. Ltd. in Mumbai, India. It was reported the DBP did not have any impurities. The test substance identity was NOT analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	It was reported the DBP was analytical grade and 99.99% pure.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	It was reported a toxicant free negative control group was used.	
	Metric 5: Negative Control Response	Low	The biological response of the negative control was not reported.	
	Metric 6: Randomized Allocation	Low	It was not reported how the fish were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	Little information on the preparation of the test concentrations was reported. The system was reported to be a static system. Experiments were conducted in glass tanks.	
	Metric 8: Consistency of Exposure Administration	High	All exposures were for 96h in 20L glass tanks with 10 fish per tank and three replicates per concentration. Fish were monitored daily for mortalities.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if the test concentrations were measured at any point in the study.	
	Metric 10: Exposure Duration and Frequency	High	The exposure duration was 96h, which is typical of a fish acute toxicity test.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	There were seven exposure levels, and the spacing was adequate to obtain LC50 values.	
	Metric 12: Testing at or Below Solubility Limit	Uninformative	Five out of seven concentrations of the DBP tested were above the water solubility limit. Study authors did not report if the negative control was a solvent control. According to the scope document, the water solubility of DBP is 11.2 mg/L at 25°C. DBP concentrations tested were 5, 10, 15, 20, 25, 35 and 40 mg L−1.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The fish were reported to be from the Tamil Nadu Fisheries Development Corporation in Aliyar, Tamil Nadu, India. Fingerlings were reported to be used in the study.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	The fish were acclimated to lab conditions for 30 days prior to testing.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were ten fish per test chamber with three replicates per concentration.	

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Study Citation:	Poopal, R. K., Ramesh, M., Maruthappan, V., Rajendran, R. B. (2017). Potential effects of low molecular weight phthalate esters (C16H22O4 and C12H14O4) on the freshwater fish <i>Cyprinus carpio</i> . Toxicology Research 6(4):505-520.		
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Cyprinus carpio</i> ; Juvenile		
Health Outcome:	Mortality		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	3974208		
Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
Metric 16:	Adequacy of Test Conditions	Medium	The fish were kept in 20L tanks during the study. Prior to the study they were fed rice bran and ground nut oil cake dough ad libitum. It was reported the they were kept at natural temperatures and photoperiod, but it was not specified what these were.
Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest–LC50 values.
Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. The fish were observed every 24h for mortality, and dead fish were removed immediately.
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	High	LC50 values were determined by probit analysis.
Metric 22:	Reporting of Data	Low	Only LC50 values were reported in the text. Control and exposure responses were not reported.
Metric 23:	Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability. The 95% confidence intervals were not reported.
Additional Comments:	This portion of the evaluation is on the 96h acute toxicity of DBP. Mortality was selected as the outcome of interest. The study received an unacceptable rating due to the fact that five out of seven concentrations tested were above the DBP water solubility limit, and it was not reported if the negative control contained a vehicle solvent.		

Overall Quality Determination**Uninformative**

Study Citation:	Zhao, X., Gao, Y., Qi, M. (2014). Toxicity of phthalate esters exposure to carp (<i>Cyprinus carpio</i>) and antioxidant response by biomarker. <i>Ecotoxicology</i> 23(4):626-632.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Cyprinus carpio</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2510817			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The chemical was identified by name.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical grade was reported as analytical.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent solvent control group.	
	Metric 5: Negative Control Response	Low	The biological response of the negative control groups was not reported.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.	
	Metric 8: Consistency of Exposure Administration	Low	Few details of exposure administration were reported.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured or measurements were not reported.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Low	Spacing was not reported.	
	Metric 12: Testing at or Below Solubility Limit	Low	The reported LC50 from this study was 16.3 mg/L for DBP. The solubility listed in the Final Scope for this compound is 2.69 mg/L.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Medium	There are minor reservations about the choice of source of test organisms.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.	
	Metric 15: Number of Organisms and Replicates per Group	Low	The number of test organisms and replicates were lower than the typical number.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Reporting of environmental conditions was not sufficiently reported to evaluate if adequate.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	

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Study Citation:	Zhao, X., Gao, Y., Qi, M. (2014). Toxicity of phthalate esters exposure to carp (<i>Cyprinus carpio</i>) and antioxidant response by biomarker. <i>Ecotoxicology</i> 23(4):626-632.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Cyprinus carpio</i> ; Adult
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	2510817

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	Medium	Few details regarding the execution of the study protocol for outcome assessment were reported.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	High	There were no differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22: Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: The reported LC50 from this study was 16.3 mg/L for DBP. The solubility listed in the Final Scope for this compound is 2.69 mg/L.

Overall Quality Determination

Medium

Study Citation:	Zhao, X., Gao, Y., Qi, M. (2014). Toxicity of phthalate esters exposure to carp (<i>Cyprinus carpio</i>) and antioxidant response by biomarker. <i>Ecotoxicology</i> 23(4):626-632.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Cyprinus carpio</i> ; Adult			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2510817			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The chemical was identified by name.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical grade was reported as analytical.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent solvent control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control groups was reported and adequate.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.	
	Metric 8: Consistency of Exposure Administration	Low	Few details of exposure administration were reported.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured or measurements were not reported.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for a dose response.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Medium	The number of exposure groups and spacing of exposure levels were adequate for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	Low	Two of the three treatment concentrations (3.26 and 8.15 mg/L) are above the solubility listed in the final scope for DBP at 2.6 mg/L.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Medium	There are minor reservations about the choice of source of test organisms.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.	
	Metric 15: Number of Organisms and Replicates per Group	Low	The number of test organisms and replicates were lower than the typical number.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Reporting of environmental conditions was not sufficiently reported to evaluate if adequate.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	

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Study Citation:	Zhao, X., Gao, Y., Qi, M. (2014). Toxicity of phthalate esters exposure to carp (<i>Cyprinus carpio</i>) and antioxidant response by biomarker. <i>Ecotoxicology</i> 23(4):626-632.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Cyprinus carpio</i> ; Adult
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	2510817

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	Medium	Few details regarding the execution of the study protocol for outcome assessment were reported.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	High	There were no differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: None

Overall Quality Determination**Medium**

Study Citation:	Poopal, R. K., Ramesh, M., Maruthappan, V., Rajendran, R. B. (2017). Potential effects of low molecular weight phthalate esters (C16H22O4 and C12H14O4) on the freshwater fish <i>Cyprinus carpio</i> . <i>Toxicology Research</i> 6(4):505-520.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Cyprinus carpio</i> ; Juvenile			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Oxidative stress (including redox biology)-Immune/Hematological-Nutritional and Metabolic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3974208			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The DBP was identified by name only.	
	Metric 2: Test Substance Source	Low	The source of the DBP was reported to be from Loba Chemie Pvt. Ltd. in Mumbai, India. It was reported the DBP did not have any impurities. The test substance identity was NOT analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	It was reported the DBP was analytical grade and 99.99% pure.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	It was reported a toxicant free negative control group was used as well as a control containing acetone reported as the positive control.	
	Metric 5: Negative Control Response	Low	The biological responses of the negative control and the solvent control were reported in Figures 1-12 and in Table 1 and were adequate for the outcomes of interest.	
	Metric 6: Randomized Allocation	Low	It was not reported how the fish were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	Little information on the preparation of the test concentrations was reported. The system was reported to be a static renewal treatment with daily water renewals to remove waste and renew the toxicant.	
	Metric 8: Consistency of Exposure Administration	High	All exposures were for 35 days in 100L glass aquaria with 50 fish each. There were daily renewals and samples taken at days 7, 14, 21, 28, and 35.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if the test concentrations were measured at any point in the study.	
	Metric 10: Exposure Duration and Frequency	High	The exposure duration was 35 days. This was adequate for a response to be observed.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Low	There were only two exposure levels, which is lower than is typical, but adequate for the outcomes of interest.	
	Metric 12: Testing at or Below Solubility Limit	Medium	The study authors reported the use of a positive control (acetone) and a negative control (no acetone or toxicant) in the results section but the test solution preparation details were not mentioned. The solvent concentration was not reported.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The fish were reported to be from the Tamil Nadu Fisheries Development Corporation in Aliyar, Tamil Nadu, India. Fingerlings were reported to be used in the study.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	The fish were acclimated to lab conditions for 30 days prior to testing.	

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Study Citation:	Poopal, R. K., Ramesh, M., Maruthappan, V., Rajendran, R. B. (2017). Potential effects of low molecular weight phthalate esters (C16H22O4 and C12H14O4) on the freshwater fish <i>Cyprinus carpio</i> . Toxicology Research 6(4):505-520.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Cyprinus carpio</i> ; Juvenile			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Oxidative stress (including redox biology)-Immune/Hematological-Nutritional and Metabolic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3974208			
Domain	Metric	Rating	Comments	
	Metric 15: Number of Organisms and Replicates per Group	Low	There were 50 fish per test chamber. Study authors reported a "suitable" number of replicates were used, but did not specify what these numbers were.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Medium	The fish were kept in 100L tanks during the study. Prior to the study they were fed rice bran and ground nut oil cake dough ad libitum. It was reported that they were kept at natural temperatures and photoperiod, but it was not specified what these were.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—a number of mechanistic outcomes.	
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	The study author described the statistical methods in the "statistical analysis" section.	
	Metric 22: Reporting of Data	High	Data for the control and exposure responses were reported in Table 2 and in Figures 1-12 and were adequate for the outcomes of interest.	
	Metric 23: Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability.	
Additional Comments:	This portion of the evaluation is on the effect of DBP on several mechanistic outcomes.			

Overall Quality Determination**Medium**

Study Citation:	Chen, X., Xu, S., Tan, T., Lee, S. T., Cheng, S. H., Lee, F., F.W., Xu, L., S.J., Ho, K. C. (2014). Toxicity and estrogenic endocrine disrupting activity of phthalates and their mixtures. International Journal of Environmental Research and Public Health 11(3):3156-3168.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; AB Strain; Embryo			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2298079			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The CAS numbers and structures for BBP, DBP, DEHP, DIDP, and DINP are reported.	
	Metric 2: Test Substance Source	High	Sources were listed.	
	Metric 3: Test Substance Purity	Low	Purity/grade were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Both a blank control and solvent control were used in the acute bioassays.	
	Metric 5: Negative Control Response	Low	Survival of the controls were not reported.	
	Metric 6: Randomized Allocation	Low	Random allocation was not reported.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	Dilution of the test substance into medium was not well described (unclear if embryo rearing medium was utilized) and the test substance was not renewed over 72 hr.	
	Metric 8: Consistency of Exposure Administration	Medium	Exposures appear to have been administered consistently.	
	Metric 9: Measurement of Test Substance Concentration	Low	Concentrations are reported as nominal.	
	Metric 10: Exposure Duration and Frequency	Medium	Acute exposures were 72-hr for embryos, which is slightly shorter than the standard 96-hour acute bioassay.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	Authors reported conducting exposures at 9 concentrations: 500, 100, 50, 10, 1.5, 0.6, 0.3, 0.06, 0.01 mg/L.	
	Metric 12: Testing at or Below Solubility Limit	Low	Several concentrations were above estimated solubility limits, including 3-4 treatment concentrations (BBP, DBP) or 7 concentrations (DEHP, DIDP, and DINP). Methanol was utilized to improve solubility.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	Source and strain were reported and husbandry methods were adequately described.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	Acclimation was not reported.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	Each concentration was represented by 20 embryos (one embryo per well).	
Domain 5: Outcome Assessment				
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Study Citation:	Chen, X., Xu, S., Tan, T., Lee, S. T., Cheng, S. H., Lee, F., F.W., Xu, L., S.J., Ho, K. C. (2014). Toxicity and estrogenic endocrine disrupting activity of phthalates and their mixtures. International Journal of Environmental Research and Public Health 11(3):3156-3168.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; AB Strain; Embryo
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	2298079

Domain	Metric	Rating	Comments
	Metric 16: Adequacy of Test Conditions	Low	The 24 well plates were described but there lacked details on water conditions (or use of embryo rearing medium) and temperature.
	Metric 17: Outcome Assessment Methodology	Low	It was not reported how mortality was determined in embryos.
	Metric 18: Consistency of Outcome Assessment	High	The outcome assessment appeared to be consistently conducted across treatment and control groups at 72 hr post treatment.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20: Outcomes Unrelated to Exposure	Medium	Nothing was reported to indicate that animal health or attrition interfered with the bioassay.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	There were no details on the LC50 estimation methods.
	Metric 22: Reporting of Data	Low	The authors report LC50 values for BBP and DBP (and LC50 not attained for DEHP, DINP, and DIDP). Mortality was not reported for each treatment group or for the controls.
	Metric 23: Explanation of Unexpected Outcomes	Low	No measures of variability were reported.
Additional Comments: None			

Overall Quality Determination**Medium**

Study Citation:	Pu, S. Y., Hamid, N., Ren, Y. W., Pei, D. S. (2020). Effects of phthalate acid esters on zebrafish larvae: Development and skeletal morphogenesis. Chemosphere 246:125808.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; Embryo			
Health Outcome:	Cardiovascular			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5932877			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	Chemicals were identified by CASRN.	
	Metric 2: Test Substance Source	Low	Chemicals were purchased from Sigma-Aldrich Beijing, but it was not reported if they were analytically verified.	
	Metric 3: Test Substance Purity	Low	Purity was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Controls with 0.1% DMSO were used.	
	Metric 5: Negative Control Response	Low	Mortality in controls was approximately 10%.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The test system was not described in any detail, and no further citation was given for preparation of test substances. Methods mention six-well plates with 5 mL solution per well, and six exposure concentrations per chemical. It can be inferred that the exposure was conducted in these well plates, but no details are given.	
	Metric 8: Consistency of Exposure Administration	Low	Details were not reported. It is not stated whether test substances were renewed during the study period or when dosing was performed.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured, and it is not stated whether test solutions were renewed over the course of the exposure. Based on the hydrolysis, photodegradation, and other degradation pathways of the phthalates, this is likely to have a substantial impact on results.	
	Metric 10: Exposure Duration and Frequency	Medium	The exposure duration was 7 days; frequency of dosing was not reported, but it can be inferred that no renewal of test substance took place.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	Six doses at a wide range of concentrations were chosen for each phthalate.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposures were below the solubility limit, and a 0.1% DMSO carrier was used.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of test animals was not reported.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	Acclimatization was not mentioned; embryos were transferred directly to plates at the beginning of the experiment.	
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Study Citation:	Pu, S. Y., Hamid, N., Ren, Y. W., Pei, D. S. (2020). Effects of phthalate acid esters on zebrafish larvae: Development and skeletal morphogenesis. Chemosphere 246:125808.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; Embryo
Health Outcome:	Cardiovascular
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	5932877

Domain	Metric	Rating	Comments
	Metric 15: Number of Organisms and Replicates per Group	Medium	Five embryos per concentration were tested in triplicate, for a total of fifteen per concentration. There were 30 fertilized eggs per six-well plate with 5mL of test solution.
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	Low	Reporting of housing and experimental conditions was not reported beyond the mention of six-well plates. Photoperiod, temperature, feeding, and other water quality characteristics were not mentioned.
	Metric 17: Outcome Assessment Methodology	High	Heart rate was assessed using a recording from a Nikon stereomicroscope.
	Metric 18: Consistency of Outcome Assessment	High	There was no evidence of outcomes assessed differently across exposure groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20: Outcomes Unrelated to Exposure	Medium	Outcomes unrelated to exposure were not reported.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical analysis was performed via T-test in SPSS, with normality confirmed by Kolmogorov-Smirnov.
	Metric 22: Reporting of Data	High	Outcomes were reported for each exposure group.
	Metric 23: Explanation of Unexpected Outcomes	High	Unexplained outcomes were not reported.
Additional Comments:	This evaluation is for the effect of DBP on cardiovascular outcomes such as heart rate and pericardial edema.		

Overall Quality Determination**Low**

Study Citation:	Pu, S. Y., Hamid, N., Ren, Y. W., Pei, D. S. (2020). Effects of phthalate acid esters on zebrafish larvae: Development and skeletal morphogenesis. Chemosphere 246:125808.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; Embryo			
Health Outcome:	Mechanistic-Cell signaling/function			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5932877			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemicals were identified by CASRN.
	Metric 2:	Test Substance Source	Low	Chemicals were purchased from Sigma-Aldrich Beijing, but it was not reported if they were analytically verified.
	Metric 3:	Test Substance Purity	Low	Purity was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Controls with 0.1% DMSO were used.
	Metric 5:	Negative Control Response	Low	Controls mortality was approximately 10%.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The test system was not described in any detail, and no further citation was given for preparation of test substances. Methods mention six-well plates with 5 mL solution per well, and six exposure concentrations per chemical. It can be inferred that exposure was conducted in these well plates, but no details are given.
	Metric 8:	Consistency of Exposure Administration	Low	Details were not reported. It is not stated whether test substances were renewed during the study period or when dosing was performed.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured, and it is not stated whether test solutions were renewed over the course of the exposure. Based on the hydrolysis, photodegradation, and other degradation pathways of the phthalates, this is likely to have a substantial impact on results.
	Metric 10:	Exposure Duration and Frequency	Medium	Exposure duration was 7 days; frequency of dosing was not reported but it can be inferred that no renewal of test substance took place.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	Only one concentration was used for the gene expression study.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposures were below the solubility limit and a 0.1% DMSO carrier was used.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of test animals was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	Acclimatization was not mentioned; embryos were transferred directly to plates at the beginning of the experiment.
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Study Citation:	Pu, S. Y., Hamid, N., Ren, Y. W., Pei, D. S. (2020). Effects of phthalate acid esters on zebrafish larvae: Development and skeletal morphogenesis. Chemosphere 246:125808.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; Embryo			
Health Outcome:	Mechanistic-Cell signaling/function			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5932877			
Domain		Metric	Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Low	Only one test concentration was used for the gene expression test, meaning that 5 embryos were used.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reporting of housing and experimental conditions was not reported beyond the mention of six-well plates. Photoperiod, temperature, feeding, and other water quality characteristics were not mentioned.
	Metric 17:	Outcome Assessment Methodology	High	Gene expression was quantified using qRT-PCR.
	Metric 18:	Consistency of Outcome Assessment	High	There was no evidence of outcomes assessed differently across exposure groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	Outcomes unrelated to exposure were not reported.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Uninformative	Statistical analysis was not performed and no data allowing analysis was provided for the gene expression tests.
	Metric 22:	Reporting of Data	High	Outcomes were reported for each exposure group.
	Metric 23:	Explanation of Unexpected Outcomes	Low	Measures of variability were not reported for this outcome (mean only).
Additional Comments:	This evaluation was for the gene expression assessment performed on zebrafish embryos after exposure to DBP. This portion of the evaluation received an unacceptable rating due to lack of statistical analysis.			
Overall Quality Determination		Uninformative		

Study Citation:	Pu, S. Y., Hamid, N., Ren, Y. W., Pei, D. S. (2020). Effects of phthalate acid esters on zebrafish larvae: Development and skeletal morphogenesis. Chemosphere 246:125808.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; Embryo
Health Outcome:	Behavioral
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	5932877

Domain	Metric	Rating	Comments
Domain 1: Test Substance			
	Metric 1: Test Substance Identity	High	Chemicals were identified by CASRN.
	Metric 2: Test Substance Source	Low	Chemicals were purchased from Sigma-Aldrich Beijing, but it was not reported if they were analytically verified.
	Metric 3: Test Substance Purity	Low	Purity was not reported.
Domain 2: Test Design			
	Metric 4: Negative Controls	High	Controls with 0.1% DMSO were used.
	Metric 5: Negative Control Response	Low	Mortality in controls was approximately 10%.
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization			
	Metric 7: Experimental System/Test Media Preparation	Low	The test system was not described in any detail, and no further citation was given for preparation of test substances. Methods mention six-well plates with 5 mL solution per well, and six exposure concentrations per chemical. It can be inferred that the exposure was conducted in these well plates, but no details are given.
	Metric 8: Consistency of Exposure Administration	Low	Details were not reported. It is not stated whether test substances were renewed during the study period or when dosing was performed.
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured, and it is not stated whether test solutions were renewed over the course of the exposure. Based on the hydrolysis, photodegradation, and other degradation pathways of the phthalates, this is likely to have a substantial impact on results.
	Metric 10: Exposure Duration and Frequency	Medium	The exposure duration was 7 days; frequency of dosing was not reported, but it can be inferred that no renewal of test substance took place.
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	Six doses at a wide range of concentrations were chosen for each phthalate.
	Metric 12: Testing at or Below Solubility Limit	High	Exposures were below the solubility limit, and a 0.1% DMSO carrier was used.
Domain 4: Test Organism			
	Metric 13: Test Organism Characteristics	Low	The source of test animals was not reported.
	Metric 14: Acclimatization and Pretreatment Conditions	Low	Acclimatization was not mentioned; embryos were transferred directly to plates at the beginning of the experiment.
	Metric 15: Number of Organisms and Replicates per Group	Medium	Five embryos per concentration were tested in triplicate, for a total of fifteen per concentration. There were 30 fertilized eggs per six-well plate with 5mL of test solution.
Domain 5: Outcome Assessment			

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Study Citation:	Pu, S. Y., Hamid, N., Ren, Y. W., Pei, D. S. (2020). Effects of phthalate acid esters on zebrafish larvae: Development and skeletal morphogenesis. Chemosphere 246:125808.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; Embryo			
Health Outcome:	Behavioral			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5932877			
Domain	Metric	Rating	Comments	
	Metric 16:	Adequacy of Test Conditions	Low	Reporting of housing and experimental conditions was not reported beyond the mention of six-well plates. Photoperiod, temperature, feeding, and other water quality characteristics were not mentioned.
	Metric 17:	Outcome Assessment Methodology	High	Movement rate (behavior) was assessed using a recording from a Nikon stereomicroscope.
	Metric 18:	Consistency of Outcome Assessment	High	There was no evidence of outcomes assessed differently across exposure groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	Outcomes unrelated to exposure were not reported.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical analysis was performed via T-test in SPSS, with normality confirmed by Kolmogorov-Smirnov.
	Metric 22:	Reporting of Data	High	Outcomes were reported for each exposure group.
	Metric 23:	Explanation of Unexpected Outcomes	High	Unexplained outcomes were not reported.
Additional Comments:	This evaluation is for behavior outcomes in embryos after exposure to DBP. These were measured in terms of movement after the exposure. These assessments were conducted at 24hpf.			

Overall Quality Determination**Low**

Study Citation:	Pu, S. Y., Hamid, N., Ren, Y. W., Pei, D. S. (2020). Effects of phthalate acid esters on zebrafish larvae: Development and skeletal morphogenesis. Chemosphere 246:125808.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; Embryo
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	5932877

Domain	Metric	Rating	Comments
Domain 1: Test Substance			
	Metric 1: Test Substance Identity	High	Chemicals were identified by CASRN.
	Metric 2: Test Substance Source	Low	Chemicals were purchased from Sigma-Aldrich Beijing, but it was not reported if they were analytically verified.
	Metric 3: Test Substance Purity	Low	Purity was not reported.
Domain 2: Test Design			
	Metric 4: Negative Controls	High	Controls with 0.1% DMSO were used.
	Metric 5: Negative Control Response	Low	Mortality in controls was approximately 10%.
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization			
	Metric 7: Experimental System/Test Media Preparation	Low	The test system was not described in any detail, and no further citation was given for preparation of test substances. Methods mention six-well plates with 5 mL solution per well, and six exposure concentrations per chemical. It can be inferred that the exposure was conducted in these well plates, but no details are given.
	Metric 8: Consistency of Exposure Administration	Low	Details were not reported. It is not stated whether test substances were renewed during the study period or when dosing was performed.
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured, and it is not stated whether test solutions were renewed over the course of the exposure. Based on the hydrolysis, photodegradation, and other degradation pathways of the phthalates, this is likely to have a substantial impact on results.
	Metric 10: Exposure Duration and Frequency	Medium	The exposure duration was 7 days; frequency of dosing was not reported, but it can be inferred that no renewal of test substance took place.
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	Six doses at a wide range of concentrations were chosen for each phthalate.
	Metric 12: Testing at or Below Solubility Limit	High	Exposures were below the solubility limit, and a 0.1% DMSO carrier was used.
Domain 4: Test Organism			
	Metric 13: Test Organism Characteristics	Low	The source of test animals was not reported.
	Metric 14: Acclimatization and Pretreatment Conditions	Low	Acclimatization was not mentioned; embryos were transferred directly to plates at the beginning of the experiment.
	Metric 15: Number of Organisms and Replicates per Group	Medium	Five embryos per concentration were tested in triplicate, for a total of fifteen per concentration. There were 30 fertilized eggs per six-well plate with 5mL of test solution.
Domain 5: Outcome Assessment			

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Study Citation:	Pu, S. Y., Hamid, N., Ren, Y. W., Pei, D. S. (2020). Effects of phthalate acid esters on zebrafish larvae: Development and skeletal morphogenesis. Chemosphere 246:125808.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; Embryo			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5932877			
Domain	Metric	Rating	Comments	
	Metric 16: Adequacy of Test Conditions	Low	Reporting of housing and experimental conditions was not reported beyond the mention of six-well plates. Photoperiod, temperature, feeding, and other water quality characteristics were not mentioned.	
	Metric 17: Outcome Assessment Methodology	High	Mortality was assessed via microscopy.	
	Metric 18: Consistency of Outcome Assessment	High	There was no evidence of outcomes assessed differently across exposure groups.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	Outcomes unrelated to exposure were not reported.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical analysis was performed via T-test in SPSS, with normality confirmed by Kolmogorov-Smirnov.	
	Metric 22: Reporting of Data	High	Outcomes were reported for each exposure group.	
	Metric 23: Explanation of Unexpected Outcomes	Low	Measures of variability were not reported for this outcome (mean only).	
Additional Comments:	This evaluation is for the effect of DBP on embryo mortality after seven days of exposure.			

Overall Quality Determination**Low**

Study Citation:	Pu, S. Y., Hamid, N., Ren, Y. W., Pei, D. S. (2020). Effects of phthalate acid esters on zebrafish larvae: Development and skeletal morphogenesis. Chemosphere 246:125808.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; Embryo			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5932877			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemicals were identified by CASRN.
	Metric 2:	Test Substance Source	Low	Chemicals were purchased from Sigma-Aldrich Beijing, but it was not reported if they were analytically verified.
	Metric 3:	Test Substance Purity	Low	Purity was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Controls with 0.1% DMSO were used.
	Metric 5:	Negative Control Response	Low	Mortality in controls was approximately 10%.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The test system was not described in any detail, and no further citation was given for preparation of test substances. Methods mention six-well plates with 5 mL solution per well, and six exposure concentrations per chemical. It can be inferred that the exposure was conducted in these well plates, but no details are given.
	Metric 8:	Consistency of Exposure Administration	Low	
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured, and it is not stated whether test solutions were renewed over the course of the exposure. Based on the hydrolysis, photodegradation, and other degradation pathways of the phthalates, this is likely to have a substantial impact on results.
	Metric 10:	Exposure Duration and Frequency	Medium	The exposure duration was 7 days; frequency of dosing was not reported, but it can be inferred that no renewal of test substance took place.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Six doses at a wide range of concentrations were chosen for each phthalate.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposures were below the solubility limit, and a 0.1% DMSO carrier was used.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of test animals was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	Acclimatization was not mentioned; embryos were transferred directly to plates at the beginning of the experiment.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Five embryos per concentration were tested in triplicate, for a total of fifteen per concentration. There were 30 fertilized eggs per six-well plate with 5mL of test solution.
Domain 5: Outcome Assessment				

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Study Citation:	Pu, S. Y., Hamid, N., Ren, Y. W., Pei, D. S. (2020). Effects of phthalate acid esters on zebrafish larvae: Development and skeletal morphogenesis. Chemosphere 246:125808.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; Embryo			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5932877			
Domain		Metric	Rating	Comments
	Metric 16:	Adequacy of Test Conditions	Low	Reporting of housing and experimental conditions was not reported beyond the mention of six-well plates. Photoperiod, temperature, feeding, and other water quality characteristics were not mentioned.
	Metric 17:	Outcome Assessment Methodology	Uninformative	
	Metric 18:	Consistency of Outcome Assessment	High	
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical analysis was performed via T-test in SPSS, with normality confirmed by Kolmogorov-Smirnov.
	Metric 22:	Reporting of Data	High	
	Metric 23:	Explanation of Unexpected Outcomes	Low	Measures of variability were not reported for this outcome (mean only).
Additional Comments:	This evaluation is for the effect of DBP on embryo development after seven days of exposure. This portion of the study received an unacceptable rating because the outcome assessment was unclear (See Metric 17 for detailed description).			

Overall Quality Determination**Uninformative**

Study Citation:	Thomas, D. G., Shankaran, H., Truong, L., Tanguay, R. L., Waters, K. M. (2019). Time-dependent behavioral data from zebrafish reveals novel signatures of chemical toxicity using point of departure analysis. Computational Toxicology 9:50-60.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; Embryo			
Health Outcome:	Behavioral			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	8635978			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The test substance was identified by name only.	
	Metric 2: Test Substance Source	Low	The source of the test substance was not reported, nor was it reported to be analytically verified.	
	Metric 3: Test Substance Purity	Low	The purity was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of and appropriate concurrent negative control.	
	Metric 5: Negative Control Response	Low	The negative control response was not reported.	
	Metric 6: Randomized Allocation	Low	It was not reported how the embryos were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The experimental system was not reported in great detail, nor was the preparation of the test solutions.	
	Metric 8: Consistency of Exposure Administration	Low	Little details of the exposure administration were reported.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if the test concentrations were measured at any point.	
	Metric 10: Exposure Duration and Frequency	High	The study duration was reported to be up 120hpf. Samples were also taken at 24hpf. This was adequate to observe the outcomes of interest.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	It was reported there were five test concentrations that increased by a factor of 10. There were two replicates per test concentration.	
	Metric 12: Testing at or Below Solubility Limit	High	Test concentrations were below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Medium	It was reported that zebrafish were raised at Oregon State University Sinnhuber Aquatic Laboratory in Corvallis, OR. It is unclear if the zebrafish were obtained from another lab and raised solely for this study, or if they were cultured at the facility long term.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if zebrafish were acclimated at any point.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 32 total embryos exposed to each test concentration split into two replicates.	
Domain 5: Outcome Assessment				
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Study Citation:	Thomas, D. G., Shankaran, H., Truong, L., Tanguay, R. L., Waters, K. M. (2019). Time-dependent behavioral data from zebrafish reveals novel signatures of chemical toxicity using point of departure analysis. Computational Toxicology 9:50-60.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; Embryo			
Health Outcome:	Behavioral			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	8635978			
Domain	Metric	Rating	Comments	
	Metric 16:	Adequacy of Test Conditions	Low	Little details were reported regarding organism housing or environmental conditions.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest- changes in behavior in terms of spontaneous movement and change in movement at light-to-dark transition time point.
	Metric 18:	Consistency of Outcome Assessment	High	Embryos were evaluated for changes in behavior were assessed using larval photomotor response assay using Viewpoint Zebrafish.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	Details regarding environmental conditions before and during the study were limited.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical analysis is reported in Section 3 "Theory and calculation."
	Metric 22:	Reporting of Data	Uninformative	This study is a reanalysis or replication (it is unclear from the text whether the experiment was repeated or whether the same data was reanalyzed) of HEROID 8591199; but the results for the same chemicals vary by up to 5 orders of magnitude for the same experimental results between the two studies. Further, the BMD10 values in Table S1 cannot follow from the LEL values reported in Table S5; endpoints with LELs at the lowest concentration tested are shown with "No effect" BMD10s. These inconsistencies render this pair of studies unacceptable for use.
	Metric 23:	Explanation of Unexpected Outcomes	Low	Variability was not reported.
Additional Comments:	This evaluation is for the effect of DBP on zebrafish embryo behavior. Data is reported in Table S1.This study is a reanalysis or replication (it is unclear from the text whether the experiment was repeated or whether the same data was reanalyzed) of HEROID 8591199; but the results for the same chemicals vary by up to 5 orders of magnitude for the same experimental results between the two studies. Further, the BMD10 values in Table S1 cannot follow from the LEL values reported in Table S5; endpoints with LELs at the lowest concentration tested are shown with "No effect" BMD10s. These inconsistencies render this pair of studies unacceptable for use.			
Overall Quality Determination		Uninformative		

Study Citation:	Thomas, D. G., Shankaran, H., Truong, L., Tanguay, R. L., Waters, K. M. (2019). Time-dependent behavioral data from zebrafish reveals novel signatures of chemical toxicity using point of departure analysis. Computational Toxicology 9:50-60.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; Embryo			
Health Outcome:	Immobilization			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	8635978			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The test substance was identified by name only.	
	Metric 2: Test Substance Source	Low	The source of the test substance was not reported, nor was it reported to be analytically verified.	
	Metric 3: Test Substance Purity	Low	The purity was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of and appropriate concurrent negative control.	
	Metric 5: Negative Control Response	Low	The negative control response was not reported.	
	Metric 6: Randomized Allocation	Low	It was not reported how the embryos were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The experimental system was not reported in great detail, nor was the preparation of the test solutions.	
	Metric 8: Consistency of Exposure Administration	Low	Little details of the exposure administration were reported.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if the test concentrations were measured at any point.	
	Metric 10: Exposure Duration and Frequency	High	The study duration was reported to be up 120hpf. Samples were also taken at 24hpf. This was adequate to observe the outcomes of interest.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	It was reported there were five test concentrations that increased by a factor of 10. There were two replicates per test concentration.	
	Metric 12: Testing at or Below Solubility Limit	High	Test concentrations were below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Medium	It was reported that zebrafish were raised at Oregon State University Sinnhuber Aquatic Laboratory in Corvallis, OR. It is unclear if the zebrafish were obtained from another lab and raised solely for this study, or if they were cultured at the facility long term.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if zebrafish were acclimated at any point.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 32 total embryos exposed to each test concentration split into two replicates.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Little details were reported regarding organism housing or environmental conditions.	
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Study Citation:	Thomas, D. G., Shankaran, H., Truong, L., Tanguay, R. L., Waters, K. M. (2019). Time-dependent behavioral data from zebrafish reveals novel signatures of chemical toxicity using point of departure analysis. Computational Toxicology 9:50-60.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; Embryo			
Health Outcome:	Immobilization			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	8635978			
Domain	Metric	Rating	Comments	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest-no response to touch at 120hpf as reported in the supplementary material.	
	Metric 18: Consistency of Outcome Assessment	Medium	Embryos were evaluated for touch response at 120hpf during the exposure. Details regarding how mobility was evaluated were not reported in detail.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	Details regarding environmental conditions before and during the study were limited.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical analysis is reported in Section 3 "Theory and calculation."	
	Metric 22: Reporting of Data	Uninformative	This study is a reanalysis or replication (it is unclear from the text whether the experiment was repeated or whether the same data was reanalyzed) of HEROID 8591199; but the results for the same chemicals vary by up to 5 orders of magnitude for the same experimental results between the two studies. Further, the BMD10 values in Table S1 cannot follow from the LEL values reported in Table S5; endpoints with LELs at the lowest concentration tested are shown with "No effect" BMD10s. These inconsistencies render this pair of studies unacceptable for use.	
	Metric 23: Explanation of Unexpected Outcomes	Low	Variability was not reported.	
Additional Comments:	This evaluation is for the effect of DBP on zebrafish embryo mobility/immobilization. Data is reported in Table S1.This study is a reanalysis or replication (it is unclear from the text whether the experiment was repeated or whether the same data was reanalyzed) of HEROID 8591199; but the results for the same chemicals vary by up to 5 orders of magnitude for the same experimental results between the two studies. Further, the BMD10 values in Table S1 cannot follow from the LEL values reported in Table S5; endpoints with LELs at the lowest concentration tested are shown with "No effect" BMD10s. These inconsistencies render this pair of studies unacceptable for use.			

Overall Quality Determination**Uninformative**

Study Citation:	Thomas, D. G., Shankaran, H., Truong, L., Tanguay, R. L., Waters, K. M. (2019). Time-dependent behavioral data from zebrafish reveals novel signatures of chemical toxicity using point of departure analysis. Computational Toxicology 9:50-60.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; Embryo			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	8635978			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The test substance was identified by name only.	
	Metric 2: Test Substance Source	Low	The source of the test substance was not reported, nor was it reported to be analytically verified.	
	Metric 3: Test Substance Purity	Low	The purity was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of and appropriate concurrent negative control.	
	Metric 5: Negative Control Response	Low	The negative control response was not reported.	
	Metric 6: Randomized Allocation	Low	It was not reported how the embryos were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The experimental system was not reported in great detail, nor was the preparation of the test solutions.	
	Metric 8: Consistency of Exposure Administration	Low	Little details of the exposure administration were reported.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if the test concentrations were measured at any point.	
	Metric 10: Exposure Duration and Frequency	High	The study duration was reported to be up 120hpf. Samples were also taken at 24hpf. This was adequate to observe the outcomes of interest.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	It was reported there were five test concentrations that increased by a factor of 10. There were two replicates per test concentration.	
	Metric 12: Testing at or Below Solubility Limit	High	Test concentrations were below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Medium	It was reported that zebrafish were raised at Oregon State University Sinnhuber Aquatic Laboratory in Corvallis, OR. It is unclear if the zebrafish were obtained from another lab and raised solely for this study, or if they were cultured at the facility long term.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if zebrafish were acclimated at any point.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 32 total embryos exposed to each test concentration split into two replicates.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Little details were reported regarding organism housing or environmental conditions.	
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Study Citation:	Thomas, D. G., Shankaran, H., Truong, L., Tanguay, R. L., Waters, K. M. (2019). Time-dependent behavioral data from zebrafish reveals novel signatures of chemical toxicity using point of departure analysis. Computational Toxicology 9:50-60.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; Embryo			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	8635978			
Domain	Metric	Rating	Comments	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest- changes in development/growth reported as changes in morphology. All morphology endpoints assessed are reported in Table 1.	
	Metric 18: Consistency of Outcome Assessment	Medium	Embryos were evaluated for changes in morphology at 24hpf and 120hpf. The exact methods for assessing morphology were not reported in great detail.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	Details regarding environmental conditions before and during the study were limited.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical analysis is reported in Section 3 "Theory and calculation."	
	Metric 22: Reporting of Data	Uninformative	This study is a reanalysis or replication (it is unclear from the text whether the experiment was repeated or whether the same data was reanalyzed) of HEROID 8591199; but the results for the same chemicals vary by up to 5 orders of magnitude for the same experimental results between the two studies. Further, the BMD10 values in Table S1 cannot follow from the LEL values reported in Table S5; endpoints with LELs at the lowest concentration tested are shown with "No effect" BMD10s. These inconsistencies render this pair of studies unacceptable for use.	
	Metric 23: Explanation of Unexpected Outcomes	Low	Variability was not reported.	
Additional Comments:	This evaluation is for the effect of DBP on zebrafish embryo morphology. Data is reported in Table S1.This study is a reanalysis or replication (it is unclear from the text whether the experiment was repeated or whether the same data was reanalyzed) of HEROID 8591199; but the results for the same chemicals vary by up to 5 orders of magnitude for the same experimental results between the two studies. Further, the BMD10 values in Table S1 cannot follow from the LEL values reported in Table S5; endpoints with LELs at the lowest concentration tested are shown with "No effect" BMD10s. These inconsistencies render this pair of studies unacceptable for use.			

Overall Quality Determination**Uninformative**

Study Citation:	Thomas, D. G., Shankaran, H., Truong, L., Tanguay, R. L., Waters, K. M. (2019). Time-dependent behavioral data from zebrafish reveals novel signatures of chemical toxicity using point of departure analysis. Computational Toxicology 9:50-60.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; Embryo			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	8635978			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The test substance was identified by name only.	
	Metric 2: Test Substance Source	Low	The source of the test substance was not reported, nor was it reported to be analytically verified.	
	Metric 3: Test Substance Purity	Low	The purity was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of and appropriate concurrent negative control.	
	Metric 5: Negative Control Response	Low	The negative control response was not reported.	
	Metric 6: Randomized Allocation	Low	It was not reported how the embryos were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The experimental system was not reported in great detail, nor was the preparation of the test solutions.	
	Metric 8: Consistency of Exposure Administration	Low	Little details of the exposure administration were reported.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if the test concentrations were measured at any point.	
	Metric 10: Exposure Duration and Frequency	High	The study duration was reported to be up 120hpf. Samples were also taken at 24hpf. This was adequate to observe the outcomes of interest.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	It was reported there were five test concentrations that increased by a factor of 10. There were two replicates per test concentration.	
	Metric 12: Testing at or Below Solubility Limit	High	Test concentrations were below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Medium	It was reported that zebrafish were raised at Oregon State University Sinnhuber Aquatic Laboratory in Corvallis, OR. It is unclear if the zebrafish were obtained from another lab and raised solely for this study, or if they were cultured at the facility long term.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if zebrafish were acclimated at any point.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 32 total embryos exposed to each test concentration split into two replicates.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Little details were reported regarding organism housing or environmental conditions.	
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Study Citation:	Thomas, D. G., Shankaran, H., Truong, L., Tanguay, R. L., Waters, K. M. (2019). Time-dependent behavioral data from zebrafish reveals novel signatures of chemical toxicity using point of departure analysis. Computational Toxicology 9:50-60.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; Embryo			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	8635978			
Domain	Metric	Rating	Comments	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest-mortality at both 24hpf and 120hpf as reported in the supplementary material.	
	Metric 18: Consistency of Outcome Assessment	Medium	Embryos were evaluated for mortality at 24hpf and 120hpf during the exposure. Details regarding how mortality was evaluated were not reported in detail.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	Details regarding environmental conditions before and during the study were limited.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical analysis is reported in Section 3 "Theory and calculation."	
	Metric 22: Reporting of Data	Uninformative	This study is a reanalysis or replication (it is unclear from the text whether the experiment was repeated or whether the same data was reanalyzed) of HEROID 8591199; but the results for the same chemicals vary by up to 5 orders of magnitude for the same experimental results between the two studies. Further, the BMD10 values in Table S1 cannot follow from the LEL values reported in Table S5; endpoints with LELs at the lowest concentration tested are shown with "No effect" BMD10s. These inconsistencies render this pair of studies unacceptable for use.	
	Metric 23: Explanation of Unexpected Outcomes	Low	Variability was not reported.	
Additional Comments:	This evaluation is for the effect of DBP on zebrafish embryo mortality. Data is reported in Table S1.This study is a reanalysis or replication (it is unclear from the text whether the experiment was repeated or whether the same data was reanalyzed) of HEROID 8591199; but the results for the same chemicals vary by up to 5 orders of magnitude for the same experimental results between the two studies. Further, the BMD10 values in Table S1 cannot follow from the LEL values reported in Table S5; endpoints with LELs at the lowest concentration tested are shown with "No effect" BMD10s. These inconsistencies render this pair of studies unacceptable for use.			

Overall Quality Determination**Uninformative**

Study Citation:	Truong, L., Reif, D. M., Mary, L. S., Geier, M. C., Truong, H. D., Tanguay, R. L. (2014). Multidimensional in vivo hazard assessment using zebrafish. Toxicological Sciences 137(1):212-233.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; Tropical 5D wild-type; Embryo			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	8591199			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The test substance was identified by name only.	
	Metric 2: Test Substance Source	Low	The source of the test substance was not reported. It was not reported if it was analytically verified.	
	Metric 3: Test Substance Purity	Low	The purity was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of a concurrent negative control.	
	Metric 5: Negative Control Response	High	The negative control response for mortality is reported in the supplemental material under Supplementary Figure 1.	
	Metric 6: Randomized Allocation	Low	It was not reported how the embryos were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	Embryos were statically exposed to the test chemicals in 96-well plate. Each well had one 6hpf embryo. Test concentrations were prepared using a 10-fold serial dilution. DMSO was used as a solvent in the embryo medium.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were conducted consistently across study groups. All exposures were conducted statically in 96-well plates under similar environmental conditions.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if the test substance was measured at any point in the study, nor were any values reported.	
	Metric 10: Exposure Duration and Frequency	High	The exposure administration was reported to be 120hpf. Embryos were also sampled at 24hpf for assessment of some parameters. These time points were adequate for a response.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	There were five exposure groups, each separated by an order of magnitude. This was adequate to observe a response and to compare responses between chemicals.	
	Metric 12: Testing at or Below Solubility Limit	High	Test concentrations were below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	It was reported that tropical 5D wild-type zebrafish were housed in a density of 1000 fish per 100 gallon tank at Sinnhuber Aquatic Research Laboratory, Oregon State University, Corvallis, OR. However, it is unclear if the fish were cultured here or obtained from another source. Embryos were used for this study.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	Test conditions and holding conditions appeared similar.	

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Study Citation:	Truong, L., Reif, D. M., Mary, L. S., Geier, M. C., Truong, H. D., Tanguay, R. L. (2014). Multidimensional in vivo hazard assessment using zebrafish. Toxicological Sciences 137(1):212-233.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; Tropical 5D wild-type; Embryo			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	8591199			
Domain	Metric	Rating	Comments	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There was one embryo placed in each well, and there were 32 replicates for each test concentration.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Adult fish were housed at 28C with 1000 fish in 100 gallon tanks. Fish were housed in reverse osmosis water supplemented with Instant Ocean, but other water characteristics were not reported. A 14L:10D photoperiod was used. The feeding regimen was not reported. Embryos were tested at 28C in standard embryo media, but characteristics of this were not reported.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest-mortality.	
	Metric 18: Consistency of Outcome Assessment	High	Outcomes appeared to be assessed consistently across study groups using PRAT.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	Embryos were held at a similar temperature to holding conditions.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical analysis was performed using code developed in R. Details of this can be found in the Analysis section.	
	Metric 22: Reporting of Data	Uninformative	This study was reanalyzed or replicated (it is unclear from the text whether the experiment was repeated or whether the same data was reanalyzed) in HEROID 8635978; but the results for the same chemicals vary by up to 5 orders of magnitude for the same experimental results between the two studies. Several of the phthalates exhibited no observable effect at any concentration studied in this study, but LELs at concentrations 10,000 times lower in 8635978 using identical methodology. These inconsistencies render this pair of studies unacceptable for use.	
	Metric 23: Explanation of Unexpected Outcomes	Low	Variability was not reported.	
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Study Citation:	Truong, L., Reif, D. M., Mary, L. S., Geier, M. C., Truong, H. D., Tanguay, R. L. (2014). Multidimensional in vivo hazard assessment using zebrafish. Toxicological Sciences 137(1):212-233.		
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; Tropical 5D wild-type; Embryo		
Health Outcome:	Mortality		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	8591199		
Domain	Metric	Rating	Comments
Additional Comments:	This evaluation is for the effect of DBP on embryo mortality after exposure to five test concentrations increasing by 10 fold. Embryos were exposed starting at 6hpf to 120hpf. Mortality data can be found in Supplemental Figure 1. This study was reanalyzed or replicated (it is unclear from the text whether the experiment was repeated or whether the same data was reanalyzed) in HERO ID 8635978; but the results for the same chemicals vary by up to 5 orders of magnitude for the same experimental results between the two studies. Several of the phthalates exhibited no observable effect at any concentration studied in this study, but LELs at concentrations 10,000 times lower in 8635978 using identical methodology. These inconsistencies render this pair of studies unacceptable for use.		

Overall Quality Determination**Uninformative**

Study Citation:	Truong, L., Reif, D. M., Mary, L. S., Geier, M. C., Truong, H. D., Tanguay, R. L. (2014). Multidimensional in vivo hazard assessment using zebrafish. Toxicological Sciences 137(1):212-233.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; Tropical 5D wild-type; Embryo			
Health Outcome:	Behavioral			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	8591199			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The test substance was identified by name only.	
	Metric 2: Test Substance Source	Low	The source of the test substance was not reported. It was not reported if it was analytically verified.	
	Metric 3: Test Substance Purity	Low	The purity was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of a concurrent negative control.	
	Metric 5: Negative Control Response	High	The negative control responses for behavioral outcomes are reported in the supplemental material under Supplementary Figure 1.	
	Metric 6: Randomized Allocation	Low	It was not reported how the embryos were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	Embryos were statically exposed to the test chemicals in 96-well plate. Each well had one 6hpf embryo. Test concentrations were prepared using a 10-fold serial dilution. DMSO was used as a solvent in the embryo medium.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were conducted consistently across study groups. All exposures were conducted statically in 96-well plates under similar environmental conditions.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if the test substance was measured at any point in the study, nor were any values reported.	
	Metric 10: Exposure Duration and Frequency	High	The exposure administration was reported to be 120hpf. Embryos were also sampled at 24hpf for assessment of some parameters. These time points were adequate for a response.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	There were five exposure groups, each separated by an order of magnitude. This was adequate to observe a response and to compare responses between chemicals.	
	Metric 12: Testing at or Below Solubility Limit	High	Test concentrations were below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	It was reported that tropical 5D wild-type zebrafish were housed in a density of 1000 fish per 100 gallon tank at Sinnhuber Aquatic Research Laboratory, Oregon State University, Corvallis, OR. However, it is unclear if the fish were cultured here or obtained from another source. Embryos were used for this study.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	Test conditions and holding conditions appeared similar.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There was one embryo placed in each well, and there were 32 replicates for each test concentration.	

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Study Citation:	Truong, L., Reif, D. M., Mary, L. S., Geier, M. C., Truong, H. D., Tanguay, R. L. (2014). Multidimensional in vivo hazard assessment using zebrafish. Toxicological Sciences 137(1):212-233.		
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; Tropical 5D wild-type; Embryo		
Health Outcome:	Behavioral		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	8591199		
Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
Metric 16:	Adequacy of Test Conditions	Low	Adult fish were housed at 28C with 1000 fish in 100 gallon tanks. Fish were housed in reverse osmosis water supplemented with Instant Ocean, but other water characteristics were not reported. A 14L:10D photoperiod was used. The feeding regimen was not reported. Embryos were tested at 28C in standard embryo media, but characteristics of this were not reported.
Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest-changes in behavior in terms of spontaneous movement and touch response.
Metric 18:	Consistency of Outcome Assessment	High	Outcomes appeared to be assessed consistently across study groups using PRAT.
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	High	Embryos were held at a similar temperature to holding conditions.
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	High	Statistical analysis was performed using code developed in R. Details of this can be found in the Analysis section.
Metric 22:	Reporting of Data	Uninformative	This study was reanalyzed or replicated (it is unclear from the text whether the experiment was repeated or whether the same data was reanalyzed) in HERO ID 8635978; but the results for the same chemicals vary by up to 5 orders of magnitude for the same experimental results between the two studies. Several of the phthalates exhibited no observable effect at any concentration studied in this study, but LELs at concentrations 10,000 times lower in 8635978 using identical methodology. These inconsistencies render this pair of studies unacceptable for use.
Metric 23:	Explanation of Unexpected Outcomes	Low	Variability was not reported.
Additional Comments:	This evaluation is for the effect of DBP on behavior after exposure to five test concentrations increasing by 10 fold. Embryos were exposed starting at 6hpf to 120hpf. Behavioral data can be found in Supplemental Figure 1. Behavioral responses assessed include spontaneous movement and touch response. This study was reanalyzed or replicated (it is unclear from the text whether the experiment was repeated or whether the same data was reanalyzed) in HERO ID 8635978; but the results for the same chemicals vary by up to 5 orders of magnitude for the same experimental results between the two studies. Several of the phthalates exhibited no observable effect at any concentration studied in this study, but LELs at concentrations 10,000 times lower in 8635978 using identical methodology. These inconsistencies render this pair of studies unacceptable for use.		

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Study Citation:	Truong, L., Reif, D. M., Mary, L. S., Geier, M. C., Truong, H. D., Tanguay, R. L. (2014). Multidimensional in vivo hazard assessment using zebrafish. Toxicological Sciences 137(1):212-233.		
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; Tropical 5D wild-type; Embryo		
Health Outcome:	Behavioral		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	8591199		
Domain	Metric	Rating	Comments
Overall Quality Determination		Uninformative	

Study Citation:	Truong, L., Reif, D. M., Mary, L. S., Geier, M. C., Truong, H. D., Tanguay, R. L. (2014). Multidimensional in vivo hazard assessment using zebrafish. Toxicological Sciences 137(1):212-233.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; Tropical 5D wild-type; Embryo			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	8591199			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The test substance was identified by name only.	
	Metric 2: Test Substance Source	Low	The source of the test substance was not reported. It was not reported if it was analytically verified.	
	Metric 3: Test Substance Purity	Low	The purity was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of a concurrent negative control.	
	Metric 5: Negative Control Response	High	The negative control responses for several development and growth outcomes are reported in the supplemental material under Supplementary Figure 1.	
	Metric 6: Randomized Allocation	Low	It was not reported how the embryos were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	Embryos were statically exposed to the test chemicals in 96-well plate. Each well had one 6hpf embryo. Test concentrations were prepared using a 10-fold serial dilution. DMSO was used as a solvent in the embryo medium.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were conducted consistently across study groups. All exposures were conducted statically in 96-well plates under similar environmental conditions.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if the test substance was measured at any point in the study, nor were any values reported.	
	Metric 10: Exposure Duration and Frequency	High	The exposure administration was reported to be 120hpf. Embryos were also sampled at 24hpf for assessment of some parameters. These time points were adequate for a response.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	There were five exposure groups, each separated by an order of magnitude. This was adequate to observe a response and to compare responses between chemicals.	
	Metric 12: Testing at or Below Solubility Limit	High	Test concentrations were below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	It was reported that tropical 5D wild-type zebrafish were housed in a density of 1000 fish per 100 gallon tank at Sinnhuber Aquatic Research Laboratory, Oregon State University, Corvallis, OR. However, it is unclear if the fish were cultured here or obtained from another source. Embryos were used for this study.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	Test conditions and holding conditions appeared similar.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There was one embryo placed in each well, and there were 32 replicates for each test concentration.	

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Study Citation:	Truong, L., Reif, D. M., Mary, L. S., Geier, M. C., Truong, H. D., Tanguay, R. L. (2014). Multidimensional in vivo hazard assessment using zebrafish. Toxicological Sciences 137(1):212-233.		
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; Tropical 5D wild-type; Embryo		
Health Outcome:	Development/Growth		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	8591199		
Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
Metric 16:	Adequacy of Test Conditions	Low	Adult fish were housed at 28C with 1000 fish in 100 gallon tanks. Fish were housed in reverse osmosis water supplemented with Instant Ocean, but other water characteristics were not reported. A 14L:10D photoperiod was used. The feeding regimen was not reported. Embryos were tested at 28C in standard embryo media, but characteristics of this were not reported.
Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest-several development and growth parameters.
Metric 18:	Consistency of Outcome Assessment	High	Outcomes appeared to be assessed consistently across study groups using PRAT.
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	High	Embryos were held at a similar temperature to holding conditions.
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	High	Statistical analysis was performed using code developed in R. Details of this can be found in the Analysis section.
Metric 22:	Reporting of Data	Uninformative	This study was reanalyzed or replicated (it is unclear from the text whether the experiment was repeated or whether the same data was reanalyzed) in HERO ID 8635978; but the results for the same chemicals vary by up to 5 orders of magnitude for the same experimental results between the two studies. Several of the phthalates exhibited no observable effect at any concentration studied in this study, but LELs at concentrations 10,000 times lower in 8635978 using identical methodology. These inconsistencies render this pair of studies unacceptable for use.
Metric 23:	Explanation of Unexpected Outcomes	Low	Variability was not reported.
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Study Citation:	Truong, L., Reif, D. M., Mary, L. S., Geier, M. C., Truong, H. D., Tanguay, R. L. (2014). Multidimensional in vivo hazard assessment using zebrafish. <i>Toxicological Sciences</i> 137(1):212-233.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; Tropical 5D wild-type; Embryo
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	8591199

Domain	Metric	Rating	Comments
Additional Comments:	This evaluation is for the effect of DBP on embryo growth and development after exposure to five test concentrations increasing by 10 fold. Embryos were exposed starting at 6hpf to 120hpf. Development and growth data can be found in Supplemental Figure 1. These parameters include developmental delay and developmental changes in the notochord, yolk sac edema, body axis, eye defect, snout, jaw, otic vesicle, pericardial edema, brain, somite, pectoral fin, caudal fin, pigment, circulation, truncated body, swim bladder, and bent tail. This study was reanalyzed or replicated (it is unclear from the text whether the experiment was repeated or whether the same data was reanalyzed) in HERO ID 8635978; but the results for the same chemicals vary by up to 5 orders of magnitude for the same experimental results between the two studies. Several of the phthalates exhibited no observable effect at any concentration studied in this study, but LELs at concentrations 10,000 times lower in 8635978 using identical methodology. These inconsistencies render this pair of studies unacceptable for use.		

Overall Quality Determination**Uninformative**

Study Citation:	Ortiz-Zarragoitia, M., Trant, J. M., Cajaravillet, M. P. (2006). Effects of dibutylphthalate and ethynylestradiol on liver peroxisomes, reproduction, and development of zebrafish (<i>Danio rerio</i>). Environmental Toxicology and Chemistry 25(9):2394-2404.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; wild type; Adult			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Oxidative stress (including redox biology)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	676322			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The chemical was identified by name only.
	Metric 2:	Test Substance Source	High	The test substance was obtained from Sigma.
	Metric 3:	Test Substance Purity	High	The chemical purity was reported as >98%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	Medium	There was no significant reported mortality. The other measurements appeared normal.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations and concentrations were not measured during the study. The exposure was conducted in 7-L glass aquaria.
	Metric 8:	Consistency of Exposure Administration	High	The vehicle solvent was dimethylformamide at 6 ul/L. The static renewal with stock solution was performed daily.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Only two concentrations were used. The exposure spacing was adequate.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Medium	All pretreatment conditions were most likely the same for control and exposed organisms, but few details were reported.
	Metric 15:	Number of Organisms and Replicates per Group	Low	There were no replicates and only 10 organisms were used.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Aquaria were maintained at 28C with conditions as appropriate for <i>D. rerio</i> at a 14:10 light cycle. They were fed three times per day.
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Study Citation:	Ortiz-Zarragoitia, M., Trant, J. M., Cajaravillet, M. P. (2006). Effects of dibutylphthalate and ethynylestradiol on liver peroxisomes, reproduction, and development of zebrafish (<i>Danio rerio</i>). Environmental Toxicology and Chemistry 25(9):2394-2404.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; wild type; Adult
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Oxidative stress (including redox biology)
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	676322

Domain	Metric	Rating	Comments
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	All aquaria were maintained at 28C with conditions as appropriate for <i>D. rerio</i> at a 14:10 light cycle. They were fed three times per day.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This evaluation is for experiment 2.

Overall Quality Determination

High

Study Citation:	Ortiz-Zarragoitia, M., Trant, J. M., Cajaravillet, M. P. (2006). Effects of dibutylphthalate and ethynylestradiol on liver peroxisomes, reproduction, and development of zebrafish (<i>Danio rerio</i>). Environmental Toxicology and Chemistry 25(9):2394-2404.			
Duration:	Overall Duration: > 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; wild type; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	676322			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	Chemical was identified by name only	
	Metric 2: Test Substance Source	High	The test substance was obtained from Sigma.	
	Metric 3: Test Substance Purity	High	Chemical purity reported as >98%	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group	
	Metric 5: Negative Control Response	Low	no significant reported mortality, reproductive output not reported	
	Metric 6: Randomized Allocation	Low	Researchers did not report how F0 organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations and concentrations were not measured during the study. Exposure to the P generation was conducted in 7-L glass aquaria, and the F1 offspring were transferred to 1.5-L glass vessels for observation.	
	Metric 8: Consistency of Exposure Administration	High	Vehicle was dimethylformamide at 6 ul/L and static renewal with stock solution was performed daily.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured	
	Metric 10: Exposure Duration and Frequency	High	F1 embryos were not individually exposed, they were the offspring of P generation exposed for 15 days.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Only two concentrations were used, spacing was adequate	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source	
	Metric 14: Acclimatization and Pretreatment Conditions	Medium	all pretreatment conditions were most likely the same for control and exposed organisms but few details were reported	
	Metric 15: Number of Organisms and Replicates per Group	Low	No replicates and only 10 F0 organisms were used	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Aquaria were maintained at 28C with conditions as appropriate for <i>D. rerio</i> at a 14:10 light cycle and fed three times per day.	
	Metric 17: Outcome Assessment Methodology	Medium	The details of outcome assessment methodology were not reported	
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Study Citation:	Ortiz-Zarragoitia, M., Trant, J. M., Cajaravillet, M. P. (2006). Effects of dibutylphthalate and ethynylestradiol on liver peroxisomes, reproduction, and development of zebrafish (<i>Danio rerio</i>). <i>Environmental Toxicology and Chemistry</i> 25(9):2394-2404.
Duration:	Overall Duration: > 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; wild type; Juvenile
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	676322

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	Medium	outcomes were assessed consistently across study groups but few details were provided
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical analysis was performed and was well-described (two-way ANOVA followed by Duncan's test after testing for normality and homogeneity of variance, or nonparametric ANOVA via Kruskal-Wallis followed by Mann-Whitney U test.
	Metric 22: Reporting of Data	Low	Data for exposure-related findings were presented for only the high concentration
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: F1 survival, experiment 2

Overall Quality Determination**Medium**

Study Citation:	Ortiz-Zarragoitia, M., Trant, J. M., Cajaravillet, M. P. (2006). Effects of dibutylphthalate and ethynylestradiol on liver peroxisomes, reproduction, and development of zebrafish (<i>Danio rerio</i>). Environmental Toxicology and Chemistry 25(9):2394-2404.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; wild type; Adult			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	676322			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	Chemical was identified by name only	
	Metric 2: Test Substance Source	High	The test substance was obtained from Sigma.	
	Metric 3: Test Substance Purity	High	Chemical purity reported as >98%	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group	
	Metric 5: Negative Control Response	Low	no significant reported mortality, reproductive output not reported	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations and concentrations were not measured during the study. Exposure was conducted in 7-L glass aquaria.	
	Metric 8: Consistency of Exposure Administration	High	Vehicle was dimethylformamide at 6 ul/L and static renewal with stock solution was performed daily.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured	
	Metric 10: Exposure Duration and Frequency	Medium	The duration of exposure was suitable for the study type, reproductive success was not the primary outcome	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Only two concentrations were used, spacing was adequate	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source	
	Metric 14: Acclimatization and Pretreatment Conditions	Medium	all pretreatment conditions were most likely the same for control and exposed organisms but few details were reported	
	Metric 15: Number of Organisms and Replicates per Group	Low	No replicates and only 10 organisms were used	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Aquaria were maintained at 28C with conditions as appropriate for <i>D. rerio</i> at a 14:10 light cycle and fed three times per day.	
	Metric 17: Outcome Assessment Methodology	Medium	The details of outcome assessment methodology were not reported	
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Study Citation:	Ortiz-Zarragoitia, M., Trant, J. M., Cajaravillet, M. P. (2006). Effects of dibutylphthalate and ethynylestradiol on liver peroxisomes, reproduction, and development of zebrafish (<i>Danio rerio</i>). <i>Environmental Toxicology and Chemistry</i> 25(9):2394-2404.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; wild type; Adult
Health Outcome:	Reproductive/Teratogenic
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	676322

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	Medium	outcomes were assessed consistently across study groups but few details were provided
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	All aquaria were maintained at 28C with conditions as appropriate for <i>D. rerio</i> at a 14:10 light cycle and fed three times per day.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical analysis was performed and was well-described (two-way ANOVA followed by Duncan's test after testing for normality and homogeneity of variance, or nonparametric ANOVA via Kruskal-Wallis followed by Mann-Whitney U test.
	Metric 22: Reporting of Data	Low	Data for exposure-related findings were presented for each treatment but not the control group
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: F0 reproductive output, experiment 2

Overall Quality Determination**Medium**

Study Citation:	Chen, H., Feng, W., Chen, K., Qiu, X., Xu, H., Mao, G., Zhao, T., Ding, Y., Wu, X. (2019). Transcriptomic analysis reveals potential mechanisms of toxicity in a combined exposure to dibutyl phthalate and diisobutyl phthalate in zebrafish (<i>Danio rerio</i>) ovary. <i>Aquatic Toxicology</i> 216:105290.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; Wild type AB strain; Adult			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	6959356			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The DBP was identified by CASRN.
	Metric 2:	Test Substance Source	Low	The DBP and the DIBP were both from Sigma-Aldrich, but it was not reported if they were analytically verified.
	Metric 3:	Test Substance Purity	High	The purity was reported to be 99%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of a negative control using dechlorinated tap water as well as a solvent control containing DMSO.
	Metric 5:	Negative Control Response	High	The negative control responses were reported in Figure 1 for condition factor.
	Metric 6:	Randomized Allocation	Low	It was not reported how the zebrafish were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The exposure system was reported to be static with 50% of the experimental media replaced every 24h. Little information was provided on the preparation of the test concentrations.
	Metric 8:	Consistency of Exposure Administration	High	All exposure were conducted in 10L tanks with six adult female zebrafish. Dechlorinated tap water was used as the dilution water in all exposure groups. The exposure appeared to be conducted consistently across exposure groups.
	Metric 9:	Measurement of Test Substance Concentration	Medium	The test substance was analyzed before and after water exchanges weekly. Measured test concentrations were reported in Table S1. The analytical method was cited to another source and was not reported in this paper.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration was reported to be for 30 days. This was adequate to observe a response.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	There were three exposure groups of DBP. This is slightly lower than is typical, but comparisons were made with other test chemicals, and three exposure groups were adequate for this purpose.
	Metric 12:	Testing at or Below Solubility Limit	High	All of the DBP exposure levels were below the water solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Adult female zebrafish were reported to be from the China Zebrafish Center in Wuhan, China. They were reported to be wild-type AB strain.
	Metric 14:	Acclimatization and Pretreatment Conditions	Medium	It was reported that there was an adaptation period before the chemical exposure. However, the length of this period and the conditions were unclear.
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Study Citation:	Chen, H., Feng, W., Chen, K., Qiu, X., Xu, H., Mao, G., Zhao, T., Ding, Y., Wu, X. (2019). Transcriptomic analysis reveals potential mechanisms of toxicity in a combined exposure to dibutyl phthalate and diisobutyl phthalate in zebrafish (<i>Danio rerio</i>) ovary. <i>Aquatic Toxicology</i> 216:105290.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; Wild type AB strain; Adult
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	6959356

Domain	Metric	Rating	Comments
	Metric 15: Number of Organisms and Replicates per Group	Medium	It was reported there were six female zebrafish in each test chamber with three replicates per treatment.
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	Medium	The fish were maintained in the performing laboratory for one year prior to exposure at 27C with a 14:10 light:dark photoperiod under flow-through conditions. Feeding regimen and water quality were reported. It was unclear if the testing conditions were the same.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest- changes in condition factor as determined by wet weight and body length.
	Metric 18: Consistency of Outcome Assessment	High	The outcome assessment methodology was described adequately and performed consistently. Wet weights and body lengths were recorded after the 30d exposure.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Medium	Culture conditions were described in detail, but little was reported on testing conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical analysis was reported in Section 2.9 and was adequate.
	Metric 22: Reporting of Data	High	Data was reported for the control and exposure groups. Condition factor was reported in Figure 1A.
	Metric 23: Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.
Additional Comments:	This evaluation was for the growth outcome reported after adult female zebrafish were exposed to DBP. Condition factor was determined based off of wet weight and body length after the 30d exposure.		

Overall Quality Determination**High**

Study Citation:	Chen, H., Feng, W., Chen, K., Qiu, X., Xu, H., Mao, G., Zhao, T., Ding, Y., Wu, X. (2019). Transcriptomic analysis reveals potential mechanisms of toxicity in a combined exposure to dibutyl phthalate and diisobutyl phthalate in zebrafish (<i>Danio rerio</i>) ovary. <i>Aquatic Toxicology</i> 216:105290.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; Wild type AB strain; Adult			
Health Outcome:	Mechanistic-Cell signaling/function-Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	6959356			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The DBP was identified by CASRN.	
	Metric 2: Test Substance Source	Low	The DBP and the DIBP were both from Sigma-Aldrich, but it was not reported if they were analytically verified.	
	Metric 3: Test Substance Purity	High	The purity was reported to be 99%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of a negative control using dechlorinated tap water as well as a solvent control containing DMSO.	
	Metric 5: Negative Control Response	Low	The negative control responses for reproductive hormone levels were reported in Fig. 4. Negative control responses were not reported for changes in gene expression, but comparisons were implied.	
	Metric 6: Randomized Allocation	Low	It was not reported how the zebrafish were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The exposure system was reported to be static with 50% of the experimental media replaced every 24h. Little information was provided on the preparation of the test concentrations.	
	Metric 8: Consistency of Exposure Administration	High	All exposure were conducted in 10L tanks with six adult female zebrafish. Dechlorinated tap water was used as the dilution water in all exposure groups. The exposure appeared to be conducted consistently across exposure groups.	
	Metric 9: Measurement of Test Substance Concentration	Medium	The test substance was analyzed before and after water exchanges weekly. Measured test concentrations were reported in Table S1. The analytical method was cited to another source and was not reported in this paper.	
	Metric 10: Exposure Duration and Frequency	High	The exposure duration was reported to be for 30 days. This was adequate to observe a response.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Medium	There were three exposure groups of DBP. This is slightly lower than is typical, but comparisons were made with other test chemicals, and three exposure groups were adequate for this purpose.	
	Metric 12: Testing at or Below Solubility Limit	High	All of the DBP exposure levels were below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	Adult female zebrafish were reported to be from the China Zebrafish Center in Wuhan, China. They were reported to be wild-type AB strain.	
	Metric 14: Acclimatization and Pretreatment Conditions	Medium	It was reported that there was an adaptation period before the chemical exposure. However, the length of this period and the conditions were unclear.	
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Study Citation:	Chen, H., Feng, W., Chen, K., Qiu, X., Xu, H., Mao, G., Zhao, T., Ding, Y., Wu, X. (2019). Transcriptomic analysis reveals potential mechanisms of toxicity in a combined exposure to dibutyl phthalate and diisobutyl phthalate in zebrafish (<i>Danio rerio</i>) ovary. <i>Aquatic Toxicology</i> 216:105290.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; Wild type AB strain; Adult			
Health Outcome:	Mechanistic-Cell signaling/function-Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	6959356			
Domain	Metric	Rating	Comments	
	Metric 15: Number of Organisms and Replicates per Group	Medium	It was reported there were six female zebrafish in each test chamber with three replicates per treatment.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Medium	The fish were maintained in the performing laboratory for one year prior to exposure at 27C with a 14:10 light:dark photoperiod under flow-through conditions. Feeding regimen and water quality were reported. It was unclear if the testing conditions were the same.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest- changes in plasma E2 and T levels as well as changes in gene expression due to exposure.	
	Metric 18: Consistency of Outcome Assessment	High	The outcome assessment methodology was described adequately and performed consistently.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Medium	Culture conditions were described in detail, but little was reported on testing conditions.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical analysis was reported in Section 2.9 and was adequate.	
	Metric 22: Reporting of Data	High	Data was reported for the control and exposure groups. Plasma hormone levels were reported in Figure 4. Changes in gene expression were reported in Tables 2-5.	
	Metric 23: Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.	
Additional Comments:	This evaluation was for the mechanistic outcomes reported in this paper. Reproductive plasma hormones (E2 and T) levels were assessed after a 30d exposure to DBP. Changes in gene expression were also assessed.			
Overall Quality Determination		High		

Study Citation:	Chen, H., Feng, W., Chen, K., Qiu, X., Xu, H., Mao, G., Zhao, T., Ding, Y., Wu, X. (2019). Transcriptomic analysis reveals potential mechanisms of toxicity in a combined exposure to dibutyl phthalate and diisobutyl phthalate in zebrafish (<i>Danio rerio</i>) ovary. <i>Aquatic Toxicology</i> 216:105290.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; Wild type AB strain; Adult			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	6959356			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The DBP was identified by CASRN.	
	Metric 2: Test Substance Source	Low	The DBP and the DIBP were both from Sigma-Aldrich, but it was not reported if they were analytically verified.	
	Metric 3: Test Substance Purity	High	The purity was reported to be 99%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of a negative control using dechlorinated tap water as well as a solvent control containing DMSO.	
	Metric 5: Negative Control Response	High	The negative control responses were reported in Figure 1 for GSI and Figures 2 and 3 for histological evaluation.	
	Metric 6: Randomized Allocation	Low	It was not reported how the zebrafish were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The exposure system was reported to be static with 50% of the experimental media replaced every 24h. Little information was provided on the preparation of the test concentrations.	
	Metric 8: Consistency of Exposure Administration	High	All exposure were conducted in 10L tanks with six adult female zebrafish. Dechlorinated tap water was used as the dilution water in all exposure groups. The exposure appeared to be conducted consistently across exposure groups.	
	Metric 9: Measurement of Test Substance Concentration	Medium	The test substance was analyzed before and after water exchanges weekly. Measured test concentrations were reported in Table S1. The analytical method was cited to another source and was not reported in this paper.	
	Metric 10: Exposure Duration and Frequency	High	The exposure duration was reported to be for 30 days. This was adequate to observe a response.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Medium	There were three exposure groups of DBP. This is slightly lower than is typical, but comparisons were made with other test chemicals, and three exposures groups were adequate for this purpose.	
	Metric 12: Testing at or Below Solubility Limit	High	All of the DBP exposure levels were below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	Adult female zebrafish were reported to be from the China Zebrafish Center in Wuhan, China. They were reported to be wild-type AB strain.	
	Metric 14: Acclimatization and Pretreatment Conditions	Medium	It was reported that there was an adaptation period before the chemical exposure. However, the length of this period and the conditions were unclear.	
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Study Citation:	Chen, H., Feng, W., Chen, K., Qiu, X., Xu, H., Mao, G., Zhao, T., Ding, Y., Wu, X. (2019). Transcriptomic analysis reveals potential mechanisms of toxicity in a combined exposure to dibutyl phthalate and diisobutyl phthalate in zebrafish (Danio rerio) ovary. Aquatic Toxicology 216:105290.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; Wild type AB strain; Adult			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	6959356			
Domain	Metric	Rating	Comments	
	Metric 15: Number of Organisms and Replicates per Group	Medium	It was reported there were six female zebrafish in each test chamber with three replicates per treatment.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Medium	The fish were maintained in the performing laboratory for one year prior to exposure at 27C with a 14:10 light:dark photoperiod under flow-through conditions. Feeding regimen and water quality were reported. It was unclear if the testing conditions were the same.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest- changes in GSI and ovary histology.	
	Metric 18: Consistency of Outcome Assessment	High	The outcome assessment methodology was described adequately and performed consistently. Histological analysis was conducted according to previous research. Basic methods were described in section 2.5.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Medium	Culture conditions were described in detail, but little was reported on testing conditions.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical analysis was reported in Section 2.9 and was adequate.	
	Metric 22: Reporting of Data	High	Data was reported for the control and exposure groups. GSI was reported in Figure 1B. Data for the histological outcomes was reported in Fig 3 with statistical analysis and in Figure 2 with histological sections.	
	Metric 23: Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.	
Additional Comments:	This evaluation was for the reproductive outcomes reported after adult female zebrafish were exposed to DBP. Reproductive outcomes assessed were GSI and histological outcomes. Data for these outcomes were reported in in Figure 1B, 2 and 3.			
Overall Quality Determination		High		

Study Citation:	Chen, P., Li, S., Liu, L., Xu, N. (2015). Long-term effects of binary mixtures of 17 α -ethinyl estradiol and dibutyl phthalate in a partial life-cycle test with zebrafish (<i>Danio rerio</i>). Environmental Toxicology and Chemistry 34(3):518-526.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; TU strain; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Respiratory			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2816885			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	Chemical was identified by name only	
	Metric 2: Test Substance Source	High	The test substance identity was verified by GC-MS.	
	Metric 3: Test Substance Purity	High	Chemical purity reported as >98%	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group (solvent control containing 0.005% DMSO).	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups using a semi-static method with half-renewal daily.	
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Medium	Only two treatments but at a suitable range	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized	
	Metric 15: Number of Organisms and Replicates per Group	Medium	7-12 fish per tank with 4 replicates	
Domain 5: Outcome Assessment				
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Study Citation:	Chen, P., Li, S., Liu, L., Xu, N. (2015). Long-term effects of binary mixtures of 17 α -ethinyl estradiol and dibutyl phthalate in a partial life-cycle test with zebrafish (<i>Danio rerio</i>). Environmental Toxicology and Chemistry 34(3):518-526.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; TU strain; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Respiratory			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2816885			
Domain	Metric	Rating	Comments	
	Metric 16:	Adequacy of Test Conditions	Medium	Environmental conditions of test system were most likely conducive to maintenance of organism health but actual measured condition values for control and exposed vessels was not reported
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes
Additional Comments:	This form is to account for the gill histology.			
Overall Quality Determination		High		

Study Citation:	Chen, P., Li, S., Liu, L., Xu, N. (2015). Long-term effects of binary mixtures of 17 α -ethinyl estradiol and dibutyl phthalate in a partial life-cycle test with zebrafish (<i>Danio rerio</i>). Environmental Toxicology and Chemistry 34(3):518-526.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; TU strain; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2816885			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	High	The test substance identity was verified by GC-MS.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group (solvent control containing 0.005% DMSO).
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups using a semi-static method with half-renewal daily.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Only two treatments but at a suitable range
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized
	Metric 15:	Number of Organisms and Replicates per Group	Medium	7-12 fish per tank with 4 replicates
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Environmental conditions of test system were most likely conducive to maintenance of organism health but actual measured condition values for control and exposed vessels was not reported
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
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Study Citation:	Chen, P., Li, S., Liu, L., Xu, N. (2015). Long-term effects of binary mixtures of 17 α -ethinyl estradiol and dibutyl phthalate in a partial life-cycle test with zebrafish (<i>Danio rerio</i>). Environmental Toxicology and Chemistry 34(3):518-526.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; TU strain; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	2816885

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	High	mortalities were checked daily
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group (see supplemental data sheet)
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes
Additional Comments:	None		

Overall Quality Determination**High**

Study Citation:	Chen, P., Li, S., Liu, L., Xu, N. (2015). Long-term effects of binary mixtures of 17 α -ethinyl estradiol and dibutyl phthalate in a partial life-cycle test with zebrafish (<i>Danio rerio</i>). Environmental Toxicology and Chemistry 34(3):518-526.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; TU strain; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2816885			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The chemical was identified by name only.
	Metric 2:	Test Substance Source	High	The test substance identity was verified by GC-MS.
	Metric 3:	Test Substance Purity	High	Chemical purity was reported as >98%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group (solvent control containing 0.005% DMSO).
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups using a semi-static method with half-renewal daily.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Only two treatments were used, but they were at a suitable range.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	There were 7-12 fish per tank with four replicates.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Environmental conditions of the test system were most likely conducive to maintenance of organism health but actual measured condition values for control and exposed vessels were not reported.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
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Study Citation:	Chen, P., Li, S., Liu, L., Xu, N. (2015). Long-term effects of binary mixtures of 17 α -ethinyl estradiol and dibutyl phthalate in a partial life-cycle test with zebrafish (<i>Danio rerio</i>). Environmental Toxicology and Chemistry 34(3):518-526.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; TU strain; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2816885			
Domain	Metric	Rating	Comments	
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.	
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.	
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.	
Additional Comments:	None			

Overall Quality Determination**High**

Study Citation:	Chen, P., Li, S., Liu, L., Xu, N. (2015). Long-term effects of binary mixtures of 17 α -ethinyl estradiol and dibutyl phthalate in a partial life-cycle test with zebrafish (<i>Danio rerio</i>). Environmental Toxicology and Chemistry 34(3):518-526.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; TU strain; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2816885			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	High	The test substance identity was verified by GC-MS.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group (solvent control containing 0.005% DMSO).
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups using a semi-static method with half-renewal daily.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Only two treatments but at a suitable range
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized
	Metric 15:	Number of Organisms and Replicates per Group	Medium	7-12 fish per tank with 4 replicates
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Environmental conditions of test system were most likely conducive to maintenance of organism health but actual measured condition values for control and exposed vessels was not reported
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
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Study Citation:	Chen, P., Li, S., Liu, L., Xu, N. (2015). Long-term effects of binary mixtures of 17 α -ethinyl estradiol and dibutyl phthalate in a partial life-cycle test with zebrafish (<i>Danio rerio</i>). Environmental Toxicology and Chemistry 34(3):518-526.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; TU strain; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	2816885

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: length, weight, condition index

Overall Quality Determination**High**

Study Citation:	Chen, P., Li, S., Liu, L., Xu, N. (2015). Long-term effects of binary mixtures of 17 α -ethinyl estradiol and dibutyl phthalate in a partial life-cycle test with zebrafish (<i>Danio rerio</i>). Environmental Toxicology and Chemistry 34(3):518-526.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; TU strain; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Hepatic/Liver			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2816885			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	High	The test substance identity was verified by GC-MS.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group (solvent control containing 0.005% DMSO).
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups using a semi-static method with half-renewal daily.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Only two treatments but at a suitable range
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized
	Metric 15:	Number of Organisms and Replicates per Group	Medium	7-12 fish per tank with 4 replicates
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Environmental conditions of test system were most likely conducive to maintenance of organism health but actual measured condition values for control and exposed vessels was not reported
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
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Study Citation:	Chen, P., Li, S., Liu, L., Xu, N. (2015). Long-term effects of binary mixtures of 17 α -ethinyl estradiol and dibutyl phthalate in a partial life-cycle test with zebrafish (<i>Danio rerio</i>). Environmental Toxicology and Chemistry 34(3):518-526.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; TU strain; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Hepatic/Liver
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	2816885

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: This form is to account for the liver histology.

Overall Quality Determination

High

Study Citation:	Chen, P., Li, S., Liu, L., Xu, N. (2015). Long-term effects of binary mixtures of 17 α -ethinyl estradiol and dibutyl phthalate in a partial life-cycle test with zebrafish (<i>Danio rerio</i>). Environmental Toxicology and Chemistry 34(3):518-526.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; TU strain; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2816885			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	High	The test substance identity was verified by GC-MS.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group (solvent control containing 0.005% DMSO).
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups using a semi-static method with half-renewal daily.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Only two treatments but at a suitable range
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized
	Metric 15:	Number of Organisms and Replicates per Group	Medium	7-12 fish per tank with 4 replicates
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Environmental conditions of test system were most likely conducive to maintenance of organism health but actual measured condition values for control and exposed vessels was not reported
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
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Study Citation:	Chen, P., Li, S., Liu, L., Xu, N. (2015). Long-term effects of binary mixtures of 17 α -ethinyl estradiol and dibutyl phthalate in a partial life-cycle test with zebrafish (<i>Danio rerio</i>). Environmental Toxicology and Chemistry 34(3):518-526.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; TU strain; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Reproductive/Teratogenic
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	2816885

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: This form is to account for the gonad histology and the sex ratio determination, both of which are reproductive outcomes.

Overall Quality Determination

High

Study Citation:	Ortiz-Zarragoitia, M., Trant, J. M., Cajaravillet, M. P. (2006). Effects of dibutylphthalate and ethynylestradiol on liver peroxisomes, reproduction, and development of zebrafish (<i>Danio rerio</i>). Environmental Toxicology and Chemistry 25(9):2394-2404.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; wild type; Embryo			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Oxidative stress (including redox biology)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	676322			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only.
	Metric 2:	Test Substance Source	High	The test substance was obtained from Sigma.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	Medium	Reported mortality of 20-30% in both controls and DBP-exposed treatments.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations, and concentrations were not measured during the study. Exposure was conducted in 2-L glass vessels.
	Metric 8:	Consistency of Exposure Administration	High	Vehicle was dimethylformamide at 6 ul/L and static renewal with stock solution was performed daily.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Only two concentrations were used, spacing was adequate
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	Medium	all pretreatment conditions were most likely the same for control and exposed organisms but few details were reported
	Metric 15:	Number of Organisms and Replicates per Group	Medium	two replicates and enough organisms to make subsampling possible
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Aquaria were maintained at 28C with conditions as appropriate for <i>D. rerio</i> at a 14:10 light cycle and fed three times per day.
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Study Citation:	Ortiz-Zarragoitia, M., Trant, J. M., Cajaravillet, M. P. (2006). Effects of dibutylphthalate and ethynylestradiol on liver peroxisomes, reproduction, and development of zebrafish (<i>Danio rerio</i>). <i>Environmental Toxicology and Chemistry</i> 25(9):2394-2404.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; wild type; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Oxidative stress (including redox biology)
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	676322

Domain	Metric	Rating	Comments
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18: Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	All aquaria were maintained at 28C with conditions as appropriate for <i>D. rerio</i> at a 14:10 light cycle and fed three times per day.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group and at several time points
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes
Additional Comments: experiment 1			

Overall Quality Determination**High**

Study Citation:	Ortiz-Zarragoitia, M., Trant, J. M., Cajaravillet, M. P. (2006). Effects of dibutylphthalate and ethynylestradiol on liver peroxisomes, reproduction, and development of zebrafish (<i>Danio rerio</i>). <i>Environmental Toxicology and Chemistry</i> 25(9):2394-2404.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; wild type; Embryo			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	676322			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance	Metric 1: Test Substance Identity	Low	Chemical was identified by name only	
	Metric 2: Test Substance Source	High	The test substance was obtained from Sigma.	
	Metric 3: Test Substance Purity	High	Chemical purity reported as >98%	
Domain 2: Test Design	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group	
	Metric 5: Negative Control Response	Low	mortality rate of 20-30% was reported	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations, and concentrations were not measured during the study. Exposure was conducted in 2-L glass vessels.	
	Metric 8: Consistency of Exposure Administration	High	Vehicle was dimethylformamide at 6 ul/L and static renewal with stock solution was performed daily.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Only two concentrations were used, spacing was adequate	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit	
Domain 4: Test Organism	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source	
	Metric 14: Acclimatization and Pretreatment Conditions	Medium	all pretreatment conditions were most likely the same for control and exposed organisms but few details were reported	
	Metric 15: Number of Organisms and Replicates per Group	Medium	Two replicates and enough organisms to allow subsampling	
Domain 5: Outcome Assessment	Metric 16: Adequacy of Test Conditions	High	Aquaria were maintained at 28C with conditions as appropriate for <i>D. rerio</i> at a 14:10 light cycle and fed three times per day. if adequate	
	Metric 17: Outcome Assessment Methodology	Medium	The outcome assessment methodology were adequate	
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Study Citation:	Ortiz-Zarragoitia, M., Trant, J. M., Cajaravillet, M. P. (2006). Effects of dibutylphthalate and ethynylestradiol on liver peroxisomes, reproduction, and development of zebrafish (<i>Danio rerio</i>). Environmental Toxicology and Chemistry 25(9):2394-2404.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; wild type; Embryo
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	676322

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	Low	It was unclear if outcomes were assessed consistently across study groups, few details were provided
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical analysis was performed and was well-described (two-way ANOVA followed by Duncan's test after testing for normality and homogeneity of variance, or nonparametric ANOVA via Kruskal-Wallis followed by Mann-Whitney U test.
	Metric 22: Reporting of Data	Low	Data was not presented for each treatment
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: experiment 1

Overall Quality Determination**Medium**

Study Citation:	Ortiz-Zarragoitia, M., Trant, J. M., Cajaravillet, M. P. (2006). Effects of dibutylphthalate and ethynylestradiol on liver peroxisomes, reproduction, and development of zebrafish (<i>Danio rerio</i>). Environmental Toxicology and Chemistry 25(9):2394-2404.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; wild type; Embryo			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	676322			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	High	The test substance was obtained from Sigma.	
	Metric 3: Test Substance Purity	High	The chemical purity reported as >98%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	Low	A mortality rate of 20-30% was reported.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations, and concentrations were not measured during the study. The exposure was conducted in 2-L glass vessels.	
	Metric 8: Consistency of Exposure Administration	High	The vehicle solvent was dimethylformamide at 6 ul/L. Static renewal with stock solution was performed daily.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Only two concentrations were used. The exposure spacing was adequate.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	Medium	All pretreatment conditions were most likely the same for control and exposed organisms, but few details were reported.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	Two replicates were used, along with enough organisms to allow for subsampling.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Aquaria were maintained at 28C with conditions as appropriate for <i>D. rerio</i> at a 14:10 light cycle. They were fed three times per day.	
	Metric 17: Outcome Assessment Methodology	Medium	The outcome assessment methodology was adequate.	
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Study Citation:	Ortiz-Zarragoitia, M., Trant, J. M., Cajaravillet, M. P. (2006). Effects of dibutylphthalate and ethynylestradiol on liver peroxisomes, reproduction, and development of zebrafish (<i>Danio rerio</i>). <i>Environmental Toxicology and Chemistry</i> 25(9):2394-2404.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Danio rerio</i> ; wild type; Embryo
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	676322

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	Medium	Outcomes were assessed consistently across study groups, but few details were provided.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical analysis was performed and was well-described (two-way ANOVA followed by Duncan's test after testing for normality and homogeneity of variance, or nonparametric ANOVA via Kruskal-Wallis followed by Mann-Whitney U test).
	Metric 22: Reporting of Data	Low	Data was not presented for each treatment.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This evaluation is for experiment 1.

Overall Quality Determination

Medium

Study Citation:	Aoki, K. A., Harris, C. A., Katsiadaki, I., Sumpter, J. P. (2011). Evidence suggesting that di-n-butyl phthalate has antiandrogenic effects in fish. Environmental Toxicology and Chemistry 30(6):1338-1345.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Gasterosteus aculeataus</i> ; males; Adult			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	788294			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The test substance was identified by name only.
	Metric 2:	Test Substance Source	High	DBP was sourced from Fisher Scientific and was analytically verified via GC/MS.
	Metric 3:	Test Substance Purity	Low	The purity of the DBP was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	The study authors reported using a negative control with the solvent used in testing.
	Metric 5:	Negative Control Response	Low	The length, weight, and GSI of the control group fish were not specifically reported.
	Metric 6:	Randomized Allocation	Medium	Study authors reported the fish were allocated randomly into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	Little detail was provided on how the stock solutions were prepared. The system was flow through. Concentrations of test substance were measured during the study every 3-4 days. Exposure was conducted in 3.8L tanks (presumably made of glass, as is standard for aquaria).
	Metric 8:	Consistency of Exposure Administration	Medium	There were 8 fish per treatment group. A flow through system was used for all tanks. Water flow was the same for all tanks. The concentrations of DBP detected were not close to the nominal concentrations, creating some concern about consistency.
	Metric 9:	Measurement of Test Substance Concentration	High	Test concentrations were measured every 3-4 days. The reported concentrations were not close to the nominal concentrations, but study authors reported the endpoints were based on the measured concentrations.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration was reported to be for 22 days. This was adequate time to observe changes in growth and development.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	There were only 2 exposure groups plus a control. This was adequate to observe a response, but more exposure levels may have yielded more data. Spacing was adequate.
	Metric 12:	Testing at or Below Solubility Limit	High	Study authors reported using methanol as a vehicle solvent.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the fish was reported to be ponds near Hook, Hampshire, United Kingdom. This creates significant concern about the health of the organisms prior to testing.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the fish were acclimated to test conditions prior to the start of the study. The reported 10-day "acclimation period" was conducted during the test, while exposure to DBP was occurring. Therefore it cannot be considered a true acclimation.

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Study Citation:	Aoki, K. A., Harris, C. A., Katsiadaki, I., Sumpter, J. P. (2011). Evidence suggesting that di-n-butyl phthalate has antiandrogenic effects in fish. Environmental Toxicology and Chemistry 30(6):1338-1345.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Gasterosteus aculeatus</i> ; males; Adult
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	788294

Domain	Metric	Rating	Comments
	Metric 15: Number of Organisms and Replicates per Group	Low	8 fish were exposed per treatment (including control) for a total of 24 fish, exposed in individual containers.
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Fish were housed in 3.8L tanks in dechlorinated water and fed 3 times daily. Prior to treatment they were held at 8L:16hD photoperiod at 18-20C, which was winter conditions to lead to sexual maturity. They were switched to 16L:8D with higher light intensity to bring the fish to full sexual maturity.
	Metric 17: Outcome Assessment Methodology	Low	Methodology to assess length, weight and GSI was not described.
	Metric 18: Consistency of Outcome Assessment	Low	Since methods were not described for assessing length, weight, and GSI it is uncertain whether consistency occurred across all study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. It was not reported if the organisms were acclimated to test conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information to suggest any differences in organisms among groups that would impact the outcome assessment.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical analyses were described in the "Statistical Analysis" section and were described in detail.
	Metric 22: Reporting of Data	Medium	Length, weight, and GSI were briefly described in text in the results section, but there was no data shown in tables or figures. There were no significant differences found with these parameters.
	Metric 23: Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability.

Additional Comments: This evaluation is for Development/Growth assessing the analysis of length, weight, and GSI.

Overall Quality Determination

Medium

Study Citation:	Aoki, K. A., Harris, C. A., Katsiadaki, I., Sumpter, J. P. (2011). Evidence suggesting that di-n-butyl phthalate has antiandrogenic effects in fish. Environmental Toxicology and Chemistry 30(6):1338-1345.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Gasterosteus aculeatus</i> ; males; Adult			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Endocrine toxicity-Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	788294			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The test substance was identified by name only.
	Metric 2:	Test Substance Source	High	DBP was sourced from Fisher Scientific and was analytically verified via GC/MS.
	Metric 3:	Test Substance Purity	Low	The purity of the DBP was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	The study authors reported using a negative control with the solvent used in testing.
	Metric 5:	Negative Control Response	High	The solvent control response was reported in Figures 2 and 3 and was adequate for the outcome of interest.
	Metric 6:	Randomized Allocation	Medium	Study authors reported the fish were allocated randomly into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	Little detail was provided on how the stock solutions were prepared. The system was flow through. Concentrations of test substance were measured during the study every 3-4 days. Exposure was conducted in 3.8L tanks (presumably made of glass, as is standard for aquaria).
	Metric 8:	Consistency of Exposure Administration	Medium	There were 8 fish per treatment group. A flow through system was used for all tanks. Water flow was the same for all tanks. The concentrations of DBP detected were not close to the nominal concentrations, creating some concern about consistency.
	Metric 9:	Measurement of Test Substance Concentration	High	Test concentrations were measured every 3-4 days. The reported concentrations were not close to the nominal concentrations, but study authors reported the endpoints were based on the measured concentrations.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration was reported to be for 22 days. This was adequate time to observe nesting behavior in all the exposure concentration.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	There were only 2 exposure groups plus a control. This was adequate to observe a response, but more exposure levels may have yielded more data. Spacing was adequate.
	Metric 12:	Testing at or Below Solubility Limit	High	Study authors reported using methanol as a vehicle solvent.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the fish was reported to be ponds near Hook, Hampshire, United Kingdom. This creates significant concern about the health of the organisms prior to testing.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the fish were acclimated to test conditions prior to the start of the study. The reported 10-day "acclimation period" was conducted during the test, while exposure to DBP was occurring. Therefore it cannot be considered a true acclimation.
	Metric 15:	Number of Organisms and Replicates per Group	Low	8 fish were exposed per treatment (including control) for a total of 24 fish, exposed in individual containers.

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Study Citation:	Aoki, K. A., Harris, C. A., Katsiadaki, I., Sumpter, J. P. (2011). Evidence suggesting that di-n-butyl phthalate has antiandrogenic effects in fish. Environmental Toxicology and Chemistry 30(6):1338-1345.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Gasterosteus aculeatus</i> ; males; Adult			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Endocrine toxicity-Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	788294			
Domain	Metric	Rating	Comments	
Domain 5: Outcome Assessment				
Metric 16:	Adequacy of Test Conditions	High	Fish were housed in 3.8L tanks in dechlorinated water and fed 3 times daily. Prior to treatment they were held at 8L:16hD photoperiod at 18-20C, which was winter conditions to lead to sexual maturity. They were switched to 16L:8D with higher light intensity to bring the fish to full sexual maturity.	
Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—changes in plasma concentrations of 11 ketotestosterone and testosterone, as well as spiggin production. StAR and 3B-HSD gene expression assessment was also addressed.	
Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups.	
Domain 6: Confounding / Variable Control				
Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. It was not reported if the organisms were acclimated to test conditions.	
Metric 20:	Outcomes Unrelated to Exposure	High	Study authors reported a fish for one treatment exhibited no nesting behavior and was removed from the data because it was considered an outlier. They also discussed any potential contamination by other phthalates in the test media.	
Domain 7: Data Presentation and Analysis				
Metric 21:	Statistical Methods	High	Statistical analyses were described in the "Statistical Analysis" section and were described in detail.	
Metric 22:	Reporting of Data	Medium	Data for the exposure related finding and the control response were reported in Figures 2 and 3. Gene expression data was reported in the text only.	
Metric 23:	Explanation of Unexpected Outcomes	Medium	25th and 75th percentiles were reported surrounding the mean in all figures.	
Additional Comments:	This portion of the evaluation was on the effect of DBP on spiggen prodcutiion, ketotestosterone and testosterone plasma levels, and steroidogenic gene expression. Mechanistic outcomes were selected under biomarkers, reproduction, and endocrine system.			

Overall Quality Determination**Medium**

Study Citation:	Aoki, K. A., Harris, C. A., Katsiadaki, I., Sumpter, J. P. (2011). Evidence suggesting that di-n-butyl phthalate has antiandrogenic effects in fish. Environmental Toxicology and Chemistry 30(6):1338-1345.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Gasterosteus aculeatus</i> ; males; Adult			
Health Outcome:	Behavioral			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	788294			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The test substance was identified by name only.
	Metric 2:	Test Substance Source	High	DBP was sourced from Fisher Scientific and was analytically verified via GC/MS.
	Metric 3:	Test Substance Purity	Low	The purity of the DBP was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	The study authors reported using a negative control with the solvent used in testing.
	Metric 5:	Negative Control Response	High	The solvent control response was reported in Table 4 and was adequate for the outcome of interest.
	Metric 6:	Randomized Allocation	Medium	Study authors reported the fish were allocated randomly into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	Little detail was provided on how the stock solutions were prepared. The system was flow through. Concentrations of test substance were measured during the study every 3-4 days. Exposure was conducted in 3.8L tanks (presumably made of glass, as is standard for aquaria).
	Metric 8:	Consistency of Exposure Administration	Medium	There were eight fish per treatment group. A flow through system was used for all tanks. Water flow was the same for all tanks. The concentrations of DBP detected were not close to the nominal concentrations, creating some concern about consistency.
	Metric 9:	Measurement of Test Substance Concentration	High	Test concentrations were measured every 3-4 days. The reported concentrations were not close to the nominal concentrations, but study authors reported the endpoints were based on the measured concentrations.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration was reported to be for 22 days. This was adequate time to observe nesting behavior in all the exposure concentrations.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	There were only two exposure groups plus a control. This was adequate to observe a response, but more exposure levels may have yielded more data. Spacing was adequate.
	Metric 12:	Testing at or Below Solubility Limit	High	Study authors reported using methanol as a vehicle solvent.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the fish was reported to be ponds near Hook, Hampshire, United Kingdom. This creates significant concern about the health of the organisms prior to testing.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the fish were acclimated to test conditions prior to the start of the study. The reported 10-day "acclimation period" was conducted during the test, while exposure to DBP was occurring. Therefore it cannot be considered a true acclimation.
	Metric 15:	Number of Organisms and Replicates per Group	Low	Eight fish were exposed per treatment (including control) for a total of 24 fish, exposed in individual containers.

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Study Citation:	Aoki, K. A., Harris, C. A., Katsiadaki, I., Sumpter, J. P. (2011). Evidence suggesting that di-n-butyl phthalate has antiandrogenic effects in fish. Environmental Toxicology and Chemistry 30(6):1338-1345.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Gasterosteus aculeatus</i> ; males; Adult
Health Outcome:	Behavioral
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	788294

Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Fish were housed in 3.8L tanks in dechlorinated water and fed three times a day. Prior to treatment they were held at 8L:16hD photoperiod at 18-20C, which was winter conditions to lead to sexual maturity. They were switched to 16L:8D with higher light intensity to bring the fish to full sexual maturity.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—nesting behavior.
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. Males were observed for nesting behavior by photographs of nest building progress every two days. Threads and gravel for nesting material were added to all exposure groups on day 10.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. It was not reported if the organisms were acclimated to test conditions.
	Metric 20: Outcomes Unrelated to Exposure	High	Study authors reported a fish for one treatment exhibited no nesting behavior and was removed from the data because it was considered an outlier. They also discussed any potential contamination by other phthalates in the test media.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical analyses were described in the "Statistical Analysis" section and were described in detail.
	Metric 22: Reporting of Data	High	Data for the exposure related finding and the control response were reported in Figure 4.
	Metric 23: Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability.

Additional Comments: This portion of the evaluation was on the nesting behavior of *G. aculeatus* during exposure to DBP. The behavior outcome was chosen for this portion.

Overall Quality Determination

Medium

Study Citation:	Adams, W. J., Biddinger, G. R., Robillard, K. A., Gorsuch, J. W. (1995). A summary of the acute toxicity of 14 phthalate esters to representative aquatic organisms. Environmental Toxicology and Chemistry 14(9):1569-1574.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Lepomis macrochirus</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1321996; Linked HERO ID(s): 1321996, 1316224			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The test substance was reported without a CASRN.	
	Metric 2: Test Substance Source	Low	The source was reported as provided by a manufacturer from commercially available batches. The manufacture name and batch number were not provided. No analytical data was reported.	
	Metric 3: Test Substance Purity	High	The test substance was at least 95% pure.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	A negative control was reported.	
	Metric 5: Negative Control Response	High	The negative control response was acceptable.	
	Metric 6: Randomized Allocation	Low	The allocation method was not reported.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The experimental system was well described. However, headspace or measures to prevent volatilization were not reported.	
	Metric 8: Consistency of Exposure Administration	High	Exposure administration was consistent across groups.	
	Metric 9: Measurement of Test Substance Concentration	Medium	Sample extracts were analyzed by gas chromatography at the start and end of the test. In static studies, final test concentrations frequently were 50% of the initial concentrations. Loss of the phthalate esters was thought to be principally due to adsorption to the test vessels.	
	Metric 10: Exposure Duration and Frequency	High	Duration and frequency of exposure were appropriate for this test.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	Exposure levels were appropriate. A range finding test was performed.	
	Metric 12: Testing at or Below Solubility Limit	High	The test was performed at or below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source was not reported.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	An appropriate acclimation for this test was reported.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	Each test was performed using duplicate test concentrations with 10 organisms per test vessel.	
Domain 5: Outcome Assessment				
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Study Citation:	Adams, W. J., Biddinger, G. R., Robillard, K. A., Gorsuch, J. W. (1995). A summary of the acute toxicity of 14 phthalate esters to representative aquatic organisms. Environmental Toxicology and Chemistry 14(9):1569-1574.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Lepomis macrochirus</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1321996; Linked HERO ID(s): 1321996, 1316224			
Domain	Metric	Rating	Comments	
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions were appropriate for the test.	
	Metric 17: Outcome Assessment Methodology	High	The intended outcomes were reported.	
	Metric 18: Consistency of Outcome Assessment	High	Outcome assessment was consistent across groups.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	Environmental conditions were consistent across groups.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There were no reported differences between groups.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical methods were performed and described.	
	Metric 22: Reporting of Data	Medium	Only treatment endpoints were reported.	
	Metric 23: Explanation of Unexpected Outcomes	High	No unexpected outcomes were reported.	
Additional Comments:	None			
Overall Quality Determination		High		

Study Citation:	Bionomics,, EG&G (1983). Exhibit III: Acute toxicity of thirteen phthalate esters to bluegill (<i>Lepomis macrochirus</i>).			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Lepomis macrochirus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1316201			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The test substance- Dibutyl phthalate (DBP) was identified by chemical name and CASRN (84-74-2).	
	Metric 2: Test Substance Source	Low	The source was not reported and the test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Low	Purity and/or grade of test substance were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was adequate.	
	Metric 6: Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations and/or minimize loss of test substance before and during the exposure. Measured concentrations deviated from reported nominal concentrations.	
	Metric 8: Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	Medium	Exposure concentrations were measured at the initiation and termination of the experiment. Measured concentrations deviated from nominal concentrations.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type- 96 hour acute toxicity test.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were justified for a dose response by study authors and adequate to address the purpose of the study. For Dibutyl phthalate (DBP), a preliminary test was conducted, which indicated that it was not toxic below the water solubility limit. A corroborative test was then conducted exposing the bluegill to a single replicated concentration of Dibutyl phthalate (DBP) representing its limit of water solubility.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were at the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions and all pretreatment conditions were the same for control and exposed organisms.	
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Study Citation:	Bionomics,, EG&G (1983). Exhibit III: Acute toxicity of thirteen phthalate esters to bluegill (<i>Lepomis macrochirus</i>).			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Lepomis macrochirus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1316201			
Domain	Metric	Rating	Comments	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were ten bluegill in each test jar, and they were tested in duplicates.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Organism housing and environmental conditions were conducive to maintenance of health. The biomass loading was appropriate.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome(s) of interest.	
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study on outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	N/A	There was no mortality at the concentration tested. Therefore, statistical analysis was not conducted.	
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group. Negative findings were reported quantitatively (Table 4).	
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.	
Additional Comments:	Non-definitive LC 50 values were reported based on a corroborative test (following negative findings from a preliminary test at concentrations below the limit of water solubility) conducted by exposing bluegills to a single replicated concentration of Dibutyl phthalate (DBP) representing it's limit of water solubility.			
Overall Quality Determination		High		

Study Citation:	Buccafusco, R. J., Ells, S. J., Leblanc, G. A. (1981). Acute toxicity of priority pollutants to bluegill (<i>Lepomis macrochirus</i>). Bulletin of Environmental Contamination and Toxicology 26(4):446-452.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Lepomis macrochirus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	18064; Linked HERO ID(s): 7508, 18050, 18064, 18110, 628983			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The source was not reported.	
	Metric 3: Test Substance Purity	Medium	Purity of the test substance was reported as greater than 80%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group.	
	Metric 5: Negative Control Response	Low	The biological response of the negative control groups was not reported.	
	Metric 6: Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations. They also noted that "most of the LC50 values reported... do not reflect [t]he actual concentrations of the chemical which were in solution in the diluent", because "the acute toxicity of most of the chemicals tested was at concentrations above their water solubility".	
	Metric 8: Consistency of Exposure Administration	Medium	Only general methods of exposure administration were reported so assessment was difficult to determine.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured. They also noted that "most of the LC50 values reported... do not reflect [t]he actual concentrations of the chemical which were in solution in the diluent", because "the acute toxicity of most of the chemicals tested was at concentrations above their water solubility".	
	Metric 10: Exposure Duration and Frequency	High	Standard test durations were used (24h and 96h).	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Low	No information is provided on the number of exposure groups and spacing of exposure levels, but cited methods suggest using a minimum of five treatment levels.	
	Metric 12: Testing at or Below Solubility Limit	Medium	The authors reported this as "precipitate." However, the highest reported LC50 value is 2.1 mg/L, compared to the solubility reported in the DBP Final Scope of 11.2 mg/L at 25C.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Medium	There are minor uncertainties about the source and characteristics of test organisms because the authors use a generalized description for all reported tests.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms for the 48 hours prior to testing.	
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Study Citation:	Buccafusco, R. J., Ells, S. J., Leblanc, G. A. (1981). Acute toxicity of priority pollutants to bluegill (<i>Lepomis macrochirus</i>). Bulletin of Environmental Contamination and Toxicology 26(4):446-452.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Lepomis macrochirus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	18064; Linked HERO ID(s): 7508, 18050, 18064, 18110, 628983			
Domain	Metric	Rating	Comments	
	Metric 15: Number of Organisms and Replicates per Group	Low	Ten fish were used but no replicates were reported.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health.	
	Metric 17: Outcome Assessment Methodology	Low	Details regarding the execution of the study protocol for outcome assessment were not reported.	
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical methods were adequately described (method of moving average angles or Wilcoxon log probit).	
	Metric 22: Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group, but results were described in the tables.	
	Metric 23: Explanation of Unexpected Outcomes	Medium	Occurrence of unexpected outcomes was not addressed.	
Additional Comments:	None			
Overall Quality Determination		Medium		

Study Citation:	Aktiengesellschaft,, BASF (1989). Report on the study of the acute toxicity of dibutylphthalat on the Golden Orfe (<i>Leuciscus idus</i> L., golden variety).			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Leuciscus idus</i> L.; Golden variety; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Behavioral			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	10817969			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The DBP was identified by name only.
	Metric 2:	Test Substance Source	Low	The source of the DBP was not reported, though BASF Aktiengesellschaft is the reporting laboratory.
	Metric 3:	Test Substance Purity	High	
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of a solvent control only that contained 1mL/L of acetone. A dilution water only control was not reported.
	Metric 5:	Negative Control Response	High	The negative control response was reported in the table on page 7, where symptoms were reported, of the report and was appropriate for the outcome of interest.
	Metric 6:	Randomized Allocation	Low	It was not reported if the fish were randomly allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The test substance was prepared by dissolving it in acetone at a rate of 1.0g/mL, and this was added to the test water. Detailed methods were not provided. The system was reported to be flow-through system, but flow rate was not reported. 10-L glass aquaria. Test aquaria were reported to be 30 x 22 x 24cm, and fish were loaded at a rate of 3.4g of fish per liter of water. Flow rates and preparation of the test concentrations were not described in detail. All exposures were conducted at 19-20C with a 16L:8D photoperiod.
	Metric 8:	Consistency of Exposure Administration	Low	
	Metric 9:	Measurement of Test Substance Concentration	Low	Study authors did not report if the test concentrations were measured at any point in the study.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration was reported to be 96h, which is typical of a fish acute toxicity test.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	There were 6 exposure levels. Only the two highest concentrations were reported to have changes in behavior observed. This indicates that a different spacing may have yielded more data, especially between the 4.64mg/L and the 10mg/L exposure levels.
	Metric 12:	Testing at or Below Solubility Limit	High	All of the test concentrations were below the water solubility limit of the DBP. A vehicle solvent was also used, and the solvent control results were appropriate.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The fish were reported to be from Fischzucht Paul Eggers D-2354 Hohenwestedt, FRG. The age was not reported, but they were reported to be 7.3cm long and weighed 3.4g.

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Study Citation:	Aktiengesellschaft,, BASF (1989). Report on the study of the acute toxicity of dibutylphthalat on the Golden Orfe (<i>Leuciscus idus</i> L., golden variety).			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Leuciscus idus</i> L.; Golden variety; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Behavioral			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	10817969			
Domain	Metric	Rating	Comments	
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The fish were held for 4 months prior to testing and were acclimated to test conditions for 3 days.
	Metric 15:	Number of Organisms and Replicates per Group	Low	It was reported there were 10 fish per test concentration. The number of replicates was not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	The fish were kept at a loading rate of 3.4g of fish per liter of water. They were fed ad libitum until one day before the start of the test. They had a 16L:8D photoperiod. The water used was reconstituted fresh water, and the water quality characteristics were reported.
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported. It was not reported how the fish were assessed for behavioral changes.
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol were not reported for the behavioral portion of the study.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment. The organisms were acclimated to test conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Uninformative	Study authors did not report conducting statistical analysis on any of the behavioral changes that were noted. Behavioral outcomes were not quantified in any manner; they were simply noted in the table on page 7 of the report.
	Metric 22:	Reporting of Data	Medium	Data was reported for every exposure level in the table on page 7. Note that the behavioral outcomes were not quantified, they were simply noted in this table.
	Metric 23:	Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability.
Additional Comments:	This evaluation was on the acute toxicity of DBP on golden orfe. Study authors reported on behavioral changes in the fish in the table on page 7 of the report. These behavioral changes were simply noted, not quantified. This meant statistical analysis was not conducted for this outcome, thus the unacceptable rating.			

Overall Quality Determination**Uninformative**

Study Citation:	Aktiengesellschaft,, BASF (1989). Report on the study of the acute toxicity of dibutylphthalat on the Golden Orfe (<i>Leuciscus idus</i> L., golden variety).			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Leuciscus idus</i> L.; Golden variety; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	10817969			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The DBP was identified by name only.
	Metric 2:	Test Substance Source	Low	The source of the DBP was not reported, though BASF Aktiengesellschaft is the reporting laboratory.
	Metric 3:	Test Substance Purity	High	The purity was reported to be greater than 99.0%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of a solvent control that contained 1mL/L of acetone. A dilution water only control was not reported.
	Metric 5:	Negative Control Response	High	The negative control response was reported in the table on page 7 of the report and was appropriate for the outcome of interest.
	Metric 6:	Randomized Allocation	Low	It was not reported if the fish were randomly allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The test substance was prepared by dissolving it in acetone at a rate of 1.0g/mL, and this was added to the test water. Detailed methods were not provided. The system was reported to be a flow-through system, but flow rate was not reported. The test vessels were 10-L glass aquaria.
	Metric 8:	Consistency of Exposure Administration	Low	Test aquaria were reported to be 30 x 22 x 24cm, and fish were loaded at a rate of 3.4g of fish per liter of water. Flow rates and preparation of the test concentrations were not described in detail. All exposures were conducted at 19-20C with a 16L:8D photoperiod.
	Metric 9:	Measurement of Test Substance Concentration	Low	Study authors did not report if the test concentrations were measured at any point in the study.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration was reported to be 96h, which is typical of a fish acute toxicity test.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	There were six exposure levels. However, only the highest concentration had mortalities. This indicates that a different spacing may have yielded more data, especially between the 4.64mg/L and the 10mg/L exposure levels.
	Metric 12:	Testing at or Below Solubility Limit	High	All of the test concentrations were below the water solubility limit for DBP. A vehicle solvent was also used, and the solvent control results were appropriate.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The fish were reported to be from Fischzucht Paul Eggers D-2354 Hohenwestedt, FRG. The age was not reported, but they were reported to be 7.3cm long and weighed 3.4g.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The fish were held for four months prior to testing and were acclimated to test conditions for three days.
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Study Citation:	Aktiengesellschaft,, BASF (1989). Report on the study of the acute toxicity of dibutylphthalat on the Golden Orfe (Leuciscus idus L., golden variety).			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Leuciscus idus</i> L.; Golden variety; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	10817969			
Domain	Metric	Rating	Comments	
	Metric 15: Number of Organisms and Replicates per Group	Low	It was reported there were 10 fish per test concentration. The number of replicates was not reported.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	The fish were kept at a loading rate of 3.4g of fish per liter of water. They were fed ad libitum until one day before the start of the test. They had a 16L:8D photoperiod. The water used was reconstituted fresh water, and the water quality characteristics were reported.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest–mortality. LC50 values for 1, 4, 24, 48, 72, and 96h were reported.	
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. Mortalities were assessed at 1h, 4h, 24h, 48h, 72h, and 96h.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment. The organisms were acclimated to test conditions.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to the exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Study authors reported using probit analysis to estimate LC50 values.	
	Metric 22: Reporting of Data	High	Data was reported for all exposure levels and the controls on page 7 and was adequate for the outcomes of interest. LC50 values for each observation interval were reported on pages 11-16.	
	Metric 23: Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability.	
Additional Comments:	This evaluation was on the acute toxicity of DBP on golden orfe. Study authors calculated the LC50 values for 1, 4, 24, 48, 72, and 96h. Mortality was the outcome of interest.			
Overall Quality Determination		Medium		

Study Citation:	Bhatia, H., Kumar, A., Du, J., Chapman, J., McLaughlin, M. J. (2013). Di-n-butyl phthalate causes antiestrogenic effects in female murray rainbowfish (<i>Melanotaenia fluviatilis</i>). Environmental Toxicology and Chemistry 32(10):2335-2344.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Melanotaenia fluviatilis</i> ; Adult			
Health Outcome:	Hepatic/Liver			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1639196			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	High	The test substance identity was verified by GC-MS.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group, and a positive vehicle control (0.001% methanol).
	Metric 5:	Negative Control Response	Medium	No mortality in any treatments reported, some growth results were only in text as not significant.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail. Exposure was conducted in glass aquaria.
	Metric 8:	Consistency of Exposure Administration	Medium	Details of exposure administration were reported and exposures were administered consistently across study groups (spiked daily) but how stock solutions were mixed was not reported
	Metric 9:	Measurement of Test Substance Concentration	Medium	Exposure concentrations were measured by GC-MS but were lower than nominal concentrations.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response (125, 250, 500, and 1000 ug/L DBP), 2 replicates containing 4 fish each.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions and all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Low	Only 4 fish in duplicate were used.
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Study Citation:	Bhatia, H., Kumar, A., Du, J., Chapman, J., McLaughlin, M. J. (2013). Di-n-butyl phthalate causes antiestrogenic effects in female murray rainbowfish (<i>Melanotaenia fluviatilis</i>). Environmental Toxicology and Chemistry 32(10):2335-2344.		
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Melanotaenia fluviatilis</i> ; Adult		
Health Outcome:	Hepatic/Liver		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	1639196		
Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcomes of interest.
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for growth measurements were limited.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described (one-way ANOVA).
	Metric 22: Reporting of Data	Medium	Some results were in text only, others were in table 2.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments:	This form was added to account for the hepatic outcome of the HSI.		

Overall Quality Determination**High**

Study Citation:	Bhatia, H., Kumar, A., Du, J., Chapman, J., McLaughlin, M. J. (2013). Di-n-butyl phthalate causes antiestrogenic effects in female murray rainbowfish (<i>Melanotaenia fluviatilis</i>). Environmental Toxicology and Chemistry 32(10):2335-2344.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Melanotaenia fluviatilis</i> ; Adult			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1639196			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The chemical was identified by name and CAS#.
	Metric 2:	Test Substance Source	High	The test substance identity was verified by GC-MS.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group, and a positive vehicle control (0.001% methanol).
	Metric 5:	Negative Control Response	Medium	No mortality in any treatments was reported. Some growth results were mentioned in the text as not significant.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail. Exposure was conducted in glass aquaria.
	Metric 8:	Consistency of Exposure Administration	Medium	Details of exposure administration were reported and exposures were administered consistently across study groups (spiked daily), but how stock solutions were mixed was not reported.
	Metric 9:	Measurement of Test Substance Concentration	Medium	Exposure concentrations were measured by GC-MS but were lower than nominal concentrations.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response (125, 250, 500, and 1000 ug/L DBP). There were two replicates containing four fish each.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions and all pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Low	Only four fish in each duplicate were used.
Domain 5: Outcome Assessment				
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Study Citation:	Bhatia, H., Kumar, A., Du, J., Chapman, J., McLaughlin, M. J. (2013). Di-n-butyl phthalate causes antiestrogenic effects in female murray rainbowfish (<i>Melanotaenia fluviatilis</i>). Environmental Toxicology and Chemistry 32(10):2335-2344.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Melanotaenia fluviatilis</i> ; Adult
Health Outcome:	Reproductive/Teratogenic
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	1639196

Domain	Metric	Rating	Comments
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcomes of interest.
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for growth measurements were limited.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described (one-way ANOVA).
	Metric 22: Reporting of Data	Medium	Some results were in text only, while others were in table 2.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This form was added to account for the reproductive outcomes obtained from histology and for the GSI.

Overall Quality Determination

High

Study Citation:	Bhatia, H., Kumar, A., Du, J., Chapman, J., McLaughlin, M. J. (2013). Di-n-butyl phthalate causes antiestrogenic effects in female murray rainbowfish (Melanotaenia fluviatilis). Environmental Toxicology and Chemistry 32(10):2335-2344.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Melanotaenia fluviatilis</i> ; Adult			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1639196			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	High	The test substance identity was verified by GC-MS.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group, and a positive vehicle control (0.001% methanol).
	Metric 5:	Negative Control Response	Medium	No mortality in any treatments reported, some growth results were only in text as not significant.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail. Exposure was conducted in glass aquaria.
	Metric 8:	Consistency of Exposure Administration	Medium	Details of exposure administration were reported and exposures were administered consistently across study groups (spiked daily) but how stock solutions were mixed was not reported
	Metric 9:	Measurement of Test Substance Concentration	Medium	Exposure concentrations were measured by GC-MS but were lower than nominal concentrations.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response (125, 250, 500, and 1000 ug/L DBP), 2 replicates containing 4 fish each.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions and all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Low	Only 4 fish in duplicate were used.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health.
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Study Citation:	Bhatia, H., Kumar, A., Du, J., Chapman, J., McLaughlin, M. J. (2013). Di-n-butyl phthalate causes antiestrogenic effects in female murray rainbowfish (<i>Melanotaenia fluviatilis</i>). Environmental Toxicology and Chemistry 32(10):2335-2344.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Melanotaenia fluviatilis</i> ; Adult			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1639196			
Domain	Metric	Rating	Comments	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcomes of interest.	
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for growth measurements were limited.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical methods were adequately described (one-way ANOVA).	
	Metric 22: Reporting of Data	Medium	Some results were in text only, others were in table 2.	
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.	
Additional Comments:	None			

Overall Quality Determination**High**

Study Citation:	Bhatia, H., Kumar, A., Du, J., Chapman, J., McLaughlin, M. J. (2013). Di-n-butyl phthalate causes antiestrogenic effects in female murray rainbowfish (<i>Melanotaenia fluviatilis</i>). Environmental Toxicology and Chemistry 32(10):2335-2344.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Melanotaenia fluviatilis</i> ; Adult			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Endocrine toxicity			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1639196			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	High	The test substance identity was verified by GC-MS.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	No mortality in any treatments reported, mechanistic results were reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	Medium	Details of exposure administration were reported and exposures were administered consistently across study groups (spiked daily) but how chemical was mixed was not reported
	Metric 9:	Measurement of Test Substance Concentration	Medium	Exposure concentrations were measured but were lower than nominal concentrations
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions and all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Low	Only 4 fish in duplicate were used
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcomes of interest
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Study Citation:	Bhatia, H., Kumar, A., Du, J., Chapman, J., McLaughlin, M. J. (2013). Di-n-butyl phthalate causes antiestrogenic effects in female murray rainbowfish (<i>Melanotaenia fluviatilis</i>). Environmental Toxicology and Chemistry 32(10):2335-2344.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Melanotaenia fluviatilis</i> ; Adult			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Endocrine toxicity			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1639196			
Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	High	Details regarding the execution of the study protocol for mechanistic measurements were adequate
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes
Additional Comments: Biomarkers were added to the mechanistic outcome to account for vitellogenin levels.				

Overall Quality Determination**High**

Study Citation:	Bhatia, H., Kumar, A., Du, J., Chapman, J., McLaughlin, M. J. (2013). Di-n-butyl phthalate causes antiestrogenic effects in female murray rainbowfish (<i>Melanotaenia fluviatilis</i>). Environmental Toxicology and Chemistry 32(10):2335-2344.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Melanotaenia fluviatilis</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1639196			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	Chemical was identified by name and CAS#	
	Metric 2: Test Substance Source	High	The test substance identity was verified by GC-MS.	
	Metric 3: Test Substance Purity	Low	Purity and/or grade of test substance were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group	
	Metric 5: Negative Control Response	High	No mortality in any treatments were reported	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail	
	Metric 8: Consistency of Exposure Administration	Medium	Details of exposure administration were reported and exposures were administered consistently across study groups (spiked daily) but how chemical was mixed was not reported	
	Metric 9: Measurement of Test Substance Concentration	Medium	Exposure concentrations were measured but were lower than nominal concentrations	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions and all pretreatment conditions were the same for control and exposed organisms	
	Metric 15: Number of Organisms and Replicates per Group	Low	Only 4 fish in duplicate were used	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest	
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Study Citation:	Bhatia, H., Kumar, A., Du, J., Chapman, J., McLaughlin, M. J. (2013). Di-n-butyl phthalate causes antiestrogenic effects in female murray rainbowfish (<i>Melanotaenia fluviatilis</i>). Environmental Toxicology and Chemistry 32(10):2335-2344.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Melanotaenia fluviatilis</i> ; Adult
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	1639196

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for mortality were limited
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described
	Metric 22: Reporting of Data	Low	Text reported no mortalities
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: None

Overall Quality Determination**High**

Study Citation:	Bhatia, H., Kumar, A., Ogino, Y., Gregg, A., Chapman, J., McLaughlin, M. J., Iguchi, T. (2014). Di-n-butyl phthalate causes estrogenic effects in adult male Murray rainbowfish (<i>Melanotaenia fluviatilis</i>). <i>Aquatic Toxicology</i> 149(Elsevier):103-115.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Melanotaenia fluviatilis</i> ; Males; Adult			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2509291			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The DBP was identified by CASRN.	
	Metric 2: Test Substance Source	Low	The DBP was reported to be from Sigma Aldrich, but it did not appear to be analytically verified.	
	Metric 3: Test Substance Purity	Low	The purity/grade of the DBP was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of a concurrent negative control as well as a solvent control.	
	Metric 5: Negative Control Response	High	The negative control responses were reported in Figures 1-4 and were adequate for the outcomes of interest.	
	Metric 6: Randomized Allocation	Low	It was not reported how the fish were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The system was reported to be semi-static with daily renewals, but the preparation of the test media was not reported.	
	Metric 8: Consistency of Exposure Administration	Medium	All exposures occurred in 35 x 18 x 24cm aquaria containing 5L of test solution with 4 fish per tank. All exposures were conducted at 21 C with a 16L:8D photoperiod. But measured concentrations did not appear to be close to nominal concentrations in some instances.	
	Metric 9: Measurement of Test Substance Concentration	High	The test substance was measured using GC-MS.	
	Metric 10: Exposure Duration and Frequency	High	The duration of the exposure was reported to be 7 days, and this was adequate for a dose response.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	There were 4 exposure levels plus a negative control and a solvent control. The spacing was appropriate to observe a dose response.	
	Metric 12: Testing at or Below Solubility Limit	High	Study authors reported using a vehicle solvent, and the solvent control response was adequate.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The fish were reported to be from Aquarium Industries in Victoria, Australia. Fish were reported to be adult males.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	The fish were acclimated for 14d prior to the start of the test.	

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Study Citation:	Bhatia, H., Kumar, A., Ogino, Y., Gregg, A., Chapman, J., McLaughlin, M. J., Iguchi, T. (2014). Di-n-butyl phthalate causes estrogenic effects in adult male Murray rainbowfish (<i>Melanotaenia fluviatilis</i>). <i>Aquatic Toxicology</i> 149(Elsevier):103-115.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Melanotaenia fluviatilis</i> ; Males; Adult
Health Outcome:	Reproductive/Teratogenic
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	2509291

Domain	Metric	Rating	Comments
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 4 fish per test chamber and 2 aquaria per treatment.
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Fish were kept at 21C in artificial river water with a 16L:8D photoperiod. They were fed frozen brine shrimp at a rate of 4% (w/w) body weight.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—histological changes in the testes.
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. Tissues were fixed after the 7 day exposure period. Assessment was discussed in detail in the paper.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were reported in the "Data Analysis" section.
	Metric 22: Reporting of Data	High	Data for the exposure response and the control response were reported in Figures 1-4 and were adequate.
	Metric 23: Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in Figures 3 and 4.
Additional Comments: This evaluation was on the histopathological effects of DBP on Murray rainbowfish testes. Reproduction was selected as the outcome of interest.			

Overall Quality Determination**High**

Study Citation:	Bhatia, H., Kumar, A., Ogino, Y., Gregg, A., Chapman, J., McLaughlin, M. J., Iguchi, T. (2014). Di-n-butyl phthalate causes estrogenic effects in adult male Murray rainbowfish (<i>Melanotaenia fluviatilis</i>). <i>Aquatic Toxicology</i> 149(Elsevier):103-115.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Melanotaenia fluviatilis</i> ; Males; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2509291			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The DBP was identified by CASRN.	
	Metric 2: Test Substance Source	Low	The DBP was reported to be from Sigma Aldrich, but it did not appear to be analytically verified.	
	Metric 3: Test Substance Purity	Low	The purity/grade of the DBP was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of a concurrent negative control as well as a solvent control.	
	Metric 5: Negative Control Response	High	The negative control response was reported in section 3.3.	
	Metric 6: Randomized Allocation	Low	It was not reported how the fish were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The system was reported to be semi-static with daily renewals, but the preparation of the test media was not reported.	
	Metric 8: Consistency of Exposure Administration	Medium	All exposures occurred in 35 x 18 x 24cm aquaria containing 5L of test solution with 4 fish per tank. All exposures were conducted at 21 C with a 16L:8D photoperiod. But measured concentrations did not appear to be close to nominal concentrations in some instances.	
	Metric 9: Measurement of Test Substance Concentration	High	The test substance was measured using GC-MS.	
	Metric 10: Exposure Duration and Frequency	High	The duration of the exposure was reported to be 7 days, and this was adequate for a dose response.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	There were 4 exposure levels plus a negative control and a solvent control. The spacing was appropriate to observe a dose response.	
	Metric 12: Testing at or Below Solubility Limit	High	Study authors reported using a vehicle solvent, and the solvent control response was adequate.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The fish were reported to be from Aquarium Industries in Victoria, Australia. Fish were reported to be adult males.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	The fish were acclimated for 14d prior to the start of the test.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 4 fish per test chamber and 2 aquaria per treatment.	
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Study Citation:	Bhatia, H., Kumar, A., Ogino, Y., Gregg, A., Chapman, J., McLaughlin, M. J., Iguchi, T. (2014). Di-n-butyl phthalate causes estrogenic effects in adult male Murray rainbowfish (<i>Melanotaenia fluviatilis</i>). <i>Aquatic Toxicology</i> 149(Elsevier):103-115.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Melanotaenia fluviatilis</i> ; Males; Adult
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	2509291

Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Fish were kept at 21C in artificial river water with a 16L:8D photoperiod. They were fed frozen brine shrimp at a rate of 4% (w/w) body weight.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—mortality.
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were reported in the "Data Analysis" section.
	Metric 22: Reporting of Data	High	Data for the exposure response and the control response were reported in Section 3.3.
	Metric 23: Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in Figures 3 and 4.

Additional Comments: Section 3.3 reported no mortality among the fish. This form was created for that outcome.

Overall Quality Determination

High

Study Citation:	Bhatia, H., Kumar, A., Ogino, Y., Gregg, A., Chapman, J., McLaughlin, M. J., Iguchi, T. (2014). Di-n-butyl phthalate causes estrogenic effects in adult male Murray rainbowfish (<i>Melanotaenia fluviatilis</i>). <i>Aquatic Toxicology</i> 149(Elsevier):103-115.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Melanotaenia fluviatilis</i> ; Males; Adult			
Health Outcome:	Hepatic/Liver			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2509291			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The DBP was identified by CASRN.	
	Metric 2: Test Substance Source	Low	The DBP was reported to be from Sigma Aldrich, but it did not appear to be analytically verified.	
	Metric 3: Test Substance Purity	Low	The purity/grade of the DBP was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of a concurrent negative control as well as a solvent control.	
	Metric 5: Negative Control Response	High	The negative control responses were reported in Table 3 and were adequate for the outcome of interest.	
	Metric 6: Randomized Allocation	Low	It was not reported how the fish were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The system was reported to be semi-static with daily renewals, but the preparation of the test media was not reported.	
	Metric 8: Consistency of Exposure Administration	Medium	All exposures occurred in 35 x 18 x 24cm aquaria containing 5L of test solution with four fish per tank. All exposures were conducted at 21 C with a 16L:8D photoperiod. But measured concentrations did not appear to be close to nominal concentrations in some instances.	
	Metric 9: Measurement of Test Substance Concentration	High	The test substance was measured using GC-MS.	
	Metric 10: Exposure Duration and Frequency	High	The duration of the exposure was reported to be seven days, and this was adequate for a dose response.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	There were four exposure levels plus a negative control and a solvent control. The spacing was appropriate to observe a dose response.	
	Metric 12: Testing at or Below Solubility Limit	High	Study authors reported using a vehicle solvent, and the solvent control response was adequate.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The fish were reported to be from Aquarium Industries in Victoria, Australia. Fish were reported to be adult males.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	The fish were acclimated for 14d prior to the start of the test.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were four fish per test chamber and two aquaria per treatment.	
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Study Citation:	Bhatia, H., Kumar, A., Ogino, Y., Gregg, A., Chapman, J., McLaughlin, M. J., Iguchi, T. (2014). Di-n-butyl phthalate causes estrogenic effects in adult male Murray rainbowfish (<i>Melanotaenia fluviatilis</i>). <i>Aquatic Toxicology</i> 149(Elsevier):103-115.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Melanotaenia fluviatilis</i> ; Males; Adult
Health Outcome:	Hepatic/Liver
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	2509291

Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Fish were kept at 21C in artificial river water with a 16L:8D photoperiod. They were fed frozen brine shrimp at a rate of 4% (w/w) body weight.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—HSI.
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. Tissues were fixed after the seven-day exposure period. Assessment was discussed in detail in the paper.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were reported in the "Data Analysis" section.
	Metric 22: Reporting of Data	High	Data for the exposure response and the control response were reported in Table 3.
	Metric 23: Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in Figures 3 and 4.

Additional Comments: This evaluation was for the HSI reported in Table 3. The outcome selected was the hepatic outcome.

Overall Quality Determination

High

Study Citation:	Bhatia, H., Kumar, A., Ogino, Y., Gregg, A., Chapman, J., McLaughlin, M. J., Iguchi, T. (2014). Di-n-butyl phthalate causes estrogenic effects in adult male Murray rainbowfish (<i>Melanotaenia fluviatilis</i>). <i>Aquatic Toxicology</i> 149(Elsevier):103-115.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Melanotaenia fluviatilis</i> ; Males; Adult			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2509291			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The DBP was identified by CASRN.	
	Metric 2: Test Substance Source	Low	The DBP was reported to be from Sigma Aldrich, but it did not appear to be analytically verified.	
	Metric 3: Test Substance Purity	Low	The purity/grade of the DBP was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of a concurrent negative control as well as a solvent control.	
	Metric 5: Negative Control Response	High	The negative control responses were reported in Figures 5-7 and was adequate for the outcome of interest.	
	Metric 6: Randomized Allocation	Low	It was not reported how the fish were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The system was reported to be semi-static with daily renewals, but the preparation of the test media was not reported.	
	Metric 8: Consistency of Exposure Administration	Medium	All exposures occurred in 35 x 18 x 24cm aquaria containing 5L of test solution with 4 fish per tank. All exposures were conducted at 21 C with a 16L:8D photoperiod. But measured concentrations did not appear to be close to nominal concentrations in some instances.	
	Metric 9: Measurement of Test Substance Concentration	High	The test substance was measured using GC-MS.	
	Metric 10: Exposure Duration and Frequency	High	The duration of the exposure was reported to be 7 days, and this was adequate for a dose response.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	There were 4 exposure levels plus a negative control and a solvent control. The spacing was appropriate to observe a dose response.	
	Metric 12: Testing at or Below Solubility Limit	High	Study authors reported using a vehicle solvent, and the solvent control response was adequate.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The fish were reported to be from Aquarium Industries in Victoria, Australia. Fish were reported to be adult males.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	The fish were acclimated for 14d prior to the start of the test.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 4 fish per test chamber and 2 aquaria per treatment.	
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Study Citation:	Bhatia, H., Kumar, A., Ogino, Y., Gregg, A., Chapman, J., McLaughlin, M. J., Iguchi, T. (2014). Di-n-butyl phthalate causes estrogenic effects in adult male Murray rainbowfish (<i>Melanotaenia fluviatilis</i>). <i>Aquatic Toxicology</i> 149(Elsevier):103-115.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Melanotaenia fluviatilis</i> ; Males; Adult
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	2509291

Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Fish were kept at 21C in artificial river water with a 16L:8D photoperiod. They were fed frozen brine shrimp at a rate of 4% (w/w) body weight.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—gene expression/transcription.
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were reported in the "Data Analysis" section.
	Metric 22: Reporting of Data	High	Data for the exposure response and the control response were reported in Figures 5-7 and were adequate.
	Metric 23: Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in Figures 5-7.
Additional Comments: This evaluation was on the effects of DBP on gene expression. The mechanistic biomarker outcome was selected.			

Overall Quality Determination**High**

Study Citation:	Bhatia, H., Kumar, A., Ogino, Y., Gregg, A., Chapman, J., McLaughlin, M. J., Iguchi, T. (2014). Di-n-butyl phthalate causes estrogenic effects in adult male Murray rainbowfish (<i>Melanotaenia fluviatilis</i>). <i>Aquatic Toxicology</i> 149(Elsevier):103-115.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Melanotaenia fluviatilis</i> ; Males; Adult			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2509291			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The DBP was identified by CASRN.
	Metric 2:	Test Substance Source	Low	The DBP was reported to be from Sigma Aldrich, but it did not appear to be analytically verified.
	Metric 3:	Test Substance Purity	Low	The purity/grade of the DBP was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of a concurrent negative control as well as a solvent control.
	Metric 5:	Negative Control Response	High	The negative control responses were reported in Table 3 and was adequate for the outcome of interest.
	Metric 6:	Randomized Allocation	Low	It was not reported how the fish were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The system was reported to be semi-static with daily renewals, but the preparation of the test media was not reported.
	Metric 8:	Consistency of Exposure Administration	Medium	All exposures occurred in 35 x 18 x 24cm aquaria containing 5L of test solution with 4 fish per tank. All exposures were conducted at 21 C with a 16L:8D photoperiod. But measured concentrations did not appear to be close to nominal concentrations in some instances.
	Metric 9:	Measurement of Test Substance Concentration	High	The test substance was measured using GC-MS.
	Metric 10:	Exposure Duration and Frequency	High	The duration of the exposure was reported to be 7 days, and this was adequate for a dose response.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	There were 4 exposure levels plus a negative control and a solvent control. The spacing was appropriate to observe a dose response.
	Metric 12:	Testing at or Below Solubility Limit	High	Study authors reported using a vehicle solvent, and the solvent control response was adequate.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The fish were reported to be from Aquarium Industries in Victoria, Australia. Fish were reported to be adult males.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The fish were acclimated for 14d prior to the start of the test.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	There were 4 fish per test chamber and 2 aquaria per treatment.
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Study Citation:	Bhatia, H., Kumar, A., Ogino, Y., Gregg, A., Chapman, J., McLaughlin, M. J., Iguchi, T. (2014). Di-n-butyl phthalate causes estrogenic effects in adult male Murray rainbowfish (<i>Melanotaenia fluviatilis</i>). <i>Aquatic Toxicology</i> 149(Elsevier):103-115.		
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Melanotaenia fluviatilis</i> ; Males; Adult		
Health Outcome:	Development/Growth		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	2509291		
Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
Metric 16:	Adequacy of Test Conditions	High	Fish were kept at 21C in artificial river water with a 16L:8D photoperiod. They were fed frozen brine shrimp at a rate of 4% (w/w) body weight.
Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—body weight and organ weight changes.
Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. Body weights and lengths were taken at the end of the 7 days, and organs weights were also taken at this time to calculate GSI and HSI and CF.
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	High	Statistical methods were reported in the "Data Analysis" section.
Metric 22:	Reporting of Data	High	Data for the exposure response and the control response were reported in Table 3 and were adequate.
Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in Table 3.
Additional Comments:	This evaluation was on the effects of DBP on organ weights and body weight and length of the Murray rainbow fish. Development/growth was selected as the outcome of interest.		
Overall Quality Determination		High	

Study Citation:	Bhatia, H., Kumar, A., Chapman, J. C., McLaughlin, M. J. (2015). Long-term exposures to di-n-butyl phthalate inhibit body growth and impair gonad development in juvenile Murray rainbowfish (<i>Melanotaenia fluviatilis</i>). Journal of Applied Toxicology 35(7):806-816.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Melanotaenia fluviatilis</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2816886			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The test substance was identified by CASRN.
	Metric 2:	Test Substance Source	Low	The source was reported to be Sigma Aldrich in Australia, but it was not reported if it was analytically verified.
	Metric 3:	Test Substance Purity	Low	The purity of the DBP was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	It was reported a concurrent negative "water" control was used as well as a solvent control.
	Metric 5:	Negative Control Response	Low	The response of the negative control was not reported for mortality.
	Metric 6:	Randomized Allocation	Low	It was not reported how the fish were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	Limited details were provided on the preparation of the stock solution and the test concentrations.
	Metric 8:	Consistency of Exposure Administration	Medium	Exposures were conducted in 1L for the first 30d and then in 2L for the rest of the study with four fish per glass beaker. Temperature and photoperiod remained the same for the duration of the study.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using GC-MS.
	Metric 10:	Exposure Duration and Frequency	High	Exposures were for up to 90 days with sampling at 30, 60, and 90 days. This was adequate to observe a response.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	There were only three exposure levels, which is lower than normal, but still showed a response.
	Metric 12:	Testing at or Below Solubility Limit	High	It was reported that DMSO was used as a vehicle solvent. The solvent control response was appropriate.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Organisms were from the Aquarium Industries in Victoria, Australia. Juvenile fish were used for this study.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Fish were acclimated to test conditions for two weeks prior to the start of this study.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	There were four fish per test chamber and four replicates per treatment per time interval.

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Study Citation:	Bhatia, H., Kumar, A., Chapman, J. C., McLaughlin, M. J. (2015). Long-term exposures to di-n-butyl phthalate inhibit body growth and impair gonad development in juvenile Murray rainbowfish (<i>Melanotaenia fluviatilis</i>). Journal of Applied Toxicology 35(7):806-816.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Melanotaenia fluviatilis</i> ; Juvenile
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	2816886

Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Fish were reported to be kept at a loading rate of 0.5g/L at a temperature of 23C with a 16L:8D photoperiod. They were fed artemia daily at 4% (w/w).
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—mortality.
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. The fish were sampled at each time interval and were measured and weighed at this point.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Medium	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were described in the "Statistical Analyses" section of the paper.
	Metric 22: Reporting of Data	Low	Results for mortality were reported in the text only.
	Metric 23: Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in Figure 1.
Additional Comments: This portion of the evaluation was on the effect of DBP on the mortality of <i>M. fluviatilis</i> . Mortality was selected as the outcome of interest.			

Overall Quality Determination

High

Study Citation:	Bhatia, H., Kumar, A., Chapman, J. C., McLaughlin, M. J. (2015). Long-term exposures to di-n-butyl phthalate inhibit body growth and impair gonad development in juvenile Murray rainbowfish (<i>Melanotaenia fluviatilis</i>). Journal of Applied Toxicology 35(7):806-816.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Melanotaenia fluviatilis</i> ; Juvenile
Health Outcome:	Reproductive/Teratogenic
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	2816886

Domain	Metric	Rating	Comments
Domain 1: Test Substance			
	Metric 1: Test Substance Identity	High	The test substance was identified by CASRN.
	Metric 2: Test Substance Source	Low	The source was reported to be Sigma Aldrich in Australia, but it was not reported if it was analytically verified.
	Metric 3: Test Substance Purity	Low	The purity of the DBP was not reported.
Domain 2: Test Design			
	Metric 4: Negative Controls	High	It was reported a concurrent negative "water" control was used as well as a solvent control.
	Metric 5: Negative Control Response	High	The negative control response was reported in Figures 2 and 3 and was adequate for the outcomes of interest.
	Metric 6: Randomized Allocation	Low	It was not reported how the fish were allocated into study groups.
Domain 3: Exposure Characterization			
	Metric 7: Experimental System/Test Media Preparation	Low	Limited details were provided on the preparation of the stock solution and the test concentrations.
	Metric 8: Consistency of Exposure Administration	Medium	Exposures were conducted in 1L for the first 30d and then in 2L for the rest of the study with 4 fish per glass beaker. Temperature and photoperiod remained the same for the duration of the study.
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured using GC-MS.
	Metric 10: Exposure Duration and Frequency	High	Exposures were for up to 90 days with sampling at 30, 60, and 90 days. This was adequate to observe a response.
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Medium	There were only 3 exposure levels, which is lower than normal, but still showed a response.
	Metric 12: Testing at or Below Solubility Limit	High	It was reported that DMSO was used as a vehicle solvent. The solvent control response was appropriate.
Domain 4: Test Organism			
	Metric 13: Test Organism Characteristics	High	Organisms were from the Aquarium Industries in Victoria, Australia. Juvenile fish were used for this study.
	Metric 14: Acclimatization and Pretreatment Conditions	High	Fish were acclimated to test conditions for 2 weeks prior to the start of this study.
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 4 fish per test chamber and 4 replicates per treatment per time interval.
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Study Citation:	Bhatia, H., Kumar, A., Chapman, J. C., McLaughlin, M. J. (2015). Long-term exposures to di-n-butyl phthalate inhibit body growth and impair gonad development in juvenile Murray rainbowfish (<i>Melanotaenia fluviatilis</i>). Journal of Applied Toxicology 35(7):806-816.		
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Melanotaenia fluviatilis</i> ; Juvenile		
Health Outcome:	Reproductive/Teratogenic		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	2816886		
Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
Metric 16:	Adequacy of Test Conditions	High	Fish were reported to be kept at a loading rate of 0.5g/L at a temperature of 23C with a 16L:8D photoperiod. They were fed artemia daily at 4% (w/w).
Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—histopathological changes in the gonads.
Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. The fish were sampled at each time interval and were fixed investigation of gonad development by whole body sectioning. Parameters were set for staging the gonads.
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	Medium	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	High	Statistical methods were described in the "Statistical Analyses" section of the paper.
Metric 22:	Reporting of Data	High	The control response and the exposure response were adequate and were reported in Figures 2-4.
Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.
Additional Comments:	This portion of the evaluation was on the effect of DBP on the gonads of the <i>M. fluviatilis</i> . Histopathology was used to assess the organs. Reproduction was selected as the outcome of interest.		
Overall Quality Determination		High	

Study Citation:	Bhatia, H., Kumar, A., Chapman, J. C., McLaughlin, M. J. (2015). Long-term exposures to di-n-butyl phthalate inhibit body growth and impair gonad development in juvenile Murray rainbowfish (<i>Melanotaenia fluviatilis</i>). Journal of Applied Toxicology 35(7):806-816.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Melanotaenia fluviatilis</i> ; Juvenile
Health Outcome:	Mechanistic-Endocrine toxicity
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	2816886

Domain	Metric	Rating	Comments
Domain 1: Test Substance			
	Metric 1: Test Substance Identity	High	The test substance was identified by CASRN.
	Metric 2: Test Substance Source	Low	The source was reported to be Sigma Aldrich in Australia, but it was not reported if it was analytically verified.
	Metric 3: Test Substance Purity	Low	The purity of the DBP was not reported.
Domain 2: Test Design			
	Metric 4: Negative Controls	High	It was reported a concurrent negative "water" control was used as well as a solvent control.
	Metric 5: Negative Control Response	High	The negative control response was reported in Figure 5 and was adequate for the outcomes of interest.
	Metric 6: Randomized Allocation	Low	It was not reported how the fish were allocated into study groups.
Domain 3: Exposure Characterization			
	Metric 7: Experimental System/Test Media Preparation	Low	Limited details were provided on the preparation of the stock solution and the test concentrations.
	Metric 8: Consistency of Exposure Administration	Medium	Exposures were conducted in 1L for the first 30d and then in 2L for the rest of the study with 4 fish per glass beaker. Temperature and photoperiod remained the same for the duration of the study.
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured using GC-MS.
	Metric 10: Exposure Duration and Frequency	High	Exposures were for up to 90 days with sampling at 30, 60, and 90 days. This was adequate to observe a response.
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Medium	There were only 3 exposure levels, which is lower than normal, but still showed a response.
	Metric 12: Testing at or Below Solubility Limit	High	It was reported that DMSO was used as a vehicle solvent. The solvent control response was appropriate.
Domain 4: Test Organism			
	Metric 13: Test Organism Characteristics	High	Organisms were from the Aquarium Industries in Victoria, Australia. Juvenile fish were used for this study.
	Metric 14: Acclimatization and Pretreatment Conditions	High	Fish were acclimated to test conditions for 2 weeks prior to the start of this study.
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 4 fish per test chamber and 4 replicates per treatment per time interval.
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Study Citation:	Bhatia, H., Kumar, A., Chapman, J. C., McLaughlin, M. J. (2015). Long-term exposures to di-n-butyl phthalate inhibit body growth and impair gonad development in juvenile Murray rainbowfish (<i>Melanotaenia fluviatilis</i>). Journal of Applied Toxicology 35(7):806-816.		
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Melanotaenia fluviatilis</i> ; Juvenile		
Health Outcome:	Mechanistic-Endocrine toxicity		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	2816886		
Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Fish were reported to be kept at a loading rate of 0.5g/L at a temperature of 23C with a 16L:8D photoperiod. They were fed artemia daily at 4% (w/w).
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—hormone levels.
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. The fish were sampled at each time interval and were snap frozen until enzyme analysis. EIA kits were used to assess the hormone levels.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Medium	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were described in the "Statistical Analyses" section of the paper.
	Metric 22: Reporting of Data	High	The control response and the exposure response were adequate and were reported in Figure 5.
	Metric 23: Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in Figure 5.
Additional Comments:	This portion of the evaluation was on the effect of DBP on hormone levels in <i>M. fluviatilis</i> . The mechanistic endocrine outcome was selected as the outcome of interest. Mortality was also mentioned in this study but was not quantified anywhere, so an evaluation was not completed for it.		
Overall Quality Determination	High		

Study Citation:	Bhatia, H., Kumar, A., Chapman, J. C., McLaughlin, M. J. (2015). Long-term exposures to di-n-butyl phthalate inhibit body growth and impair gonad development in juvenile Murray rainbowfish (<i>Melanotaenia fluviatilis</i>). Journal of Applied Toxicology 35(7):806-816.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Melanotaenia fluviatilis</i> ; Juvenile
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	2816886

Domain	Metric	Rating	Comments
Domain 1: Test Substance			
	Metric 1: Test Substance Identity	High	The test substance was identified by CASRN.
	Metric 2: Test Substance Source	Low	The source was reported to be Sigma Aldrich in Australia, but it was not reported if it was analytically verified.
	Metric 3: Test Substance Purity	Low	The purity of the DBP was not reported.
Domain 2: Test Design			
	Metric 4: Negative Controls	High	It was reported a concurrent negative "water" control was used as well as a solvent control.
	Metric 5: Negative Control Response	High	The negative control response was reported in Figure 1 and was adequate for the outcomes of interest.
	Metric 6: Randomized Allocation	Low	It was not reported how the fish were allocated into study groups.
Domain 3: Exposure Characterization			
	Metric 7: Experimental System/Test Media Preparation	Low	Limited details were provided on the preparation of the stock solution and the test concentrations.
	Metric 8: Consistency of Exposure Administration	Medium	Exposures were conducted in 1L for the first 30d and then in 2L for the rest of the study with 4 fish per glass beaker. Temperature and photoperiod remained the same for the duration of the study.
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured using GC-MS.
	Metric 10: Exposure Duration and Frequency	High	Exposures were for up to 90 days with sampling at 30, 60, and 90 days. This was adequate to observe a response.
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Medium	There were only 3 exposure levels, which is lower than normal, but still showed a response.
	Metric 12: Testing at or Below Solubility Limit	High	It was reported that DMSO was used as a vehicle solvent. The solvent control response was appropriate.
Domain 4: Test Organism			
	Metric 13: Test Organism Characteristics	High	Organisms were from the Aquarium Industries in Victoria, Australia. Juvenile fish were used for this study.
	Metric 14: Acclimatization and Pretreatment Conditions	High	Fish were acclimated to test conditions for 2 weeks prior to the start of this study.
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 4 fish per test chamber and 4 replicates per treatment per time interval.
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Study Citation:	Bhatia, H., Kumar, A., Chapman, J. C., McLaughlin, M. J. (2015). Long-term exposures to di-n-butyl phthalate inhibit body growth and impair gonad development in juvenile Murray rainbowfish (<i>Melanotaenia fluviatilis</i>). Journal of Applied Toxicology 35(7):806-816.		
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Melanotaenia fluviatilis</i> ; Juvenile		
Health Outcome:	Development/Growth		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	2816886		
Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Fish were reported to be kept at a loading rate of 0.5g/L at a temperature of 23C with a 16L:8D photoperiod. They were fed artemia daily at 4% (w/w).
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—the growth of the fish in terms of length and weight.
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. The fish were sampled at each time interval and were measured and weighed at this point.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Medium	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were described in the "Statistical Analyses" section of the paper.
	Metric 22: Reporting of Data	High	The control response and the exposure response were adequate and were reported in Figure 1.
	Metric 23: Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in Figure 1.
Additional Comments:	This portion of the evaluation was on the effect of DBP on the weight and length of <i>M. fluviatilis</i> . Development and growth was selected as the outcome of interest.		
Overall Quality Determination		High	

Study Citation:	EnviroSystem, (1991). Early life-stage toxicity of di-n-butyl phthalate (DnBP) to the rainbow trout (<i>Oncorhynchus mykiss</i>) under flow-through conditions.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oncorhynchus mykiss</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	6571362			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The test substance was identified by name only.	
	Metric 2: Test Substance Source	High	The source of the DBP was reported to be the Chemical Manufacturers Association in Washington DC, and was analytically verified by GC-MS.	
	Metric 3: Test Substance Purity	High	The purity of the DBP was reported to be 99.9%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of a negative control and a vehicle control.	
	Metric 5: Negative Control Response	Low	The negative control for the preliminary test was not reported.	
	Metric 6: Randomized Allocation	Low	It was not reported how juvenile fish were allocated into study groups in the preliminary test.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	Little information was provided on the preparation of the test concentrations.	
	Metric 8: Consistency of Exposure Administration	Low	All exposures for the preliminary test were conducted for 13 days. The test chambers for this portion of the study were not described. Little other information was provided regarding test conditions for the preliminary study.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if test concentrations were measured in the preliminary study.	
	Metric 10: Exposure Duration and Frequency	High	The test duration was reported to be 13 days. This was adequate for a preliminary test.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	There were five exposure groups as well as a negative control and a solvent control. This is typical for testing, and spacing was appropriate to observe a response.	
	Metric 12: Testing at or Below Solubility Limit	High	All test concentrations were below the water solubility limit, and a vehicle solvent was used. The solvent control had an appropriate response.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The trout were from Mt. Laassen Trout Farms and were the Hildebrand strain. They were certified to be disease free.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	Acclimation was not reported for the preliminary test.	
	Metric 15: Number of Organisms and Replicates per Group	Low	The number of fish per replicate and the number of replicates were not reported for the preliminary study.	
Domain 5: Outcome Assessment				
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Study Citation:	EnviroSystem, (1991). Early life-stage toxicity of di-n-butyl phthalate (DnBP) to the rainbow trout (<i>Oncorhynchus mykiss</i>) under flow-through conditions.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oncorhynchus mykiss</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	6571362			
Domain	Metric	Rating	Comments	
	Metric 16:	Adequacy of Test Conditions	Low	Test conditions were not reported for the preliminary study.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcomes of interest—fish survival in the terms of LC50 values for 96h and 13d.
	Metric 18:	Consistency of Outcome Assessment	Low	Minimal details were provided regarding outcome assessment for the preliminary study.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately for the preliminary study.
	Metric 22:	Reporting of Data	Low	Data were only reported in the text for the preliminary test.
	Metric 23:	Explanation of Unexpected Outcomes	Low	No measures of variability were reported for the preliminary study.
Additional Comments:	This evaluation is for the preliminary test conducted with juvenile rainbow trout. 96h and 13d LC50 values were reported. Little information regarding preliminary test protocol and procedures was reported. Mortality was the outcome of interest.			

Overall Quality Determination**Medium**

Study Citation:	EnviroSystem, (1991). Early life-stage toxicity of di-n-butyl phthalate (DnBP) to the rainbow trout (<i>Oncorhynchus mykiss</i>) under flow-through conditions.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oncorhynchus mykiss</i> ; Embryo			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	6571362			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The test substance was identified by name only.	
	Metric 2: Test Substance Source	High	The source of the DBP was reported to be the Chemical Manufacturers Association in Washington DC, and was analytically verified by GC-MS.	
	Metric 3: Test Substance Purity	High	The purity of the DBP was reported to be 99.9%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of a negative control and a vehicle control.	
	Metric 5: Negative Control Response	High	The negative control response and the vehicle control response were reported in Table 3 and in Tables B1 and B2.	
	Metric 6: Randomized Allocation	Medium	Embryos were randomly distributed to each exposure group.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	Little information was provided on the preparation of the test concentrations.	
	Metric 8: Consistency of Exposure Administration	Medium	All exposures were for 99 days unless all the organisms in the test concentration had died prior to the end of the study. All the tests were conducted in 14 x 53 x 25cm with 15L of test solution.	
	Metric 9: Measurement of Test Substance Concentration	High	The test concentrations were measured by two methods, a radiochemical analytical method and via GC/MS.	
	Metric 10: Exposure Duration and Frequency	High	The test duration was reported to be 99 days. This was adequate to observe a response across study groups.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	There were five exposure groups as well as a negative control and a solvent control. This is typical for testing, and spacing was appropriate to observe a response.	
	Metric 12: Testing at or Below Solubility Limit	High	All test concentrations were below the water solubility limit, and a vehicle solvent was used. The solvent control had an appropriate response.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The embryos and sperm were from Mt. Laassen Trout Farms and were the Hildebrand strain. They were certified to be disease free.	
	Metric 14: Acclimatization and Pretreatment Conditions	Medium	Acclimation was reported, but the duration was not reported.	
	Metric 15: Number of Organisms and Replicates per Group	Low	There were 30 embryos per test chamber with two replicates. Two replicates is less than is typical, thus the low ranking.	

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Study Citation:	EnviroSystem, (1991). Early life-stage toxicity of di-n-butyl phthalate (DnBP) to the rainbow trout (<i>Oncorhynchus mykiss</i>) under flow-through conditions.		
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Oncorhynchus mykiss</i> ; Embryo		
Health Outcome:	Mortality		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	6571362		
Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
Metric 16:	Adequacy of Test Conditions	High	Embryos were held at 10C for the first six weeks and then the temperature was gradually increased to 12.5C over week seven to the end of the study. Embryos were kept in the dark until day 43, at the start of swim-up, and then kept at a photoperiod of 14L:10D. Fry were fed starter mash and live brine shrimp.
Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcomes of interest—embryo/larval survival and percent hatch.
Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. Embryos and larvae were observed daily for mortalities.
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	High	Statistical methods were described in the "statistical analysis" section of the report.
Metric 22:	Reporting of Data	High	Mortality data for the control responses and the exposure responses were reported in Tables 3, B1, and B2 and were adequate for the outcomes of interest.
Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.
Additional Comments:	This portion of the evaluation was on the effect of DBP on embryo survival, percent hatch, and larval survival at various points in the 99 day study. Mortality was selected as the outcome of interest.		
Overall Quality Determination		High	

Study Citation:	EnviroSystem, (1991). Early life-stage toxicity of di-n-butyl phthalate (DnBP) to the rainbow trout (<i>Oncorhynchus mykiss</i>) under flow-through conditions.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oncorhynchus mykiss</i> ; Embryo			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	6571362			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The test substance was identified by name only.	
	Metric 2: Test Substance Source	Low	The source of the DBP was reported to be the Chemical Manufacturers Association in Washington DC. It was not reported if it was analytically verified.	
	Metric 3: Test Substance Purity	High	The purity of the DBP was reported to be 99.9%	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of a negative control and a vehicle control.	
	Metric 5: Negative Control Response	High	The negative control response and the vehicle control response were reported in Tables 4, 5, and in B3-B7.	
	Metric 6: Randomized Allocation	Medium	Embryos were randomly distributed to each exposure group.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	Little information was provided on the preparation of the test concentrations.	
	Metric 8: Consistency of Exposure Administration	Medium	All exposures were for 99 days unless all the organisms in the test concentration had died prior to the end of the study. All the tests were conducted in 14 x 53 x 25cm with 15L of test solution.	
	Metric 9: Measurement of Test Substance Concentration	High	The test concentrations were measured by two methods, a radiochemical analytical method and via GC/MS.	
	Metric 10: Exposure Duration and Frequency	High	The test duration was reported to be 99 days. This was adequate to observe a response across study groups.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	There were 5 exposure groups as well as a negative control and a solvent control. This is typical for testing, and spacing was appropriate to observe a response.	
	Metric 12: Testing at or Below Solubility Limit	High	All test concentrations were below the water solubility limit, and a vehicle solvent was used. The solvent control had an appropriate response.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The embryos and sperm were from Mt. Laassen Trout Farms and were the Hildebrand strain. They were certified to be disease free.	
	Metric 14: Acclimatization and Pretreatment Conditions	Medium	Acclimation was reported, but the duration was not reported.	
	Metric 15: Number of Organisms and Replicates per Group	Low	There were 30 embryos per test chamber with two replicates. Two replicates is less than is typical, thus the low ranking.	
Domain 5: Outcome Assessment				
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Study Citation:	EnviroSystem, (1991). Early life-stage toxicity of di-n-butyl phthalate (DnBP) to the rainbow trout (<i>Oncorhynchus mykiss</i>) under flow-through conditions.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oncorhynchus mykiss</i> ; Embryo			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	6571362			
Domain	Metric	Rating	Comments	
	Metric 16: Adequacy of Test Conditions	High	Embryos were held at 10C for the first 6 weeks and then the temperature was gradually increased to 12.5C over week 7 to the end of the study. Embryos were kept in the dark until day 43, at the start of swim-up, and then kept at a photoperiod of 14L:10D. Fry were fed starter mash and live brine shrimp.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—lengths and weights of larval fish, as well as any other sublethal effects.	
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. Caliper lengths and photographic lengths were taken at the end of the study, and wet weights and dry weights were both taken at the end of the study.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical methods were described in the "statistical analysis" section of the report.	
	Metric 22: Reporting of Data	High	Body weights and lengths were provided as well as other sublethal effects were reported in Tables 4, 5, and B3-B7.	
	Metric 23: Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in the tables.	
Additional Comments:	This portion of the evaluation was on the effect of DBP on fish length and width, as well as other sublethal effect. Development/growth was selected as the outcome of interest.			
Overall Quality Determination		High		

Study Citation:	Rhodes, J. E., Adams, W. J., Biddinger, G. R., Robillard, K. A., Gorsuch, J. W. (1995). Chronic toxicity of 14 phthalate esters to <i>Daphnia magna</i> and rainbow trout (<i>Oncorhynchus mykiss</i>). Environmental Toxicology and Chemistry 14(11):1967-1976.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oncorhynchus mykiss</i> ; Embryo			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	680120; Linked HERO ID(s): 1316195, 680120			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	Chemical identified by name	
	Metric 2: Test Substance Source	Low	The test substance identity was NOT analytically verified by the performing laboratory	
	Metric 3: Test Substance Purity	High	Purity reported as >95%	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control and solvent control	
	Metric 5: Negative Control Response	Low	The biological response of the negative control groups was not reported	
	Metric 6: Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study group	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The experimental system and methods for preparation of test media were described in adequate detail	
	Metric 8: Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups	
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were justified for a dose response by study authors	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized	
	Metric 15: Number of Organisms and Replicates per Group	Low	The numbers of test organisms was adequate but only two replicates were reported	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Organism environmental conditions were conducive to maintenance of organism health	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome	

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Study Citation:	Rhodes, J. E., Adams, W. J., Biddinger, G. R., Robillard, K. A., Gorsuch, J. W. (1995). Chronic toxicity of 14 phthalate esters to <i>Daphnia magna</i> and rainbow trout (<i>Oncorhynchus mykiss</i>). Environmental Toxicology and Chemistry 14(11):1967-1976.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oncorhynchus mykiss</i> ; Embryo			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	680120; Linked HERO ID(s): 1316195, 680120			
Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	High	There were no differences among groups that could influence the outcome assessment
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately
	Metric 22:	Reporting of Data	Low	Data were only reported for the most sensitive outcomes
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes
Additional Comments: None				

Overall Quality Determination**High**

Study Citation:	Rhodes, J. E., Adams, W. J., Biddinger, G. R., Robillard, K. A., Gorsuch, J. W. (1995). Chronic toxicity of 14 phthalate esters to <i>Daphnia magna</i> and rainbow trout (<i>Oncorhynchus mykiss</i>). Environmental Toxicology and Chemistry 14(11):1967-1976.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oncorhynchus mykiss</i> ; Embryo			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	680120; Linked HERO ID(s): 1316195, 680120			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The chemical was identified by name.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Test substance purity was reported as >95%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control and solvent control	
	Metric 5: Negative Control Response	Low	The biological response of the negative control groups was not reported.	
	Metric 6: Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The experimental system and methods for preparation of test media were described in adequate detail.	
	Metric 8: Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were justified for a dose response by study authors.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.	
	Metric 15: Number of Organisms and Replicates per Group	Low	The numbers of test organisms was adequate, but only two replicates were reported.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Organism housing and environmental conditions were conducive to maintenance of organism health.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome.	
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.	
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Study Citation:	Rhodes, J. E., Adams, W. J., Biddinger, G. R., Robillard, K. A., Gorsuch, J. W. (1995). Chronic toxicity of 14 phthalate esters to <i>Daphnia magna</i> and rainbow trout (<i>Oncorhynchus mykiss</i>). Environmental Toxicology and Chemistry 14(11):1967-1976.		
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Oncorhynchus mykiss</i> ; Embryo		
Health Outcome:	Development/Growth		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	680120; Linked HERO ID(s): 1316195, 680120		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
Metric 20:	Outcomes Unrelated to Exposure	High	There were no differences among groups that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately.
Metric 22:	Reporting of Data	Low	Data were only reported for the most sensitive outcomes.
Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments: None			
Overall Quality Determination		High	

Study Citation:	Bionomics,, EG&G (1983). Acute toxicity of fourteen phthalate esters to rainbow trout (<i>Salmo gairdneri</i>) under flow-through conditions (final report) report no BW-83-3-1373.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oncorhynchus mykiss</i> (<i>Salmo gairdneri</i>); Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5530771			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The test substance was identified by CASRN.	
	Metric 2: Test Substance Source	High	The test substance was obtained from General Electric Company, Hudson Falls, NY on 11 and 18 December 1981.	
	Metric 3: Test Substance Purity	High	The test substance was reported as "100% active ingredient." Though absolute purity is doubtful, this can be interpreted as an indication of very high purity.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Negative controls were included.	
	Metric 5: Negative Control Response	High	There was no unacceptable mortality in controls.	
	Metric 6: Randomized Allocation	Medium	Trout were randomly distributed among aquaria.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	A flow-through system with daily replenishment of solution was used. A detailed diluter design for mixing phthalates was described in Appendix I.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across substance groups.	
	Metric 9: Measurement of Test Substance Concentration	High	Test concentrations were measured during the experiment. Endpoints were based on measured concentrations.	
	Metric 10: Exposure Duration and Frequency	High	This was a 96-h acute exposure.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The range of concentrations and number of groups were acceptable to determine LC50 values.	
	Metric 12: Testing at or Below Solubility Limit	Medium	A dilution regimen for mixing the phthalates into solution was given in Appendix I. A combination of ultrasonication and mechanical mixing was used. Some of the high concentrations tested exhibited a visible film of undissolved phthalate. Although some measured concentrations were consistently lower than nominal, the measured concentrations were used to calculate LC50s.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	Fish were obtained from commercial suppliers in Maryland and Montana. Lot numbers were given.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	Acclimatization was for a minimum of 14 days in holding tanks.	
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Study Citation:	Bionomics,, EG&G (1983). Acute toxicity of fourteen phthalate esters to rainbow trout (<i>Salmo gairdneri</i>) under flow-through conditions (final report) report no BW-83-3-1373.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Oncorhynchus mykiss</i> (<i>Salmo gairdneri</i>); Adult
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	5530771

Domain	Metric	Rating	Comments
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were ten fish per group and two replicates per concentration.
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Test conditions were adequate for husbandry of rainbow trout.
	Metric 17: Outcome Assessment Methodology	High	The outcome was assessed appropriately (mortality).
	Metric 18: Consistency of Outcome Assessment	High	The outcome was assessed consistently among study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences in conditions among study groups.
	Metric 20: Outcomes Unrelated to Exposure	High	There were no reported outcomes unrelated to the exposure.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	The LC50 was calculated by a customized computer program using moving average angle analysis, probit analysis, or binomial probability. Details of the program were not reported.
	Metric 22: Reporting of Data	High	Data were reported for all groups.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments: None			

Overall Quality Determination**High**

Study Citation:	Erkmen, B., Benli, K., A.C., Agus, H. H., Yildirim, Z., Mert, R., Erkok, F. (2017). Impact of sublethal di-n-butyl phthalate on the aquaculture fish species Nile tilapia (<i>Oreochromis niloticus</i>): Histopathology and oxidative stress assessment. <i>Aquaculture Research</i> 48(2):675-685.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oreochromis niloticus</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3974179			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The test substance was identified by CASRN.	
	Metric 2: Test Substance Source	Low	The DBP was reported to be from Merck, but it was not reported to be analytically verified.	
	Metric 3: Test Substance Purity	Low	The purity/grade was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of a negative control.	
	Metric 5: Negative Control Response	High	The negative control response was reported in the text.	
	Metric 6: Randomized Allocation	Low	It was not reported how the fish were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The preparation of the test media and test concentration was not reported. Exposure was conducted in 60L glass aquaria.	
	Metric 8: Consistency of Exposure Administration	High	All exposures were for either 24 or 96h in 60L tanks at 23C with 10 fish per tank.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if the DBP was measured at any point in the study.	
	Metric 10: Exposure Duration and Frequency	High	The study duration was reported to be up to 96h with sampling done at 24h and 96h. This was appropriate for the outcome of interest.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	N/A	The goal of the study was not to have a dose response, but to observe the oxidative stress effect of one sublethal concentration of DBP on the fish.	
	Metric 12: Testing at or Below Solubility Limit	High	The test concentration was below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The tilapia were from the Cukurova University Freshwater Fish Production Station in Adana, Turkey and were juveniles.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	The tilapia were acclimated for 15d prior to the start of the study.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 10 fish per test chamber and 2 tanks per treatment per exposure duration.	
Domain 5: Outcome Assessment				
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Study Citation:	Erkmen, B., Benli, K., A.C., Agus, H. H., Yildirim, Z., Mert, R., Erkoc, F. (2017). Impact of sublethal di-n-butyl phthalate on the aquaculture fish species Nile tilapia (<i>Oreochromis niloticus</i>): Histopathology and oxidative stress assessment. <i>Aquaculture Research</i> 48(2):675-685.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oreochromis niloticus</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3974179			
Domain	Metric	Rating	Comments	
	Metric 16:	Adequacy of Test Conditions	High	The tilapia were kept in 60L tanks with 10 fish per tank at 23C with a 16L:8D photoperiod. They were fed commercial trout food at 2% their body weight.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest–mortality.
	Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were described under the "Data Analysis" section of the paper.
	Metric 22:	Reporting of Data	Low	Control and exposure responses were reported in the text only.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.
Additional Comments:	This portion of the evaluation was on the effect of DBP mortality.			
Overall Quality Determination		High		

Study Citation:	Erkmen, B., Benli, K., A.C., Agus, H. H., Yildirim, Z., Mert, R., Erkoc, F. (2017). Impact of sublethal di-n-butyl phthalate on the aquaculture fish species Nile tilapia (<i>Oreochromis niloticus</i>): Histopathology and oxidative stress assessment. <i>Aquaculture Research</i> 48(2):675-685.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oreochromis niloticus</i> ; Juvenile			
Health Outcome:	Respiratory			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3974179			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The test substance was identified by CASRN.	
	Metric 2: Test Substance Source	Low	The DBP was reported to be from Merck, but it was not reported to be analytically verified.	
	Metric 3: Test Substance Purity	Low	The purity/grade was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of a negative control.	
	Metric 5: Negative Control Response	High	The negative control response was reported in Figure 1 and was adequate for the outcome of interest.	
	Metric 6: Randomized Allocation	Low	It was not reported how the fish were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The preparation of the test media and test concentration was not reported. Exposure was conducted in 60L glass aquaria.	
	Metric 8: Consistency of Exposure Administration	High	All exposures were for either 24 or 96h in 60L tanks at 23C with 10 fish per tank.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if the DBP was measured at any point in the study.	
	Metric 10: Exposure Duration and Frequency	High	The study duration was reported to be up to 96h with sampling done at 24h and 96h. This was appropriate for the outcome of interest.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	N/A	The goal of the study was not to have a dose response, but to observe the histological effect of one sublethal concentration of DBP on the fish.	
	Metric 12: Testing at or Below Solubility Limit	High	The test concentration was below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The tilapia were from the Cukurova University Freshwater Fish Production Station in Adana, Turkey and were juveniles.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	The tilapia were acclimated for 15d prior to the start of the study.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 10 fish per test chamber and 2 tanks per treatment per exposure duration.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	The tilapia were kept in 60L tanks with 10 fish per tank at 23C with a 16L:8D photoperiod. They were fed commercial trout food at 2% their body weight.	
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Study Citation:	Erkmen, B., Benli, K., A.C., Agus, H. H., Yildirim, Z., Mert, R., Erkoc, F. (2017). Impact of sublethal di-n-butyl phthalate on the aquaculture fish species Nile tilapia (<i>Oreochromis niloticus</i>): Histopathology and oxidative stress assessment. Aquaculture Research 48(2):675-685.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oreochromis niloticus</i> ; Juvenile			
Health Outcome:	Respiratory			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3974179			
Domain		Metric	Rating	Comments
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—changes in gill histology.
	Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. Liver and gill tissue was fixed at the end of the study and examined via microscope for histological changes.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were described under the "Data Analysis" section of the paper.
	Metric 22:	Reporting of Data	High	Control and exposure responses are reported in Figure 1 and were adequate for the outcome of interest.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.
Additional Comments:	This portion of the evaluation was on the effect of DBP on the gills. Histopathology was performed, so the respiratory outcome was chosen.			

Overall Quality Determination**High**

Study Citation:	Erkmen, B., Benli, K., A.C., Agus, H. H., Yildirim, Z., Mert, R., Erkok, F. (2017). Impact of sublethal di-n-butyl phthalate on the aquaculture fish species Nile tilapia (<i>Oreochromis niloticus</i>): Histopathology and oxidative stress assessment. <i>Aquaculture Research</i> 48(2):675-685.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oreochromis niloticus</i> ; Juvenile			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3974179			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The test substance was identified by CASRN.	
	Metric 2: Test Substance Source	Low	The DBP was reported to be from Merck, but it was not reported to be analytically verified.	
	Metric 3: Test Substance Purity	Low	The purity/grade was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of a negative control.	
	Metric 5: Negative Control Response	High	The negative control response was reported in the text.	
	Metric 6: Randomized Allocation	Low	It was not reported how the fish were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The preparation of the test media and test concentration was not reported. Exposure was conducted in 60L glass aquaria.	
	Metric 8: Consistency of Exposure Administration	High	All exposures were for either 24 or 96h in 60L tanks at 23C with 10 fish per tank.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if the DBP was measured at any point in the study.	
	Metric 10: Exposure Duration and Frequency	High	The study duration was reported to be up to 96h with sampling done at 24h and 96h. This was appropriate for the outcome of interest.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	N/A	The goal of the study was not to have a dose response, but to observe the oxidative stress effect of one sublethal concentration of DBP on the fish.	
	Metric 12: Testing at or Below Solubility Limit	High	The test concentration was below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The tilapia were from the Cukurova University Freshwater Fish Production Station in Adana, Turkey and were juveniles.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	The tilapia were acclimated for 15d prior to the start of the study.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 10 fish per test chamber and 2 tanks per treatment per exposure duration.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	The tilapia were kept in 60L tanks with 10 fish per tank at 23C with a 16L:8D photoperiod. They were fed commercial trout food at 2% their body weight.	
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Study Citation:	Erkmen, B., Benli, K., A.C., Agus, H. H., Yildirim, Z., Mert, R., Erkoc, F. (2017). Impact of sublethal di-n-butyl phthalate on the aquaculture fish species Nile tilapia (<i>Oreochromis niloticus</i>): Histopathology and oxidative stress assessment. <i>Aquaculture Research</i> 48(2):675-685.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Oreochromis niloticus</i> ; Juvenile
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	3974179

Domain	Metric	Rating	Comments
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest–growth.
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were described under the "Data Analysis" section of the paper.
	Metric 22: Reporting of Data	Low	Control and exposure responses were reported in the text only.
	Metric 23: Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.

Additional Comments: This portion of the evaluation was on the effect of DBP development/growth.

Overall Quality Determination

High

Study Citation:	Erkmen, B., Benli, K., A.C., Agus, H. H., Yildirim, Z., Mert, R., Erkok, F. (2017). Impact of sublethal di-n-butyl phthalate on the aquaculture fish species Nile tilapia (<i>Oreochromis niloticus</i>): Histopathology and oxidative stress assessment. <i>Aquaculture Research</i> 48(2):675-685.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oreochromis niloticus</i> ; Juvenile			
Health Outcome:	Hepatic/Liver			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3974179			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The test substance was identified by CASRN.	
	Metric 2: Test Substance Source	Low	The DBP was reported to be from Merck, but it was not reported to be analytically verified.	
	Metric 3: Test Substance Purity	Low	The purity/grade was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of a negative control.	
	Metric 5: Negative Control Response	High	The negative control response was reported in Figure 2 and was adequate for the outcome of interest.	
	Metric 6: Randomized Allocation	Low	It was not reported how the fish were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The preparation of the test media and test concentration was not reported. Exposure was conducted in 60L glass aquaria.	
	Metric 8: Consistency of Exposure Administration	High	All exposures were for either 24 or 96h in 60L tanks at 23C with 10 fish per tank.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if the DBP was measured at any point in the study.	
	Metric 10: Exposure Duration and Frequency	High	The study duration was reported to be up to 96h with sampling done at 24h and 96h. This was appropriate for the outcome of interest.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	N/A	The goal of the study was not to have a dose response, but to observe the histological effect of one sublethal concentration of DBP on the fish.	
	Metric 12: Testing at or Below Solubility Limit	High	The test concentration was below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The tilapia were from the Cukurova University Freshwater Fish Production Station in Adana, Turkey and were juveniles.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	The tilapia were acclimated for 15d prior to the start of the study.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 10 fish per test chamber and 2 tanks per treatment per exposure duration.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	The tilapia were kept in 60L tanks with 10 fish per tank at 23C with a 16L:8D photoperiod. They were fed commercial trout food at 2% their body weight.	
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Study Citation:	Erkmen, B., Benli, K., A.C., Agus, H. H., Yildirim, Z., Mert, R., Erkoc, F. (2017). Impact of sublethal di-n-butyl phthalate on the aquaculture fish species Nile tilapia (<i>Oreochromis niloticus</i>): Histopathology and oxidative stress assessment. <i>Aquaculture Research</i> 48(2):675-685.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oreochromis niloticus</i> ; Juvenile			
Health Outcome:	Hepatic/Liver			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3974179			
Domain		Metric	Rating	Comments
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—changes in liver histology.
	Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. Liver and gill tissue was fixed at the end of the study and examined via microscope for histological changes.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were described under the "Data Analysis" section of the paper.
	Metric 22:	Reporting of Data	High	Control and exposure responses are reported in Figure 2 and were adequate for the outcome of interest.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.
Additional Comments:	This portion of the evaluation was on the effect of DBP on the liver. Histopathology was performed, so the liver/hepatic outcome was chosen.			

Overall Quality Determination**High**

Study Citation:	Erkmen, B., Benli, K., A.C., Agus, H. H., Yildirim, Z., Mert, R., Erkoc, F. (2017). Impact of sublethal di-n-butyl phthalate on the aquaculture fish species Nile tilapia (<i>Oreochromis niloticus</i>): Histopathology and oxidative stress assessment. <i>Aquaculture Research</i> 48(2):675-685.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oreochromis niloticus</i> ; Juvenile			
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)-Immune/Hematological			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3974179			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The test substance was identified by CASRN.	
	Metric 2: Test Substance Source	Low	The DBP was reported to be from Merck, but it was not reported to be analytically verified.	
	Metric 3: Test Substance Purity	Low	The purity/grade was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of a negative control.	
	Metric 5: Negative Control Response	High	The negative control response was reported in Table 1 for MDA and GSH and was adequate for the outcome of interest. HCT control responses were presented in the text.	
	Metric 6: Randomized Allocation	Low	It was not reported how the fish were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The preparation of the test media and test concentration was not reported. Exposure was conducted in 60L glass aquaria.	
	Metric 8: Consistency of Exposure Administration	High	All exposures were for either 24 or 96h in 60L tanks at 23C with 10 fish per tank.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if the DBP was measured at any point in the study.	
	Metric 10: Exposure Duration and Frequency	High	The study duration was reported to be up to 96h with sampling done at 24h and 96h. This was appropriate for the outcome of interest.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	N/A	The goal of the study was not to have a dose response, but to observe the oxidative stress effect of one sublethal concentration of DBP on the fish.	
	Metric 12: Testing at or Below Solubility Limit	High	The test concentration was below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The tilapia were from the Cukurova University Freshwater Fish Production Station in Adana, Turkey and were juveniles.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	The tilapia were acclimated for 15d prior to the start of the study.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were ten fish per test chamber and two tanks per treatment per exposure duration.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	The tilapia were kept in 60L tanks with 10 fish per tank at 23C with a 16L:8D photoperiod. They were fed commercial trout food at 2% their body weight.	
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Study Citation:	Erkmen, B., Benli, K., A.C., Agus, H. H., Yildirim, Z., Mert, R., Erkoc, F. (2017). Impact of sublethal di-n-butyl phthalate on the aquaculture fish species Nile tilapia (<i>Oreochromis niloticus</i>): Histopathology and oxidative stress assessment. <i>Aquaculture Research</i> 48(2):675-685.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oreochromis niloticus</i> ; Juvenile			
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)-Immune/Hematological			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3974179			
Domain	Metric	Rating	Comments	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcomes of interest—changes in HCT, GSH, and MDA.	
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical methods were described under the "Data Analysis" section of the paper.	
	Metric 22: Reporting of Data	Medium	Control and exposure responses for oxidative stress were reported in Table 1. The HCT results for the control and the exposure were only reported in the text.	
	Metric 23: Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in Table 1 and in the text.	
Additional Comments:	This portion of the evaluation was on the effect of DBP on MDA, GSH, and HCT. The mechanistic outcomes for oxidative stress and hematological/immunity were chosen.			
Overall Quality Determination		High		

Study Citation:	Khalil, , S. R., Elhakim, Abd, Y., El-Murr, A. E. (2016). Sublethal concentrations of di-n-butyl phthalate promote biochemical changes and DNA damage in juvenile Nile tilapia (<i>Oreochromis niloticus</i>). Japanese Journal of Veterinary Research 64(1):67-80.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oreochromis niloticus</i> ; Juvenile			
Health Outcome:	Renal/Kidney			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3350208			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance	Metric 1: Test Substance Identity	High	Chemical was identified by name and CAS#.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity reported as 99% by the manufacturer.	
Domain 2: Test Design	Metric 4: Negative Controls	Low	Study authors reported using an appropriate concurrent control group, but did not indicate if that control group represented DMSO or not. So, no solvent control or negative control was indicated, just a control. Also, authors did not report the amount of DMSO used for treatment groups.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.	
	Metric 6: Randomized Allocation	Medium	The biological response of the negative control group was reported and reasonable for assessed outcomes.	
Domain 3: Exposure Characterization	Metric 7: Experimental System/Test Media Preparation	Low	The study provided few details on exposure media preparation. The acclimation period detailed glass aquaria but no details on vessel media type for sub-lethal tests were provided.	
	Metric 8: Consistency of Exposure Administration	Medium	No mention of irregularities in exposure administration.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Medium	The number of exposure groups and spacing of exposure levels were suitable for a dose response although only concentrations were tested.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations (5.9 and 3.9 mg/L) were at or below the water solubility limit (11.2 mg/L).	
Domain 4: Test Organism	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source. Mean size was reported as 10.6 grams.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions for two weeks.	
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Study Citation:	Khalil, , S. R., Elhakim, Abd, Y., El-Murr, A. E. (2016). Sublethal concentrations of di-n-butyl phthalate promote biochemical changes and DNA damage in juvenile Nile tilapia (<i>Oreochromis niloticus</i>). Japanese Journal of Veterinary Research 64(1):67-80.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Oreochromis niloticus</i> ; Juvenile
Health Outcome:	Renal/Kidney
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	3350208

Domain	Metric	Rating	Comments
	Metric 15: Number of Organisms and Replicates per Group	Medium	Exposures were conducted with two replicates of 10 fish each for treatment and control exposures.
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described with ANOVA analysis and a Duncans post-hoc.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group in Figure 5.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments: This form is for the histopathological assessment of kidney tissue.			

Overall Quality Determination**Medium**

Study Citation:	Khalil, , S. R., Elhakim, Abd, Y., El-Murr, A. E. (2016). Sublethal concentrations of di-n-butyl phthalate promote biochemical changes and DNA damage in juvenile Nile tilapia (<i>Oreochromis niloticus</i>). Japanese Journal of Veterinary Research 64(1):67-80.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oreochromis niloticus</i> ; Juvenile			
Health Outcome:	Hepatic/Liver			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3350208			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	Chemical was identified by name and CAS#.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity reported as 99% by the manufacturer.	
Domain 2: Test Design				
	Metric 4: Negative Controls	Low	Study authors reported using an appropriate concurrent control group, but did not indicate if that control group represented DMSO or not. So, no solvent control or negative control was indicated, just a control. Also, authors did not report the amount of DMSO used for treatment groups.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.	
	Metric 6: Randomized Allocation	Medium	The biological response of the negative control group was reported and reasonable for assessed outcomes.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided few details on exposure media preparation. The acclimation period detailed glass aquaria but no details on vessel media type for sub-lethal tests were provided.	
	Metric 8: Consistency of Exposure Administration	Medium	No mention of irregularities in exposure administration.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Medium	The number of exposure groups and spacing of exposure levels were suitable for a dose response although only concentrations were tested.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations (5.9 and 3.9 mg/L) were at or below the water solubility limit (11.2 mg/L).	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source. Mean size was reported as 10.6 grams.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions for two weeks.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	Exposures were conducted with two replicates of 10 fish each for treatment and control exposures.	

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Study Citation:	Khalil, , S. R., Elhakim, Abd, Y., El-Murr, A. E. (2016). Sublethal concentrations of di-n-butyl phthalate promote biochemical changes and DNA damage in juvenile Nile tilapia (<i>Oreochromis niloticus</i>). Japanese Journal of Veterinary Research 64(1):67-80.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Oreochromis niloticus</i> ; Juvenile
Health Outcome:	Hepatic/Liver
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	3350208

Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described with ANOVA analysis and a Duncans post-hoc.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group in Figure 4.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments: This form is for the histopathological assessment of liver tissue.			

Overall Quality Determination**Medium**

Study Citation:	Khalil, , S. R., Elhakim, Abd, Y., El-Murr, A. E. (2016). Sublethal concentrations of di-n-butyl phthalate promote biochemical changes and DNA damage in juvenile Nile tilapia (<i>Oreochromis niloticus</i>). Japanese Journal of Veterinary Research 64(1):67-80.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oreochromis niloticus</i> ; Juvenile			
Health Outcome:	Respiratory			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3350208			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#.
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99% by the manufacturer.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Study authors reported using an appropriate concurrent control group, but did not indicate if that control group represented DMSO or not. So, no solvent control or negative control was indicated, just a control. Also, authors did not report the amount of DMSO used for treatment groups.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.
	Metric 6:	Randomized Allocation	Medium	The biological response of the negative control group was reported and reasonable for assessed outcomes.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided few details on exposure media preparation. The acclimation period detailed glass aquaria but no details on vessel media type for sub-lethal tests were provided.
	Metric 8:	Consistency of Exposure Administration	Medium	No mention of irregularities in exposure administration.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	The number of exposure groups and spacing of exposure levels were suitable for a dose response although only concentrations were tested.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations (5.9 and 3.9 mg/L) were at or below the water solubility limit (11.2 mg/L).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source. Mean size was reported as 10.6 grams.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions for two weeks.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Exposures were conducted with two replicates of 10 fish each for treatment and control exposures.

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Study Citation:	Khalil, , S. R., Elhakim, Abd, Y., El-Murr, A. E. (2016). Sublethal concentrations of di-n-butyl phthalate promote biochemical changes and DNA damage in juvenile Nile tilapia (<i>Oreochromis niloticus</i>). Japanese Journal of Veterinary Research 64(1):67-80.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Oreochromis niloticus</i> ; Juvenile
Health Outcome:	Respiratory
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	3350208

Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described with ANOVA analysis and a Duncans post-hoc.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group in Figure 5.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments: This form is for the histopathological assessment of gill tissue.			

Overall Quality Determination**Medium**

Study Citation:	Khalil, , S. R., Elhakim, Abd, Y., El-Murr, A. E. (2016). Sublethal concentrations of di-n-butyl phthalate promote biochemical changes and DNA damage in juvenile Nile tilapia (<i>Oreochromis niloticus</i>). Japanese Journal of Veterinary Research 64(1):67-80.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oreochromis niloticus</i> ; Juvenile			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Oxidative stress (including redox biology)-Cardiovascular-Endocrine toxicity-Liver toxicology			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3350208			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99% by the manufacturer.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Study authors reported using an appropriate concurrent control group, but did not indicate if that control group represented DMSO or not. So, no solvent control or negative control was indicated, just a control. Also, authors did not report the amount of DMSO used for treatment groups.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.
	Metric 6:	Randomized Allocation	Medium	The biological response of the negative control group was reported and reasonable for assessed outcomes.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided few details on exposure media preparation. The acclimation period detailed glass aquaria but no details on vessel media type for sub-lethal tests were provided.
	Metric 8:	Consistency of Exposure Administration	Medium	No mention of irregularities in exposure administration.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	The number of exposure groups and spacing of exposure levels were suitable for a dose response although only concentrations were tested.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations (5.9 and 3.9 mg/L) were at or below the water solubility limit (11.2 mg/L).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source. Mean size was reported as 10.6 grams.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions for two weeks.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Exposures were conducted with two replicates of 10 fish each for treatment and control exposures.

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Study Citation:	Khalil, , S. R., Elhakim, Abd, Y., El-Murr, A. E. (2016). Sublethal concentrations of di-n-butyl phthalate promote biochemical changes and DNA damage in juvenile Nile tilapia (<i>Oreochromis niloticus</i>). Japanese Journal of Veterinary Research 64(1):67-80.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Oreochromis niloticus</i> ; Juvenile
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Oxidative stress (including redox biology)-Cardiovascular-Endocrine toxicity-Liver toxicology
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	3350208

Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described with ANOVA analysis and a Duncans post-hoc.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This form represents outcomes associated with SOD, GSH, MDA, and Comet assay (quantitatively used as an indirect measure of DNA damage, Fig 3).

Overall Quality Determination

Medium

Study Citation:	Khalil, , S. R., Elhakim, Abd, Y., El-Murr, A. E. (2016). Sublethal concentrations of di-n-butyl phthalate promote biochemical changes and DNA damage in juvenile Nile tilapia (<i>Oreochromis niloticus</i>). Japanese Journal of Veterinary Research 64(1):67-80.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oreochromis niloticus</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3350208			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The chemical was identified by name and CAS#.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity was reported as 99% from the manufacturer.	
Domain 2: Test Design				
	Metric 4: Negative Controls	Low	Study authors reported using an appropriate concurrent control group, but did not indicate if that control group represented DMSO or not. So, no solvent control or negative control was indicated, just a control. Also, authors did not report the amount of DMSO used for treatment groups.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.	
	Metric 6: Randomized Allocation	Medium	The biological response of the negative control group was reported and reasonable for assessed outcomes.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided few details on exposure media preparation. The acclimation period detailed glass aquaria but there were no details on vessel media type for the mortality tests.	
	Metric 8: Consistency of Exposure Administration	Medium	There was no mention of irregularities in exposure administration. Authors did not report if the control group also had DMSO.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured, and water replacement was not detailed for the mortality portion of the study.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	Low	Five of the lower concentrations of DBP were below solubility listed in the scope (11.2), while seven of the treatment concentrations were above solubility.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source. Mean weight was reported as 10.6 grams.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions for two weeks.	
	Metric 15: Number of Organisms and Replicates per Group	Low	No replicates were used. Guideline, OCSPP 850.1075, recommends two replicates of ten fish each.	
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Study Citation:	Khalil, , S. R., Elhakim, Abd, Y., El-Murr, A. E. (2016). Sublethal concentrations of di-n-butyl phthalate promote biochemical changes and DNA damage in juvenile Nile tilapia (<i>Oreochromis niloticus</i>). Japanese Journal of Veterinary Research 64(1):67-80.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Oreochromis niloticus</i> ; Juvenile
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	3350208

Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Statistical methods were adequately described, however, the LC50 method used did not allow for the calculation of error terms surrounding the LC50 value obtained.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments: An LC50 value was obtained (11.8 mg/L) that is over the solubility for this compound (11.2 mg/L).			

Overall Quality Determination**Medium**

Study Citation:	Patyna, P. J. (1999). Reproductive effects of phthalate esters in Japanese medaka (<i>Oryzias latipes</i>). Doctoral Dissertation:137.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Embryo			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5489073; Linked HERO ID(s): 5489073, 680110			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Nomenclature was reported in section 2.1. The structure was identified in a figure within the dissertation. CAS number was not reported.
	Metric 2:	Test Substance Source	High	GCMS was reported on the compound in section 2.1 and the lot number was provided. Source was reported as from Sigma Aldrich.
	Metric 3:	Test Substance Purity	High	A purity of 99% via GCMS was reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	The authors reported performing control and solvent controls.
	Metric 5:	Negative Control Response	Low	No control responses for development are reported for control groups.
	Metric 6:	Randomized Allocation	Low	Allocation was not random.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	A Static non renewal 10 day exposure was conducted. At 10 days, replacement water was free of the compound. Authors used teflon rearing containers to reduce the leeching of test compound to the containers.
	Metric 8:	Consistency of Exposure Administration	High	exposures were the same across treatments and control groups.
	Metric 9:	Measurement of Test Substance Concentration	Medium	Measured concentrations are reported in table 4.2/4.3, but developmental observations were not reported for each concentration.
	Metric 10:	Exposure Duration and Frequency	High	The exposure (10 days) was reported to cover the normal period of embryo development for this species. Modal hatch was recorded between 11-14 days.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	The incidence of lesions are reported below and above the LC50 for DBP, so it is not able to relate to a dose-response gradient.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The source was listed as from Carolina Biological Supply. These fish were bred to produce embryos used in the study.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Embryo media and test water conditions were reported on page 45/158.
	Metric 15:	Number of Organisms and Replicates per Group	Low	The authors report the exposures were done in triplicate, however, sample numbers reported in the results do not reflect replicated sample numbers (Table 4.2-4.3) pages 90,91/158.

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Study Citation:	Patyna, P. J. (1999). Reproductive effects of phthalate esters in Japanese medaka (<i>Oryzias latipes</i>). Doctoral Dissertation:137.		
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 4 - 10 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Embryo		
Health Outcome:	Development/Growth		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	5489073; Linked HERO ID(s): 5489073, 680110		
Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
Metric 16:	Adequacy of Test Conditions	Medium	Embryo housing was detailed for the 10 day exposure, however, authors did not report water quality results to ensure adequate environmental conditions during embryogenesis.
Metric 17:	Outcome Assessment Methodology	Low	The authors described morphological lesions from DBP exposure but failed to present data on concentration specific incidences.
Metric 18:	Consistency of Outcome Assessment	Low	It is not clear how developmental morphology was scored at each concentration and control for the lesions listed in table 4.5 (page 94/158). Data are not presented.
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	High	There was no indication that reported differences were from environmental conditions or other confounding variables.
Metric 20:	Outcomes Unrelated to Exposure	Medium	No information to suggest differences among treatment and control groups attributed to animal health or attrition.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	Uninformative	The authors report the presence of lesions with no data on quantifying at difference exposure concentrations.
Metric 22:	Reporting of Data	Low	There is not enough data on incidence of lesions to relate to a dose-response gradient.
Metric 23:	Explanation of Unexpected Outcomes	Low	Not enough data are presented for lesions at specific concentrations to determine.
Additional Comments: This evaluation form is relevant to the morphological lesions results given on page 94/158.			

Overall Quality Determination**Uninformative**

Study Citation:	Patyna, P. J. (1999). Reproductive effects of phthalate esters in Japanese medaka (<i>Oryzias latipes</i>). Doctoral Dissertation:137.		
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 4 - 10 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Embryo		
Health Outcome:	Renal/Kidney		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	5489073; Linked HERO ID(s): 5489073, 680110		
Domain	Metric	Rating	Comments
Domain 1: Test Substance			
	Metric 1: Test Substance Identity	High	Nomenclature was reported in section 2.1. The structure was identified in a figure within the dissertation. CAS number was not reported.
	Metric 2: Test Substance Source	High	GCMS was reported on the compound in section 2.1 and the lot number was provided. Source was reported as from Sigma Aldrich.
	Metric 3: Test Substance Purity	High	A purity of 99% via GCMS was reported.
Domain 2: Test Design			
	Metric 4: Negative Controls	High	The authors reported performing control and solvent controls.
	Metric 5: Negative Control Response	High	Control responses are acceptable and presented in Table 4.2 and 4.3 (Pages 90,91/158).
	Metric 6: Randomized Allocation	Low	Allocation was not random.
Domain 3: Exposure Characterization			
	Metric 7: Experimental System/Test Media Preparation	High	A Static non renewal 10 day exposure was conducted. At 10 days, replacement water was free of the compound. Authors used teflon rearing containers to reduce the leeching of test compound to the containers.
	Metric 8: Consistency of Exposure Administration	High	Exposures were the same across treatments and control groups.
	Metric 9: Measurement of Test Substance Concentration	High	The test compound was verified and concentration gradients are reported as measured concentrations in Tables 4.2 and 4.3 (pages 90,91/158).
	Metric 10: Exposure Duration and Frequency	High	The exposure (10 days) was reported to cover the normal period of embryo development for this species. Modal hatch was recorded between 11-14 days.
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Medium	The authors report five measured concentrations that span relevance in no effects to complete mortality. No indication of prior range finding were reported.
	Metric 12: Testing at or Below Solubility Limit	High	All concentrations reported for DBP were under the solubility reported in the final scope (11 mg/l).
Domain 4: Test Organism			
	Metric 13: Test Organism Characteristics	High	The source was listed as from Carolina Biological Supply. These fish were bred to produce embryos used in the study.
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms. Embryo media and test water conditions were reported on page 45/158.
	Metric 15: Number of Organisms and Replicates per Group	Low	The authors report the exposures were done in triplicate, however, sample numbers reported in the results do not reflect replicated sample numbers (Table 4.2-4.3) pages 90,91/158.
Domain 5: Outcome Assessment			

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Study Citation:	Patyna, P. J. (1999). Reproductive effects of phthalate esters in Japanese medaka (<i>Oryzias latipes</i>). Doctoral Dissertation:137.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Embryo			
Health Outcome:	Renal/Kidney			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5489073; Linked HERO ID(s): 5489073, 680110			
Domain	Metric	Rating	Comments	
	Metric 16:	Adequacy of Test Conditions	Medium	Embryo housing was detailed for the 10 day exposure, however, authors did not report water quality results to ensure adequate environmental conditions during embryogenesis.
	Metric 17:	Outcome Assessment Methodology	High	Outcome assessment methodology reported the intended outcomes (urinary bladder lesions) appropriately for these embryo exposures.
	Metric 18:	Consistency of Outcome Assessment	High	The assessments were conducted similarly across treatment and control groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There was no indication that reported differences were from environmental conditions or other confounding variables.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	No information to suggest differences among treatment and control groups attributed to animal health or attrition.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Authors report using USEPA Probit analysis ver 1.4 for LC and EC50 calculations.
	Metric 22:	Reporting of Data	Low	Authors report EC50 for urinary bladder lesions (page 50/158) with no Confidence Intervals and no other components of the dose-response curve (NOEC, LOEC).
	Metric 23:	Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability (e.g., SE, SD, confidence intervals) and/or insufficient information was provided to determine if excessive variability or unexpected outcomes occurred.
Additional Comments:	This evaluation form is relevant to the urinary bladder inflation results results given in Table 4.2.			
Overall Quality Determination		High		

Study Citation:	Patyna, P. J. (1999). Reproductive effects of phthalate esters in Japanese medaka (<i>Oryzias latipes</i>). Doctoral Dissertation:137.		
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 4 - 10 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Embryo		
Health Outcome:	Mortality		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	5489073; Linked HERO ID(s): 5489073, 680110		
Domain	Metric	Rating	Comments
Domain 1: Test Substance			
Metric 1:	Test Substance Identity	High	Nomenclature was reported in section 2.1. The structure was identified in a figure within the dissertation. CAS number was not reported.
Metric 2:	Test Substance Source	High	GCMS was reported on the compound in section 2.1 and the lot number was provided. Source was reported as from Sigma Aldrich.
Metric 3:	Test Substance Purity	High	A purity of 99% via GCMS was reported.
Domain 2: Test Design			
Metric 4:	Negative Controls	High	The authors reported performing control and solvent controls.
Metric 5:	Negative Control Response	High	Control responses are acceptable and presented in Table 4.2 and 4.3 (Pages 90,91/158).
Metric 6:	Randomized Allocation	Low	Allocation was not random.
Domain 3: Exposure Characterization			
Metric 7:	Experimental System/Test Media Preparation	High	A Static non renewal 10 day exposure was conducted. At 10 days, replacement water was free of the compound. Authors used teflon rearing containers to reduce the leeching of test compound to the containers.
Metric 8:	Consistency of Exposure Administration	High	Exposures were the same across treatments and control groups.
Metric 9:	Measurement of Test Substance Concentration	High	The test compound was verified and concentration gradients are reported as measured concentrations in Tables 4.2 and 4.3 (pages 90,91/158).
Metric 10:	Exposure Duration and Frequency	High	The exposure (10 days) was reported to cover the normal period of embryo development for this species. Modal hatch was recorded between 11-14 days.
Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	The authors report five measured concentrations that span relevance in no effects to complete mortality. No indication of prior range finding were reported.
Metric 12:	Testing at or Below Solubility Limit	High	All concentrations reported for DBP were under the solubility reported in the final scope (11 mg/l).
Domain 4: Test Organism			
Metric 13:	Test Organism Characteristics	High	The source was listed as from Carolina Biological Supply. These fish were bred to produce embryos used in the study.
Metric 14:	Acclimatization and Pretreatment Conditions	High	Embryo media and test water conditions were reported on page 45/158.
Metric 15:	Number of Organisms and Replicates per Group	Low	The authors report the exposures were done in triplicate, however, sample numbers reported in the results do not reflect replicated sample numbers (Table 4.2-4.3) pages 90,91/158.
Domain 5: Outcome Assessment			
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Study Citation:	Patyna, P. J. (1999). Reproductive effects of phthalate esters in Japanese medaka (<i>Oryzias latipes</i>). Doctoral Dissertation:137.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Embryo			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5489073; Linked HERO ID(s): 5489073, 680110			
Domain	Metric	Rating	Comments	
	Metric 16: Adequacy of Test Conditions	Medium	Embryo housing was detailed for the 10 day exposure, however, authors did not report water quality results to ensure adequate environmental conditions during embryogenesis.	
	Metric 17: Outcome Assessment Methodology	High	Outcome assessment methodology reported the intended outcomes (hatch, mortality, development) appropriately for these embryo exposures.	
	Metric 18: Consistency of Outcome Assessment	High	The assessments were conducted similarly across treatment and control groups.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	There was no indication that reported differences were from environmental conditions or other confounding variables.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	No information to suggest differences among treatment and control groups attributed to animal health or attrition.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Authors report using USEPA Probit analysis ver 1.4 for LC and EC50 calculations.	
	Metric 22: Reporting of Data	Low	Authors report LC50 for mortality with no Confidence Intervals and no other components of the dose-response curve (NOEC, LOEC). Percent 3 day survival was also given (Table 4.3) without any measures of variability.	
	Metric 23: Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability (e.g., SE, SD, confidence intervals) and/or insufficient information was provided to determine if excessive variability or unexpected outcomes occurred.	
Additional Comments:	Authors report LC50 for mortality with no Confidence Intervals and no other components of the dose-response curve (NOEC, LOEC).			
Overall Quality Determination		High		

Study Citation:	Patyna, P. J. (1999). Reproductive effects of phthalate esters in Japanese medaka (<i>Oryzias latipes</i>). Doctoral Dissertation:137.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Embryo			
Health Outcome:	Cardiovascular			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5489073; Linked HERO ID(s): 5489073, 680110			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
Metric 1:	Test Substance Identity	High	Nomenclature was reported in section 2.1. The structure was identified in a figure within the dissertation. The CAS number was not reported.	
Metric 2:	Test Substance Source	High	GCMS was reported on the compound in section 2.1 and the lot number was provided. The source was reported as from Sigma Aldrich.	
Metric 3:	Test Substance Purity	High	A purity of 99% via GCMS was reported.	
Domain 2: Test Design				
Metric 4:	Negative Controls	High	The authors reported performing control and solvent controls.	
Metric 5:	Negative Control Response	Low	No control responses for development are reported for control groups.	
Metric 6:	Randomized Allocation	Low	Allocation was not random.	
Domain 3: Exposure Characterization				
Metric 7:	Experimental System/Test Media Preparation	High	A static non-renewal 10-day exposure was conducted. At 10 days, replacement water was free of the compound. Authors used teflon rearing containers to reduce the leeching of test compound to the containers.	
Metric 8:	Consistency of Exposure Administration	High	Exposures were the same across treatments and control groups.	
Metric 9:	Measurement of Test Substance Concentration	Medium	Measured concentrations are reported in table 4.2/4.3, but developmental observations were not reported for each concentration.	
Metric 10:	Exposure Duration and Frequency	High	The exposure (10 days) was reported to cover the normal period of embryo development for this species. Modal hatch was recorded between 11-14 days.	
Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	The incidence of lesions are reported below and above the LC50 for DBP, so it is not able to relate to a dose-response gradient.	
Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit.	
Domain 4: Test Organism				
Metric 13:	Test Organism Characteristics	High	The source was listed as from Carolina Biological Supply. These fish were bred to produce embryos used in the study.	
Metric 14:	Acclimatization and Pretreatment Conditions	High	Embryo media and test water conditions were reported on page 45/158.	
Metric 15:	Number of Organisms and Replicates per Group	Low	The authors report the exposures were done in triplicate, however, sample numbers reported in the results do not reflect replicated sample numbers (Table 4.2-4.3) pages 90, 91/158.	
Domain 5: Outcome Assessment				
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Study Citation:	Patyna, P. J. (1999). Reproductive effects of phthalate esters in Japanese medaka (<i>Oryzias latipes</i>). Doctoral Dissertation:137.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Embryo			
Health Outcome:	Cardiovascular			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5489073; Linked HERO ID(s): 5489073, 680110			
Domain	Metric	Rating	Comments	
	Metric 16: Adequacy of Test Conditions	Medium	Embryo housing was detailed for the 10-day exposure, however, authors did not report water quality results to ensure adequate environmental conditions during embryogenesis.	
	Metric 17: Outcome Assessment Methodology	Low	The authors described morphological lesions from DBP exposure but failed to present data on concentration specific incidences.	
	Metric 18: Consistency of Outcome Assessment	Low	It is not clear how developmental morphology was scored at each concentration and control for the lesions listed in table 4.5 (page 94/158). Data are not presented.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	There was no indication that reported differences were from environmental conditions or other confounding variables.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information to suggest differences among treatment and control groups attributed to animal health or attrition.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	N/A	The results were qualitative.	
	Metric 22: Reporting of Data	Uninformative	Not sufficient reporting of results (Table 4.5).	
	Metric 23: Explanation of Unexpected Outcomes	Low	Not enough data are presented for cardio effects at specific concentrations to determine if there are any unexpected outcomes.	
Additional Comments:	This evaluation form is relevant to the blood flow and edema histo results.			

Overall Quality Determination**Uninformative**

Study Citation:	EAG Laboratories, (2018). Dibutyl phthalate: Medaka extended one generation reproduction test (final report).			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	10064186			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	Chemical was identified by name and CAS#	
	Metric 2: Test Substance Source	High	The test substance identity was analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity reported as 99.3%	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors used a concurrent negative control group	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations	
	Metric 8: Consistency of Exposure Administration	Medium	Reporting omissions are unlikely to have a substantial impact on results.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured or measurements were not reported.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The test organisms were not adequately described and assumed to be same source as definitive test	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health	
	Metric 17: Outcome Assessment Methodology	Low	Environmental conditions were not sufficiently reported to evaluate if adequate	
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Study Citation:	EAG Laboratories, (2018). Dibutyl phthalate: Medaka extended one generation reproduction test (final report).			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	10064186			
Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Sufficient data were provided to conduct an independent statistical analysis.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes
Additional Comments: range finder				
Overall Quality Determination			Medium	

Study Citation:	EAG Laboratories, (2018). Dibutyl phthalate: Medaka extended one generation reproduction test (final report).			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	10064186			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The chemical was identified by name and CAS#.	
	Metric 2: Test Substance Source	High	The test substance identity was analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity was reported as 99.3%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors used a concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.	
	Metric 8: Consistency of Exposure Administration	Medium	Reporting omissions are unlikely to have a substantial impact on the results.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured or measurements were not reported.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The test organisms were not adequately described, and they were assumed to be from the same source as the definitive test.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health.	
	Metric 17: Outcome Assessment Methodology	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.	
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported.	
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Study Citation:	EAG Laboratories, (2018). Dibutyl phthalate: Medaka extended one generation reproduction test (final report).
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	10064186

Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	High	Sufficient data were provided to conduct an independent statistical analysis.
Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This evaluation is for a range-finder test.

Overall Quality Determination

Medium

Study Citation:	EAG Laboratories, (2018). Dibutyl phthalate: Medaka extended one generation reproduction test (final report).			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult			
Health Outcome:	Skin & Connective Tissue			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	10064186			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	Chemical was identified by name and CAS#	
	Metric 2: Test Substance Source	High	The test substance identity was analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity reported as 99.3%	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable	
	Metric 6: Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail	
	Metric 8: Consistency of Exposure Administration	High	exposures were administered consistently across study groups	
	Metric 9: Measurement of Test Substance Concentration	Medium	Exposure concentrations were measured using appropriate analytical technologies and methods, however measured concentrations were consistently lower than nominals	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source	
	Metric 14: Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest (page 37)	
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Study Citation:	EAG Laboratories, (2018). Dibutyl phthalate: Medaka extended one generation reproduction test (final report).			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult			
Health Outcome:	Skin & Connective Tissue			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	10064186			
Domain	Metric	Rating	Comments	
	Metric 18: Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions	
	Metric 20: Outcomes Unrelated to Exposure	High	there were no differences among groups	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical methods were adequately described	
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group	
	Metric 23: Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained	
Additional Comments:	This is for the anal fin papillae evaluation.			

Overall Quality Determination**High**

Study Citation:	EAG Laboratories, (2018). Dibutyl phthalate: Medaka extended one generation reproduction test (final report).		
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult		
Health Outcome:	Hepatic/Liver		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	10064186		
Domain	Metric	Rating	Comments
Domain 1: Test Substance			
	Metric 1: Test Substance Identity	High	The chemical was identified by name and CAS#.
	Metric 2: Test Substance Source	High	The test substance identity was analytically verified by the performing laboratory.
	Metric 3: Test Substance Purity	High	Chemical purity was reported as 99.3%.
Domain 2: Test Design			
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable.
	Metric 6: Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups.
Domain 3: Exposure Characterization			
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.
	Metric 9: Measurement of Test Substance Concentration	Medium	Exposure concentrations were measured using appropriate analytical technologies and methods, however measured concentrations were consistently lower than nominals.
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type.
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit.
Domain 4: Test Organism			
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14: Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions.
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest (pages 37-39).
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups..
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Study Citation:	EAG Laboratories, (2018). Dibutyl phthalate: Medaka extended one generation reproduction test (final report).		
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult		
Health Outcome:	Hepatic/Liver		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	10064186		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
Metric 20:	Outcomes Unrelated to Exposure	High	There were no differences among groups.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	High	Statistical methods were adequately described.
Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group (Appendix 8).
Metric 23:	Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.
Additional Comments: This is for the liver histopathology evaluation.			
Overall Quality Determination		High	

Study Citation:	EAG Laboratories, (2018). Dibutyl phthalate: Medaka extended one generation reproduction test (final report).		
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult		
Health Outcome:	Renal/Kidney		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	10064186		
Domain	Metric	Rating	Comments
Domain 1: Test Substance			
	Metric 1: Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2: Test Substance Source	High	The test substance identity was analytically verified by the performing laboratory.
	Metric 3: Test Substance Purity	High	Chemical purity reported as 99.3%
Domain 2: Test Design			
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable
	Metric 6: Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization			
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8: Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9: Measurement of Test Substance Concentration	Medium	Exposure concentrations were measured using appropriate analytical technologies and methods, however measured concentrations were consistently lower than nominals
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit
Domain 4: Test Organism			
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14: Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest (pages 37-39)
	Metric 18: Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
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Study Citation:	EAG Laboratories, (2018). Dibutyl phthalate: Medaka extended one generation reproduction test (final report).		
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult		
Health Outcome:	Renal/Kidney		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	10064186		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	High	Statistical methods were adequately described
Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group (Appendix 8)
Metric 23:	Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained
Additional Comments: This is for the kidney histopathology evaluation.			
Overall Quality Determination		High	

Study Citation:	EAG Laboratories, (2018). Dibutyl phthalate: Medaka extended one generation reproduction test (final report).		
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult		
Health Outcome:	Endocrine		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	10064186		
Domain	Metric	Rating	Comments
Domain 1: Test Substance			
	Metric 1: Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2: Test Substance Source	High	The test substance identity was analytically verified by the performing laboratory.
	Metric 3: Test Substance Purity	High	Chemical purity reported as 99.3%
Domain 2: Test Design			
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable
	Metric 6: Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization			
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8: Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9: Measurement of Test Substance Concentration	Medium	Exposure concentrations were measured using appropriate analytical technologies and methods, however measured concentrations were consistently lower than nominals
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit
Domain 4: Test Organism			
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14: Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest (pages 37-39)
	Metric 18: Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
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Study Citation:	EAG Laboratories, (2018). Dibutyl phthalate: Medaka extended one generation reproduction test (final report).		
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult		
Health Outcome:	Endocrine		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	10064186		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	High	Statistical methods were adequately described
Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group (Appendix 8)
Metric 23:	Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained
Additional Comments: This is for the thyroid gland histopathology evaluation.			
Overall Quality Determination		High	

Study Citation:	EAG Laboratories, (2018). Dibutyl phthalate: Medaka extended one generation reproduction test (final report).		
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult		
Health Outcome:	Mortality		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	10064186		
Domain	Metric	Rating	Comments
Domain 1: Test Substance			
	Metric 1: Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2: Test Substance Source	High	The test substance identity was analytically verified by the performing laboratory.
	Metric 3: Test Substance Purity	High	Chemical purity reported as 99.3%
Domain 2: Test Design			
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable
	Metric 6: Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization			
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8: Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9: Measurement of Test Substance Concentration	Medium	Exposure concentrations were measured using appropriate analytical technologies and methods, however measured concentrations were consistently lower than nominals
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit
Domain 4: Test Organism			
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14: Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18: Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
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Study Citation:	EAG Laboratories, (2018). Dibutyl phthalate: Medaka extended one generation reproduction test (final report).		
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult		
Health Outcome:	Mortality		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	10064186		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	High	Statistical methods were adequately described
Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
Metric 23:	Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained
Additional Comments: generational effects			
Overall Quality Determination		High	

Study Citation:	EAG Laboratories, (2018). Dibutyl phthalate: Medaka extended one generation reproduction test (final report).		
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult		
Health Outcome:	Reproductive/Teratogenic		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	10064186		
Domain	Metric	Rating	Comments
Domain 1: Test Substance			
	Metric 1: Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2: Test Substance Source	High	The test substance identity was analytically verified by the performing laboratory.
	Metric 3: Test Substance Purity	High	Chemical purity reported as 99.3%
Domain 2: Test Design			
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable
	Metric 6: Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization			
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8: Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9: Measurement of Test Substance Concentration	Medium	Exposure concentrations were measured using appropriate analytical technologies and methods, however measured concentrations were consistently lower than nominals
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit
Domain 4: Test Organism			
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14: Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18: Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
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Study Citation:	EAG Laboratories, (2018). Dibutyl phthalate: Medaka extended one generation reproduction test (final report).		
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult		
Health Outcome:	Reproductive/Teratogenic		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	10064186		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	High	Statistical methods were adequately described
Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
Metric 23:	Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained
Additional Comments: generational effects			
Overall Quality Determination		High	

Study Citation:	EAG Laboratories, (2018). Dibutyl phthalate: Medaka extended one generation reproduction test (final report).		
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult		
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Endocrine toxicity-Reproductive/Teratogenic		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	10064186		
Domain	Metric	Rating	Comments
Domain 1: Test Substance			
	Metric 1: Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2: Test Substance Source	High	The test substance identity was analytically verified by the performing laboratory.
	Metric 3: Test Substance Purity	High	Chemical purity reported as 99.3%
Domain 2: Test Design			
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable
	Metric 6: Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization			
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8: Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9: Measurement of Test Substance Concentration	Medium	Exposure concentrations were measured using appropriate analytical technologies and methods, however measured concentrations were consistently lower than nominals
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit
Domain 4: Test Organism			
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14: Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18: Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
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Study Citation:	EAG Laboratories, (2018). Dibutyl phthalate: Medaka extended one generation reproduction test (final report).		
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult		
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Endocrine toxicity-Reproductive/Teratogenic		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	10064186		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	High	Statistical methods were adequately described
Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
Metric 23:	Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained
Additional Comments: generational effects			

Overall Quality Determination**High**

Study Citation:	EAG Laboratories, (2018). Dibutyl phthalate: Medaka extended one generation reproduction test (final report).		
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult		
Health Outcome:	Development/Growth		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	10064186		
Domain	Metric	Rating	Comments
Domain 1: Test Substance			
	Metric 1: Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2: Test Substance Source	High	The test substance identity was analytically verified by the performing laboratory.
	Metric 3: Test Substance Purity	High	Chemical purity reported as 99.3%
Domain 2: Test Design			
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable
	Metric 6: Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization			
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8: Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9: Measurement of Test Substance Concentration	Medium	Exposure concentrations were measured using appropriate analytical technologies and methods, however measured concentrations were consistently lower than nominals
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit
Domain 4: Test Organism			
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14: Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18: Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
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Study Citation:	EAG Laboratories, (2018). Dibutyl phthalate: Medaka extended one generation reproduction test (final report).		
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult		
Health Outcome:	Development/Growth		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	10064186		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	High	Statistical methods were adequately described
Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
Metric 23:	Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained
Additional Comments: generational effects			
Overall Quality Determination		High	

Study Citation:	Patyna, P. J. (1999). Reproductive effects of phthalate esters in Japanese medaka (<i>Oryzias latipes</i>). Doctoral Dissertation:137.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult			
Health Outcome:	Renal/Kidney			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5489073; Linked HERO ID(s): 5489073, 680110			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
Metric 1:	Test Substance Identity	High	Nomenclature was reported in section 2.1. The structure was identified in a figure within the dissertation. CAS number was not reported.	
Metric 2:	Test Substance Source	High	GCMS was reported on the compound in section 2.1 and the lot number was provided. Source was reported as from Sigma Aldrich.	
Metric 3:	Test Substance Purity	High	A purity of 99% via GCMS was reported.	
Domain 2: Test Design				
Metric 4:	Negative Controls	High	The authors reported performing a control diet with ethanol as a carrier.	
Metric 5:	Negative Control Response	High	Control kidney histology is presented in figure 6.4.4 on page 136/158.	
Metric 6:	Randomized Allocation	Low	Allocation was not reported as random.	
Domain 3: Exposure Characterization				
Metric 7:	Experimental System/Test Media Preparation	High	The diet study with multiple generations of exposure was described in detail on page 46/158. The experimental systems are reported and water quality parameters (temp, Dissolved oxygen, pH) were observed in this flow-through system.	
Metric 8:	Consistency of Exposure Administration	High	Exposures were the same across treatments and control groups.	
Metric 9:	Measurement of Test Substance Concentration	High	The test compound in the diet was verified with GC-MS and was reported as final concentrations in the feed as well as the delivery dose for mg/kg/day as 12.2, 65, and 776 mg/kg/d DBP.	
Metric 10:	Exposure Duration and Frequency	High	The dietary treatments were administered after 14 days post hatch and till 180 DPH.	
Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Three exposure concentrations were used. The authors acknowledge on page 68/158 that these concentrations are all mostly likely "several orders of magnitude" above environmentally relevant concentrations.	
Metric 12:	Testing at or Below Solubility Limit	N/A	Dietary study, solubility not applicable.	
Domain 4: Test Organism				
Metric 13:	Test Organism Characteristics	High	The source was listed as from Carolina Biological Supply. These fish were bred to produce embryos used in the study.	
Metric 14:	Acclimatization and Pretreatment Conditions	High	The fish were acclimated for two weeks prior to the dietary study beginning. Test conditions were the same before and after the start of the study. (page 46/158).	
Metric 15:	Number of Organisms and Replicates per Group	Low	The treatment and control groups were conducted in duplicate.	
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Study Citation:	Patyna, P. J. (1999). Reproductive effects of phthalate esters in Japanese medaka (<i>Oryzias latipes</i>). Doctoral Dissertation:137.		
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult		
Health Outcome:	Renal/Kidney		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	5489073; Linked HERO ID(s): 5489073, 680110		
Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
Metric 16:	Adequacy of Test Conditions	High	Housing conditions for the flow through system with this diet study were appropriate for this bioassay.
Metric 17:	Outcome Assessment Methodology	High	Abnormal kidney pathology was noted for the highest dietary treatment and is presented on page 67/158 of the results and shown in figures 6.4.5 and 6.4.4. (pages 139,140/158)
Metric 18:	Consistency of Outcome Assessment	High	The assessment was conducted similarly across treatment and control groups.
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	High	There was no indication that reported differences were from environmental conditions or other confounding variables.
Metric 20:	Outcomes Unrelated to Exposure	Medium	No information to suggest differences among treatment and control groups attributed to animal health or attrition.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	N/A	The results are presented as pathology/histology operations among treatment concentrations. Traditional statistics were not applied.
Metric 22:	Reporting of Data	Low	Results were not quantitative. Abnormal kidney pathology was noted for the highest dietary treatment and is presented on page 67/158 of the results and shown in figures 6.4.5 and 6.4.4. (pages 139,140/158)
Metric 23:	Explanation of Unexpected Outcomes	High	No unexpected outcomes appear to have been encountered.
Additional Comments:	It was reported that the highest dose of DBP tested induced kidney toxicity, manifested by tubular degeneration and glomerular congestion; results were qualitative and the figures in the pdf were not clear to verify findings.		
Overall Quality Determination		Low	

Study Citation:	Patyna, P. J. (1999). Reproductive effects of phthalate esters in Japanese medaka (<i>Oryzias latipes</i>). Doctoral Dissertation:137.		
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult		
Health Outcome:	Development/Growth		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	5489073; Linked HERO ID(s): 5489073, 680110		
Domain	Metric	Rating	Comments
Domain 1: Test Substance			
	Metric 1: Test Substance Identity	High	Nomenclature was reported in section 2.1. The structure was identified in a figure within the dissertation. CAS number was not reported.
	Metric 2: Test Substance Source	High	GCMS was reported on the compound in section 2.1 and the lot number was provided. Source was reported as from Sigma Aldrich.
	Metric 3: Test Substance Purity	High	A purity of 99% via GCMS was reported.
Domain 2: Test Design			
	Metric 4: Negative Controls	High	The authors reported performing a control diet with ethanol as a carrier.
	Metric 5: Negative Control Response	High	Control growth/final weight at 180 days of feeding exposure within Table 6.1 on page 100/158.
	Metric 6: Randomized Allocation	Low	Allocation was not reported as random.
Domain 3: Exposure Characterization			
	Metric 7: Experimental System/Test Media Preparation	High	The diet study with multiple generations of exposure was described in detail on page 46/158. The experimental systems are reported and water quality parameters (temp, Dissolved oxygen, pH) were observed in this flow-through system.
	Metric 8: Consistency of Exposure Administration	High	Exposures were the same across treatments and control groups.
	Metric 9: Measurement of Test Substance Concentration	High	The test compound in the diet was verified with GC-MS and was reported as final concentrations in the feed as well as the delivery dose for mg/kg/day as 12.2, 65, and 776 mg/kg/d DBP.
	Metric 10: Exposure Duration and Frequency	High	The dietary treatments were administered after 14 days post hatch and till 180 DPH.
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Medium	Three exposure concentrations were used. The authors acknowledge on page 68/158 that these concentrations are all mostly likely "several orders of magnitude" above environmentally relevant concentrations.
	Metric 12: Testing at or Below Solubility Limit	N/A	Dietary study, solubility not applicable.
Domain 4: Test Organism			
	Metric 13: Test Organism Characteristics	High	The source was listed as from Carolina Biological Supply. These fish were bred to produce embryos used in the study.
	Metric 14: Acclimatization and Pretreatment Conditions	High	The fish were acclimated for two weeks prior to the dietary study beginning. Test conditions were the same before and after the start of the study. (page 46/158).
	Metric 15: Number of Organisms and Replicates per Group	Low	The treatment and control groups were conducted in duplicate.
Domain 5: Outcome Assessment			

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Study Citation:	Patyna, P. J. (1999). Reproductive effects of phthalate esters in Japanese medaka (<i>Oryzias latipes</i>). Doctoral Dissertation:137.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5489073; Linked HERO ID(s): 5489073, 680110			
Domain	Metric	Rating	Comments	
	Metric 16:	Adequacy of Test Conditions	High	Housing conditions for the flow through system with this diet study were appropriate for this bioassay.
	Metric 17:	Outcome Assessment Methodology	High	Outcome assessment methodology reported the intended outcomes (growth) appropriately for these dietary exposures.
	Metric 18:	Consistency of Outcome Assessment	High	The assessments were conducted similarly across treatment and control groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There was no indication that reported differences were from environmental conditions or other confounding variables.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	No information to suggest differences among treatment and control groups attributed to animal health or attrition.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately. Authors did a poor job reporting the output from statistical tests and lack significance results (p values, DF, F scores).
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	No unexpected outcomes appear to have been encountered.
Additional Comments:	This evaluation form is relevant to growth endpoints- total body weight, gonad weight and gonadal somatic index results (Table 6.1) for F1 generation.			

Overall Quality Determination**High**

Study Citation:	Patyna, P. J. (1999). Reproductive effects of phthalate esters in Japanese medaka (<i>Oryzias latipes</i>). Doctoral Dissertation:137.		
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult		
Health Outcome:	Development/Growth		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	5489073; Linked HERO ID(s): 5489073, 680110		
Domain	Metric	Rating	Comments
Domain 1: Test Substance			
	Metric 1: Test Substance Identity	High	Nomenclature was reported in section 2.1. The structure was identified in a figure within the dissertation. CAS number was not reported.
	Metric 2: Test Substance Source	High	GCMS was reported on the compound in section 2.1 and the lot number was provided. Source was reported as from Sigma Aldrich.
	Metric 3: Test Substance Purity	High	A purity of 99% via GCMS was reported.
Domain 2: Test Design			
	Metric 4: Negative Controls	High	The authors reported performing a control diet with ethanol as a carrier.
	Metric 5: Negative Control Response	High	Control growth/final weight at 180 days of feeding exposure within Table 6.1 on page 100/158.
	Metric 6: Randomized Allocation	Low	Allocation was not reported as random.
Domain 3: Exposure Characterization			
	Metric 7: Experimental System/Test Media Preparation	High	The diet study with multiple generations of exposure was described in detail on page 46/158. The experimental systems are reported and water quality parameters (temp, Dissolved oxygen, pH) were observed in this flow-through system.
	Metric 8: Consistency of Exposure	High	Exposures were the same across treatments and control groups.
	Metric 9: Administration Measurement of Test Substance Concentration	High	The test compound in the diet was verified with GC-MS and was reported as final concentrations in the feed as well as the delivery dose for mg/kg/day as 12.2, 65, and 776 mg/kg/d DBP.
	Metric 10: Exposure Duration and Frequency	High	The dietary treatments were administered after 14 days post hatch and till 180 DPH.
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Three exposure concentrations were used. The authors acknowledge on page 68/158 that these concentrations are all mostly likely "several orders of magnitude" above environmentally relevant concentrations.
	Metric 12: Testing at or Below Solubility Limit	N/A	Dietary study, solubility not applicable.
Domain 4: Test Organism			
	Metric 13: Test Organism Characteristics	High	The source was listed as from Carolina Biological Supply. These fish were bred to produce embryos used in the study.
	Metric 14: Acclimatization and Pretreatment Conditions	High	The fish were acclimated for two weeks prior to the dietary study beginning. Test conditions were the same before and after the start of the study. (page 46/158).
	Metric 15: Number of Organisms and Replicates per Group	Low	The treatment and control groups were conducted in duplicate.
Domain 5: Outcome Assessment			

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Study Citation:	Patyna, P. J. (1999). Reproductive effects of phthalate esters in Japanese medaka (<i>Oryzias latipes</i>). Doctoral Dissertation:137.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5489073; Linked HERO ID(s): 5489073, 680110			
Domain	Metric	Rating	Comments	
	Metric 16:	Adequacy of Test Conditions	High	Housing conditions for the flow through system with this diet study were appropriate for this bioassay.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome(s) of interest.
	Metric 18:	Consistency of Outcome Assessment	High	The assessments were conducted similarly across treatment and control groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There was no indication that reported differences were from environmental conditions or other confounding variables.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	No information to suggest differences among treatment and control groups attributed to animal health or attrition.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately. Authors did a poor job reporting the output from statistical tests and lack significance results (p values, DF, F scores).
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group (Table 6.1).
	Metric 23:	Explanation of Unexpected Outcomes	High	No unexpected outcomes appear to have been encountered.
Additional Comments:	No differences in weight within the F2 generation were observed from the control to the 12 and 65 mg/kg/d treatment groups.			

Overall Quality Determination**High**

Study Citation:	Patyna, P. J. (1999). Reproductive effects of phthalate esters in Japanese medaka (<i>Oryzias latipes</i>). Doctoral Dissertation:137.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5489073; Linked HERO ID(s): 5489073, 680110			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Nomenclature was reported in section 2.1. The structure was identified in a figure within the dissertation. CAS number was not reported.
	Metric 2:	Test Substance Source	High	GCMS was reported on the compound in section 2.1 and the lot number was provided. Source was reported as from Sigma Aldrich.
	Metric 3:	Test Substance Purity	High	A purity of 99% via GCMS was reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	The authors reported performing a control diet with ethanol as a carrier.
	Metric 5:	Negative Control Response	High	Control Egg production and GSI values at 180 days of feeding exposure are presented within Table 6.1, 6.2 on page 100,101/158. Page 140/158 has a figure with control egg output.
	Metric 6:	Randomized Allocation	Low	Allocation was not reported as random.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The diet study with multiple generations of exposure was described in detail on page 46/158. The experimental systems are reported and water quality parameters (temp, Dissolved oxygen, pH) were observed in this flow-through system.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were the same across treatments and control groups.
	Metric 9:	Measurement of Test Substance Concentration	High	The test compound in the diet was verified with GC-MS and was reported as final concentrations in the feed as well as the delivery dose for mg/kg/day as 12.2, 65, and 776 mg/kg/d DBP.
	Metric 10:	Exposure Duration and Frequency	High	The dietary treatments were administered after 14 days post hatch and till 180 DPH.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Three exposure concentrations were used. The authors acknowledge on page 68/158 that these concentrations are all mostly likely "several orders of magnitude" above environmentally relevant concentrations.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Dietary study, solubility not applicable.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The source was listed as from Carolina Biological Supply. These fish were bred to produce embryos used in the study.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The fish were acclimated for two weeks prior to the dietary study beginning. Test conditions were the same before and after the start of the study. (page 46/158).
	Metric 15:	Number of Organisms and Replicates per Group	Low	The treatment and control groups were conducted in duplicate.
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Study Citation:	Patyna, P. J. (1999). Reproductive effects of phthalate esters in Japanese medaka (<i>Oryzias latipes</i>). Doctoral Dissertation:137.		
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult		
Health Outcome:	Reproductive/Teratogenic		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	5489073; Linked HERO ID(s): 5489073, 680110		
Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Housing conditions for the flow through system with this diet study were appropriate for this bioassay.
	Metric 17: Outcome Assessment Methodology	High	Outcome assessment methodology reported the intended outcomes (GSI, Reproductive output) appropriately for these dietary exposures. figure 6.7 on page 141/158 reports eggs per female with notations for significance from control (for the F1 generation).
	Metric 18: Consistency of Outcome Assessment	High	The assessment was conducted similarly across treatment and control groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There was no indication that reported differences were from environmental conditions or other confounding variables.
	Metric 20: Outcomes Unrelated to Exposure	Medium	No information to suggest differences among treatment and control groups attributed to animal health or attrition.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Statistical analysis was performed but not described adequately. Authors did a poor job reporting the output from statistical tests and lack significance results (p values, DF, F scores).
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	No unexpected outcomes appear to have been encountered.
Additional Comments:	This evaluation form is relevant to reproductive output endpoints - total egg production and egg production per female for F1 generation; Results given in Table 6.2.		
Overall Quality Determination		High	

Study Citation:	Patyna, P. J. (1999). Reproductive effects of phthalate esters in Japanese medaka (<i>Oryzias latipes</i>). Doctoral Dissertation:137.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5489073; Linked HERO ID(s): 5489073, 680110			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
Metric 1:	Test Substance Identity	High	Nomenclature was reported in section 2.1. The structure was identified in a figure within the dissertation. CAS number was not reported.	
Metric 2:	Test Substance Source	High	GCMS was reported on the compound in section 2.1 and the lot number was provided. Source was reported as from Sigma Aldrich.	
Metric 3:	Test Substance Purity	High	A purity of 99% via GCMS was reported.	
Domain 2: Test Design				
Metric 4:	Negative Controls	High	The authors reported performing a control diet with ethanol as a carrier.	
Metric 5:	Negative Control Response	High	Control Egg production and GSI values at 180 days of feeding exposure are presented within Table 6.1, 6.2 on page 100,101/158. Page 140/158 has a figure with control egg output.	
Metric 6:	Randomized Allocation	Low	Allocation was not reported as random.	
Domain 3: Exposure Characterization				
Metric 7:	Experimental System/Test Media Preparation	High	The diet study with multiple generations of exposure was described in detail on page 46/158. The experimental systems are reported and water quality parameters (temp, Dissolved oxygen, pH) were observed in this flow-through system.	
Metric 8:	Consistency of Exposure Administration	High	Exposures were the same across treatments and control groups.	
Metric 9:	Measurement of Test Substance Concentration	High	The test compound in the diet was verified with GC-MS and was reported as final concentrations in the feed as well as the delivery dose for mg/kg/day as 12.2, 65, and 776 mg/kg/d DBP.	
Metric 10:	Exposure Duration and Frequency	High	The dietary treatments were administered after 14 days post hatch and till 180 DPH.	
Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Three exposure concentrations were used. The authors acknowledge on page 68/158 that these concentrations are all mostly likely "several orders of magnitude" above environmentally relevant concentrations.	
Metric 12:	Testing at or Below Solubility Limit	N/A	Dietary study, solubility not applicable.	
Domain 4: Test Organism				
Metric 13:	Test Organism Characteristics	High	The source was listed as from Carolina Biological Supply. These fish were bred to produce embryos used in the study.	
Metric 14:	Acclimatization and Pretreatment Conditions	High	The fish were acclimated for two weeks prior to the dietary study beginning. Test conditions were the same before and after the start of the study. (page 46/158).	
Metric 15:	Number of Organisms and Replicates per Group	Low	The treatment and control groups were conducted in duplicate.	
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Study Citation:	Patyna, P. J. (1999). Reproductive effects of phthalate esters in Japanese medaka (<i>Oryzias latipes</i>). Doctoral Dissertation:137.		
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult		
Health Outcome:	Reproductive/Teratogenic		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	5489073; Linked HERO ID(s): 5489073, 680110		
Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Housing conditions for the flow through system with this diet study were appropriate for this bioassay.
	Metric 17: Outcome Assessment Methodology	High	Outcome assessment methodology reported the intended outcomes (Reproductive output- Total egg production and Egg production per female; Table 6.2) appropriately for these dietary exposures. figure 6.6 on page 140/158 reports eggs per female with notations for significance from control.
	Metric 18: Consistency of Outcome Assessment	High	The assessment was conducted similarly across treatment and control groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There was no indication that reported differences were from environmental conditions or other confounding variables.
	Metric 20: Outcomes Unrelated to Exposure	Medium	No information to suggest differences among treatment and control groups attributed to animal health or attrition.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Statistical analysis was performed but not described adequately. Authors did a poor job reporting the output from statistical tests and lack significance results (p values, DF, F scores).
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	No unexpected outcomes appear to have been encountered.
Additional Comments:	This evaluation form is relevant to reproductive output endpoints - total egg production and egg production per female for F0 generation; Results given in Table 6.2		
Overall Quality Determination		High	

Study Citation:	Patyna, P. J. (1999). Reproductive effects of phthalate esters in Japanese medaka (<i>Oryzias latipes</i>). Doctoral Dissertation:137.		
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult		
Health Outcome:	Reproductive/Teratogenic		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	5489073; Linked HERO ID(s): 5489073, 680110		
Domain	Metric	Rating	Comments
Domain 1: Test Substance			
Metric 1:	Test Substance Identity	High	Nomenclature was reported in section 2.1. The structure was identified in a figure within the dissertation. CAS number was not reported.
Metric 2:	Test Substance Source	High	GCMS was reported on the compound in section 2.1 and the lot number was provided. Source was reported as from Sigma Aldrich.
Metric 3:	Test Substance Purity	High	A purity of 99% via GCMS was reported.
Domain 2: Test Design			
Metric 4:	Negative Controls	High	The authors reported performing a control diet with ethanol as a carrier.
Metric 5:	Negative Control Response	High	Control Egg production and GSI values at 180 days of feeding exposure are presented within Table 6.1, 6.2 on page 100,101/158. Page 140/158 has a figure with control egg output.
Metric 6:	Randomized Allocation	Low	Allocation was not reported as random.
Domain 3: Exposure Characterization			
Metric 7:	Experimental System/Test Media Preparation	High	The diet study with multiple generations of exposure was described in detail on page 46/158. The experimental systems are reported and water quality parameters (temp, Dissolved oxygen, pH) were observed in this flow-through system.
Metric 8:	Consistency of Exposure Administration	High	Exposures were the same across treatments and control groups.
Metric 9:	Measurement of Test Substance Concentration	High	The test compound in the diet was verified with GC-MS and was reported as final concentrations in the feed as well as the delivery dose for mg/kg/day as 12.2, 65, and 776 mg/kg/d DBP.
Metric 10:	Exposure Duration and Frequency	High	The dietary treatments were administered after 14 days post hatch and till 180 DPH.
Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	Two exposure concentrations were used. The previous highest feeding group (776 mg/kg/d) did not produce progeny for the F2 generation. The authors acknowledge on page 68/158 that these concentrations are all mostly likely "several orders of magnitude" above environmentally relevant concentrations.
Metric 12:	Testing at or Below Solubility Limit	N/A	Dietary study, solubility not applicable.
Domain 4: Test Organism			
Metric 13:	Test Organism Characteristics	High	The source was listed as from Carolina Biological Supply. These fish were bred to produce embryos used in the study.
Metric 14:	Acclimatization and Pretreatment Conditions	High	The fish were acclimated for two weeks prior to the dietary study beginning. Test conditions were the same before and after the start of the study. (page 46/158).
Metric 15:	Number of Organisms and Replicates per Group	Low	The treatment and control groups were conducted in duplicate.
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Study Citation:	Patyna, P. J. (1999). Reproductive effects of phthalate esters in Japanese medaka (<i>Oryzias latipes</i>). Doctoral Dissertation:137.		
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult		
Health Outcome:	Reproductive/Teratogenic		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	5489073; Linked HERO ID(s): 5489073, 680110		
Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Housing conditions for the flow through system with this diet study were appropriate for this bioassay.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome(s) of interest.
	Metric 18: Consistency of Outcome Assessment	High	The assessment was conducted similarly across treatment and control groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There was no indication that reported differences were from environmental conditions or other confounding variables.
	Metric 20: Outcomes Unrelated to Exposure	Medium	No information to suggest differences among treatment and control groups attributed to animal health or attrition.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Statistical analysis was performed but not described adequately. Authors did a poor job reporting the output from statistical tests and lack significance results (p values, DF, F scores).
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23: Explanation of Unexpected Outcomes	High	No unexpected outcomes appear to have been encountered.
Additional Comments:	There were no significant differences in reproduction for the F2 generation between control, 12, and 65 mg/kg. The previous highest feeding group (776 mg/kg/d) did not produce progeny for the F2 generation.		
Overall Quality Determination		High	

Study Citation:	Patyna, P. J. (1999). Reproductive effects of phthalate esters in Japanese medaka (<i>Oryzias latipes</i>). Doctoral Dissertation:137.		
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult		
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	5489073; Linked HERO ID(s): 5489073, 680110		
Domain	Metric	Rating	Comments
Domain 1: Test Substance			
	Metric 1: Test Substance Identity	High	Nomenclature was reported in section 2.1. The structure was identified in a figure within the dissertation.
	Metric 2: Test Substance Source	High	GCMS was reported on the compound in section 2.1 and the lot number was provided. Source was reported as from Sigma Aldrich.
	Metric 3: Test Substance Purity	High	A purity of 99% via GCMS was reported.
Domain 2: Test Design			
	Metric 4: Negative Controls	High	The authors reported performing a control diet with ethanol as a carrier.
	Metric 5: Negative Control Response	High	Control VTG is reported in results section on page 65/158 and figure 6.1 on page 128/158.
	Metric 6: Randomized Allocation	Low	Allocation was not reported as random.
Domain 3: Exposure Characterization			
	Metric 7: Experimental System/Test Media Preparation	High	The diet study with multiple generations of exposure was described in detail on page 46/158. The experimental systems are reported and water quality parameters (temp, Dissolved oxygen, pH) were observed in this flow-through system.
	Metric 8: Consistency of Exposure	High	Exposures were the same across treatments and control groups.
	Metric 9: Administration Measurement of Test Substance Concentration	High	The test compound in the diet was verified with GC-MS and was reported as final concentrations in the feed as well as the delivery dose for mg/kg/day as 12.2, 65, and 776 mg/kg/d DBP.
	Metric 10: Exposure Duration and Frequency	High	The dietary treatments were administered after 14 days post hatch and till 180 DPH.
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Three exposure concentrations were used. The authors acknowledge on page 68/158 that these concentrations are all mostly likely "several orders of magnitude" above environmentally relevant concentrations.
	Metric 12: Testing at or Below Solubility Limit	N/A	Dietary study, solubility not applicable.
Domain 4: Test Organism			
	Metric 13: Test Organism Characteristics	High	The source was listed as from Carolina Biological Supply. These fish were bred to produce embryos used in the study.
	Metric 14: Acclimatization and Pretreatment Conditions	High	The fish were acclimated for two weeks prior to the dietary study beginning. Test conditions were the same before and after the start of the study. (page 46/158).
	Metric 15: Number of Organisms and Replicates per Group	Low	The treatment and control groups were conducted in duplicate.
Domain 5: Outcome Assessment			

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Study Citation:	Patyna, P. J. (1999). Reproductive effects of phthalate esters in Japanese medaka (<i>Oryzias latipes</i>). Doctoral Dissertation:137.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5489073; Linked HERO ID(s): 5489073, 680110			
Domain	Metric	Rating	Comments	
	Metric 16: Adequacy of Test Conditions	High	Housing conditions for the flow through system with this diet study were appropriate for this bioassay.	
	Metric 17: Outcome Assessment Methodology	Medium	Methods for western blot analysis for vitellogenin were provided and this method provided qualitative data.	
	Metric 18: Consistency of Outcome Assessment	High	The assessment was conducted similarly across treatment and control groups.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	There was no indication that reported differences were from environmental conditions or other confounding variables.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	No information to suggest differences among treatment and control groups attributed to animal health or attrition.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	N/A	VTG was not quantified but rather reported for its presence/absence (Western Blots) in males exposed to dietary concentrations of DBP.	
	Metric 22: Reporting of Data	Low	Results were not quantitative. Western blot figures were not clear to interpret results but were described in the text.	
	Metric 23: Explanation of Unexpected Outcomes	Low	Insufficient information was provided to determine if excessive variability occurred.	
Additional Comments:	No VTG was detected for DBP treatment group in F0 males.			

Overall Quality Determination**Low**

Study Citation:	Patyna, P. J. (1999). Reproductive effects of phthalate esters in Japanese medaka (<i>Oryzias latipes</i>). Doctoral Dissertation:137.		
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult		
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	5489073; Linked HERO ID(s): 5489073, 680110		
Domain	Metric	Rating	Comments
Domain 1: Test Substance			
	Metric 1: Test Substance Identity	High	Nomenclature was reported in section 2.1. The structure was identified in a figure within the dissertation.
	Metric 2: Test Substance Source	High	GCMS was reported on the compound in section 2.1 and the lot number was provided. Source was reported as from Sigma Aldrich.
	Metric 3: Test Substance Purity	High	A purity of 99% via GCMS was reported.
Domain 2: Test Design			
	Metric 4: Negative Controls	High	The authors reported performing a control diet with ethanol as a carrier.
	Metric 5: Negative Control Response	Low	Control VTG was not reported for F1 generation. The representative western blot figures provided were not clear to interpret control data.
	Metric 6: Randomized Allocation	Low	Allocation was not reported as random.
Domain 3: Exposure Characterization			
	Metric 7: Experimental System/Test Media Preparation	High	The diet study with multiple generations of exposure was described in detail on page 46/158. The experimental systems are reported and water quality parameters (temp, Dissolved oxygen, pH) were observed in this flow-through system.
	Metric 8: Consistency of Exposure	High	Exposures were the same across treatments and control groups.
	Metric 9: Administration Measurement of Test Substance Concentration	High	The test compound in the diet was verified with GC-MS and was reported as final concentrations in the feed as well as the delivery dose for mg/kg/day as 12.2, 65, and 776 mg/kg/d DBP.
	Metric 10: Exposure Duration and Frequency	High	The dietary treatments were administered after 14 days post hatch and till 180 DPH.
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Three exposure concentrations were used. The authors acknowledge on page 68/158 that these concentrations are all mostly likely "several orders of magnitude" above environmentally relevant concentrations.
	Metric 12: Testing at or Below Solubility Limit	N/A	Dietary study, solubility not applicable.
Domain 4: Test Organism			
	Metric 13: Test Organism Characteristics	High	The source was listed as from Carolina Biological Supply. These fish were bred to produce embryos used in the study.
	Metric 14: Acclimatization and Pretreatment Conditions	High	The fish were acclimated for two weeks prior to the dietary study beginning. Test conditions were the same before and after the start of the study. (page 46/158).
	Metric 15: Number of Organisms and Replicates per Group	Low	The treatment and control groups were conducted in duplicate.
Domain 5: Outcome Assessment			

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Study Citation:	Patyna, P. J. (1999). Reproductive effects of phthalate esters in Japanese medaka (<i>Oryzias latipes</i>). Doctoral Dissertation:137.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5489073; Linked HERO ID(s): 5489073, 680110			
Domain	Metric	Rating	Comments	
	Metric 16: Adequacy of Test Conditions	High	Housing conditions for the flow through system with this diet study were appropriate for this bioassay.	
	Metric 17: Outcome Assessment Methodology	Medium	Methods for western blot analysis for vitellogenin were provided and this method provided qualitative data.	
	Metric 18: Consistency of Outcome Assessment	High	The assessment was conducted similarly across treatment and control groups.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	There was no indication that reported differences were from environmental conditions or other confounding variables.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	No information to suggest differences among treatment and control groups attributed to animal health or attrition.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	N/A	VTG was not quantified but rather reported for its presence/absence (Western Blots) in males exposed to dietary concentrations of DBP.	
	Metric 22: Reporting of Data	Low	Results were not quantitative. Representative western blot analysis figures provided in the dissertation were not clear to interpret results that were described in the text.	
	Metric 23: Explanation of Unexpected Outcomes	Low	Insufficient information was provided to determine if excessive variability or unexpected outcomes occurred.	
Additional Comments:	VTG was detected for DBP treatment group in males at the highest treatment concentration (776 mg/kg/d). (Page 66/158)			
Overall Quality Determination		Low		

Study Citation:	Patyna, P. J. (1999). Reproductive effects of phthalate esters in Japanese medaka (<i>Oryzias latipes</i>). Doctoral Dissertation:137.		
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult		
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	5489073; Linked HERO ID(s): 5489073, 680110		
Domain	Metric	Rating	Comments
Domain 1: Test Substance			
	Metric 1: Test Substance Identity	High	Nomenclature was reported in section 2.1. The structure was identified in a figure within the dissertation.
	Metric 2: Test Substance Source	High	GCMS was reported on the compound in section 2.1 and the lot number was provided. Source was reported as from Sigma Aldrich.
	Metric 3: Test Substance Purity	High	A purity of 99% via GCMS was reported.
Domain 2: Test Design			
	Metric 4: Negative Controls	High	The authors reported performing a control diet with ethanol as a carrier.
	Metric 5: Negative Control Response	Low	Control VTG was not reported for F2 generation. The representative western blot figures provided were not clear to interpret control data.
	Metric 6: Randomized Allocation	Low	Allocation was not reported as random.
Domain 3: Exposure Characterization			
	Metric 7: Experimental System/Test Media Preparation	High	The diet study with multiple generations of exposure was described in detail on page 46/158. The experimental systems are reported and water quality parameters (temp, Dissolved oxygen, pH) were observed in this flow-through system.
	Metric 8: Consistency of Exposure	High	Exposures were the same across treatments and control groups.
	Metric 9: Administration Measurement of Test Substance Concentration	High	The test compound in the diet was verified with GC-MS and was reported as final concentrations in the feed as well as the delivery dose for mg/kg/day as 12.2, 65, and 776 mg/kg/d DBP.
	Metric 10: Exposure Duration and Frequency	High	The dietary treatments were administered after 14 days post hatch and till 180 DPH.
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Three exposure concentrations were used. The authors acknowledge on page 68/158 that these concentrations are all mostly likely "several orders of magnitude" above environmentally relevant concentrations.
	Metric 12: Testing at or Below Solubility Limit	N/A	Dietary study, solubility not applicable.
Domain 4: Test Organism			
	Metric 13: Test Organism Characteristics	High	The source was listed as from Carolina Biological Supply. These fish were bred to produce embryos used in the study.
	Metric 14: Acclimatization and Pretreatment Conditions	High	The fish were acclimated for two weeks prior to the dietary study beginning. Test conditions were the same before and after the start of the study. (page 46/158).
	Metric 15: Number of Organisms and Replicates per Group	Low	The treatment and control groups were conducted in duplicate.
Domain 5: Outcome Assessment			

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Study Citation:	Patyna, P. J. (1999). Reproductive effects of phthalate esters in Japanese medaka (<i>Oryzias latipes</i>). Doctoral Dissertation:137.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5489073; Linked HERO ID(s): 5489073, 680110			
Domain	Metric	Rating	Comments	
	Metric 16: Adequacy of Test Conditions	High	Housing conditions for the flow through system with this diet study were appropriate for this bioassay.	
	Metric 17: Outcome Assessment Methodology	Medium	Methods for western blot analysis for vitellogenin were provided and this method yielded qualitative data. The highest treatment group from previous generations was not present as the F1 were not able to reproduce.	
	Metric 18: Consistency of Outcome Assessment	High	The assessment was conducted similarly across treatment and control groups.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	There was no indication that reported differences were from environmental conditions or other confounding variables.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	No information to suggest differences among treatment and control groups attributed to animal health or attrition.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	N/A	VTG was not quantified but rather reported for its presence/absence (Western Blots) in males exposed to dietary concentrations of DBP.	
	Metric 22: Reporting of Data	Low	Results were not quantitative. Figures in the dissertation were not clear to interpret results but were described in the text.	
	Metric 23: Explanation of Unexpected Outcomes	Low	Insufficient information was provided to determine if excessive variability occurred.	
Additional Comments:	Vitellogenin was not quantified but rather reported for its presence/absence (Western Blots) in males and females exposed to dietary concentrations of DBP. The highest treatment group from previous generations was not present as the F1 were not able to reproduce. Control VTG was not reported for F2 generation.			
Overall Quality Determination		Low		

Study Citation:	Patyna, P. J. (1999). Reproductive effects of phthalate esters in Japanese medaka (<i>Oryzias latipes</i>). Doctoral Dissertation:137.		
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult		
Health Outcome:	Development/Growth		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	5489073; Linked HERO ID(s): 5489073, 680110		
Domain	Metric	Rating	Comments
Domain 1: Test Substance			
	Metric 1: Test Substance Identity	High	Nomenclature was reported in section 2.1. The structure was identified in a figure within the dissertation.
	Metric 2: Test Substance Source	High	GCMS was reported on the compound in section 2.1 and the lot number was provided. Source was reported as from Sigma Aldrich.
	Metric 3: Test Substance Purity	High	A purity of 99% via GCMS was reported.
Domain 2: Test Design			
	Metric 4: Negative Controls	High	The authors reported performing a control diet with ethanol as a carrier.
	Metric 5: Negative Control Response	High	Control growth/final weight at 180 days of feeding exposure within Table 6.1 on page 100/158.
	Metric 6: Randomized Allocation	Low	Allocation was not reported as random.
Domain 3: Exposure Characterization			
	Metric 7: Experimental System/Test Media Preparation	High	The diet study with multiple generations of exposure was described in detail on page 46/158. The experimental systems are reported and water quality parameters (temp, Dissolved oxygen, pH) were observed in this flow-through system.
	Metric 8: Consistency of Exposure	High	Exposures were the same across treatments and control groups.
	Metric 9: Administration Measurement of Test Substance Concentration	High	The test compound in the diet was verified with GC-MS and was reported as final concentrations in the feed as well as the delivery dose for mg/kg/day as 12.2, 65, and 776 mg/kg/d DBP.
	Metric 10: Exposure Duration and Frequency	High	The dietary treatments were administered after 14 days post hatch and till 180 DPH.
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Low	Three exposure concentrations were used. The authors acknowledge on page 68/158 that these concentrations are all mostly likely "several orders of magnitude" above environmentally relevant concentrations.
	Metric 12: Testing at or Below Solubility Limit	N/A	Dietary study, solubility not applicable.
Domain 4: Test Organism			
	Metric 13: Test Organism Characteristics	High	The source was listed as from Carolina Biological Supply. These fish were bred to produce embryos used in the study.
	Metric 14: Acclimatization and Pretreatment Conditions	High	The fish were acclimated for two weeks prior to the dietary study beginning. Test conditions were the same before and after the start of the study. (page 46/158).
	Metric 15: Number of Organisms and Replicates per Group	Low	The treatment and control groups were conducted in duplicate and 10 animals per treatment were used.
Domain 5: Outcome Assessment			

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Study Citation:	Patyna, P. J. (1999). Reproductive effects of phthalate esters in Japanese medaka (<i>Oryzias latipes</i>). Doctoral Dissertation:137.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5489073; Linked HERO ID(s): 5489073, 680110			
Domain	Metric	Rating	Comments	
	Metric 16:	Adequacy of Test Conditions	High	Housing conditions for the flow through system with this diet study were appropriate for this bioassay.
	Metric 17:	Outcome Assessment Methodology	High	Outcome assessment methodology reported the intended outcomes (growth) appropriately for these dietary exposures.
	Metric 18:	Consistency of Outcome Assessment	High	The assessments were conducted similarly across treatment and control groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There was no indication that reported differences were from environmental conditions or other confounding variables.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	No information to suggest differences among treatment and control groups attributed to animal health or attrition.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Authors did a poor job reporting the output from statistical tests and lack significance results (p values, DF, F scores).
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group. Mean and standard deviation are given for total body weight, gonad weight and gonadal somatic index in Table 6.1
	Metric 23:	Explanation of Unexpected Outcomes	High	No unexpected outcomes appear to have been encountered.
Additional Comments:	This evaluation form is relevant to growth endpoints- total body weight, gonad weight and gonadal somatic index results (Table 6.1) for F0 generation.			

Overall Quality Determination**High**

Study Citation:	Patyna, P. J. (1999). Reproductive effects of phthalate esters in Japanese medaka (<i>Oryzias latipes</i>). Doctoral Dissertation:137.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5489073; Linked HERO ID(s): 5489073, 680110			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
Metric 1:	Test Substance Identity	High	Nomenclature was reported in section 2.1. The structure was identified in a figure within the dissertation. The CAS number was not reported.	
Metric 2:	Test Substance Source	High	GCMS was reported on the compound in section 2.1 and the lot number was provided. The source was reported as from Sigma Aldrich.	
Metric 3:	Test Substance Purity	High	A purity of 99% via GCMS was reported.	
Domain 2: Test Design				
Metric 4:	Negative Controls	High	The authors reported performing control and solvent controls.	
Metric 5:	Negative Control Response	Low	No control responses for development are reported for control groups.	
Metric 6:	Randomized Allocation	Low	Allocation was not random.	
Domain 3: Exposure Characterization				
Metric 7:	Experimental System/Test Media Preparation	High	A static non-renewal 10-day exposure was conducted. At 10 days replacement water was free of the compound. Authors used teflon rearing containers to reduce the leeching of test compound to the containers.	
Metric 8:	Consistency of Exposure Administration	High	Exposures were the same across treatments and control groups.	
Metric 9:	Measurement of Test Substance Concentration	Medium	Measured concentrations are reported in table 4.2/4.3, but developmental observations were not reported for each concentration.	
Metric 10:	Exposure Duration and Frequency	High	The exposure (10 days) was reported to cover the normal period of embryo development for this species. Modal hatch was recorded between 11-14 days.	
Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	The incidence of lesions are reported below and above the LC50 for DBP, so it is not able to relate to a dose-response gradient.	
Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit.	
Domain 4: Test Organism				
Metric 13:	Test Organism Characteristics	High	The source was listed as from Carolina Biological Supply. These fish were bred to produce embryos used in the study.	
Metric 14:	Acclimatization and Pretreatment Conditions	High	Embryo media and test water conditions were reported on page 45/158.	
Metric 15:	Number of Organisms and Replicates per Group	Low	The authors report the exposures were done in triplicate, however, sample numbers reported in the results do not reflect replicated sample numbers (Table 4.2-4.3) pages 90,91/158.	
Domain 5: Outcome Assessment				
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Study Citation:	Patyna, P. J. (1999). Reproductive effects of phthalate esters in Japanese medaka (<i>Oryzias latipes</i>). Doctoral Dissertation:137.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias latipes</i> ; Adult			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5489073; Linked HERO ID(s): 5489073, 680110			
Domain	Metric	Rating	Comments	
	Metric 16:	Adequacy of Test Conditions	Medium	Embryo housing was detailed for the 10-day exposure, however, authors did not report water quality results to ensure adequate environmental conditions during embryogenesis.
	Metric 17:	Outcome Assessment Methodology	Low	The authors described morphological lesions from DBP exposure but failed to present data on concentration specific incidences.
	Metric 18:	Consistency of Outcome Assessment	Low	It is not clear how developmental morphology was scored at each concentration and control for the lesions listed in table 4.5 (page 94/158). Data are not presented.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There was no indication that reported differences were from environmental conditions or other confounding variables.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information to suggest differences among treatment and control groups attributed to animal health or attrition.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Uninformative	The authors report the presence of lesions with no data on quantifying at different exposure concentrations.
	Metric 22:	Reporting of Data	Low	There is not enough data on incidence of lesions to relate to a dose-response gradient.
	Metric 23:	Explanation of Unexpected Outcomes	Low	Not enough data are presented for lesions at specific concentrations to determine unexpected outcomes.
Additional Comments:	This evaluation form is relevant to the morphological lesions results given on page 94/158.			

Overall Quality Determination**Uninformative**

Study Citation:	Chen, X., Xu, S., Tan, T., Lee, S. T., Cheng, S. H., Lee, F., F.W., Xu, L., S.J., Ho, K. C. (2014). Toxicity and estrogenic endocrine disrupting activity of phthalates and their mixtures. International Journal of Environmental Research and Public Health 11(3):3156-3168.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias melastigma</i> ; ChgH-EGFP; Larvae			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Receptor binding/ regulation of receptor activity-Endocrine toxicity-Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2298079			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The CAS numbers and structures for BBP, DBP, DEHP, DIDP, and DINP were reported.	
	Metric 2: Test Substance Source	High	The sources were reported.	
	Metric 3: Test Substance Purity	Low	Purity/grade were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Both a blank control and solvent control were used in the acute bioassays.	
	Metric 5: Negative Control Response	High	Control responses (blank and solvent) are shown in Figure 3. Positive control responses are shown in Figures 3, 4, and 5.	
	Metric 6: Randomized Allocation	Low	Random allocation was not reported.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	Preparation of test substances and dilution into test medium was not well described.	
	Metric 8: Consistency of Exposure Administration	High	Exposures appear to have been administered consistently.	
	Metric 9: Measurement of Test Substance Concentration	Low	Concentrations are reported as nominal.	
	Metric 10: Exposure Duration and Frequency	Low	Exposures were 24-hr for embryos, which is shorter than the typical 72-96 hrs utilized in other transgenic fish embryo studies.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Five concentrations were utilized in the pre-testing, from which one concentration per phthalate was utilized in the formal testing.	
	Metric 12: Testing at or Below Solubility Limit	High	Concentrations utilized in the formal testing did not exceed solubility (BBP and DBP) and slightly exceeded solubility (DIDP, DINP, DEHP). Methanol was utilized to increase solubility.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Medium	The source for the transgenic medaka was cited as Chen et al 2007 and Cheng and Chen 2013, but it was not well described in the cited sources.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	Acclimation of embryos in 24-well plates was not reported.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	Each concentration was tested in triplicate with eight embryos per replicate.	
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Study Citation:	Chen, X., Xu, S., Tan, T., Lee, S. T., Cheng, S. H., Lee, F., F.W., Xu, L., S.J., Ho, K. C. (2014). Toxicity and estrogenic endocrine disrupting activity of phthalates and their mixtures. International Journal of Environmental Research and Public Health 11(3):3156-3168.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Oryzias melastigma</i> ; ChgH-EGFP; Larvae
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Receptor binding/ regulation of receptor activity-Endocrine toxicity-Reproductive/Teratogenic
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	2298079

Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	Low	The 24 well plates were described but no other details on environmental conditions were provided.
	Metric 17: Outcome Assessment Methodology	Medium	Measurement of GFP in liver of embryos was described in the methods. Anesthesia of embryos prior to imaging was not described.
	Metric 18: Consistency of Outcome Assessment	High	The outcome assessment appeared to be consistently conducted across treatment and control groups at 72 hr exposure.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20: Outcomes Unrelated to Exposure	Medium	Nothing was reported to indicate that animal health or attrition interfered with the bioassay.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	The authors utilized Student's t-tests (alpha = 0.05) to determine significant difference of phthalate/E2 cotreatment activity from E2 activity. Other data analysis was performed according to ISO 20281.
	Metric 22: Reporting of Data	Medium	Solvent control and positive control data were shown in Figure 3. Response for BBP treatment was shown in Figure 4 and co-treatment response for DBP, DEHP, DIDP, and DINP are shown in Figure 5.
	Metric 23: Explanation of Unexpected Outcomes	Medium	Only one treatment concentration was reported (1.5 mg/L), but it was reported with mean +/- SEM.

Additional Comments: This form applies to BBP, DBP, DEHP, DIDP, and DINP.

Overall Quality Determination

Medium

Study Citation:	Adams, W. J., Biddinger, G. R., Robillard, K. A., Gorsuch, J. W. (1995). A summary of the acute toxicity of 14 phthalate esters to representative aquatic organisms. Environmental Toxicology and Chemistry 14(9):1569-1574.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1321996; Linked HERO ID(s): 1321996, 1316224			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The test substance nomenclature was reported without a CASRN.
	Metric 2:	Test Substance Source	Low	The source was provided by a manufacturer from commercially available batches. The manufacture name and batch number were not provided. No analytical data was reported.
	Metric 3:	Test Substance Purity	High	The test substance was at least 95% pure.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	A negative control was reported.
	Metric 5:	Negative Control Response	High	The control response was acceptable.
	Metric 6:	Randomized Allocation	Low	The allocation method was not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The experimental system was well described. Static tests were conducted in 19.6-L jars with 15 L of test solution. However, headspace or measures to prevent volatilization were not reported.
	Metric 8:	Consistency of Exposure Administration	High	Exposure administration was consistent across groups.
	Metric 9:	Measurement of Test Substance Concentration	Medium	Sample extracts were analyzed by gas chromatography at the start and end of the test. In static studies, final test concentrations frequently were 50% of the initial concentrations. Loss of the phthalate esters was thought to be principally due to adsorption to the test vessels.
	Metric 10:	Exposure Duration and Frequency	High	Duration and frequency of exposure were appropriate for the test.
	Metric 11:	Number of Exposure Groups/Spacing of Exposure Levels	High	Exposure levels were appropriate. A range finding test was performed.
	Metric 12:	Testing at or Below Solubility Limit	High	The test was performed at or below the water solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Appropriate acclimation for the test was reported.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Each test was performed using duplicate test concentrations with 10 organisms per test vessel.
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Study Citation:	Adams, W. J., Biddinger, G. R., Robillard, K. A., Gorsuch, J. W. (1995). A summary of the acute toxicity of 14 phthalate esters to representative aquatic organisms. Environmental Toxicology and Chemistry 14(9):1569-1574.		
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Juvenile		
Health Outcome:	Mortality		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	1321996; Linked HERO ID(s): 1321996, 1316224		
Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions were appropriate for the test.
	Metric 17: Outcome Assessment Methodology	High	The intended outcomes were reported.
	Metric 18: Consistency of Outcome Assessment	High	Outcome assessment was consistent across groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	Environmental conditions were consistent across groups.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There were no reported differences between groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were performed and described.
	Metric 22: Reporting of Data	Medium	Only treatment endpoints were reported.
	Metric 23: Explanation of Unexpected Outcomes	High	No unexpected outcomes were reported.
Additional Comments: None			
Overall Quality Determination		High	

Study Citation:	Adams, W. J., Biddinger, G. R., Robillard, K. A., Gorsuch, J. W. (1995). A summary of the acute toxicity of 14 phthalate esters to representative aquatic organisms. Environmental Toxicology and Chemistry 14(9):1569-1574.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1321996; Linked HERO ID(s): 1321996, 1316224			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	Test substance nomenclature reported without CASRN	
	Metric 2: Test Substance Source	Low	Reported as provided by manufacturer from commercially available batches. Manufacture name and batch number not provided. No analytical data reported.	
	Metric 3: Test Substance Purity	High	At least 95% purity	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Negative control reported	
	Metric 5: Negative Control Response	High	Control response acceptable	
	Metric 6: Randomized Allocation	Low	Allocation method not reported.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	Experimental system well described. However, headspace or measures to prevent volatilization not reported.	
	Metric 8: Consistency of Exposure Administration	High	Exposure administration consistent across groups.	
	Metric 9: Measurement of Test Substance Concentration	High	Sample extracts were analyzed by gas chromatography at start and end of test.	
	Metric 10: Exposure Duration and Frequency	High	Duration and frequency of exposure appropriate for test.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	Exposure levels appropriate. Range finding test was performed.	
	Metric 12: Testing at or Below Solubility Limit	High	Test performed at or below water solubility	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	Source not reported.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	Appropriate acclimation for test reported.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	Each test was performed using duplicate test concentrations with 10 organisms per test vessel.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions appropriate for test.	
	Metric 17: Outcome Assessment Methodology	High	Intended outcomes reported.	
	Metric 18: Consistency of Outcome Assessment	High	Outcome assessment consistent across groups.	
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Study Citation:	Adams, W. J., Biddinger, G. R., Robillard, K. A., Gorsuch, J. W. (1995). A summary of the acute toxicity of 14 phthalate esters to representative aquatic organisms. Environmental Toxicology and Chemistry 14(9):1569-1574.		
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Juvenile		
Health Outcome:	Mortality		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	1321996; Linked HERO ID(s): 1321996, 1316224		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	High	Environmental conditions were consistent across groups.
Metric 20:	Outcomes Unrelated to Exposure	Medium	No reported differences between groups.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	High	Statistical methods performed and described.
Metric 22:	Reporting of Data	Medium	Only treatment endpoints reported.
Metric 23:	Explanation of Unexpected Outcomes	High	No unexpected outcomes reported.
Additional Comments: None			
Overall Quality Determination		High	

Study Citation:	Bencic, D. C., Flick, R. W., Bell, M. E., Henderson, W. M., Huang, W., Purucker, S. T., Glinski, D. A., Blackwell, B. R., Christen, C. H., Stacy, E. H., Biales, A. D. (2024). A multiomics study following acute exposures to phthalates in larval fathead minnows (<i>Pimephales promelas</i>) – The potential application of omics data in risk evaluations under TSCA (internal use only).			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Larvae			
Health Outcome:	Behavioral			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	11581733			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance	Metric 1: Test Substance Identity	High	The chemical name and CASRN were reported.	
	Metric 2: Test Substance Source	Low	The test substance was purchased from MilliporeSigma (Burlington, MA), but no information was given on analytical verification.	
	Metric 3: Test Substance Purity	High	The purity was reported as greater than or equal to 98%.	
Domain 2: Test Design	Metric 4: Negative Controls	High	A control and a control plus O-ring group were tested.	
	Metric 5: Negative Control Response	Low	The goal of the behavior analysis was to calculate a behavior-based point of departure. While the authors reported excluding any inactivity across treatment groups from analysis, the authors did not specifically report if the control group behaved as expected.	
	Metric 6: Randomized Allocation	Medium	Larvae were randomly distributed to chemical exposure beakers and the beakers were randomly arranged on trays in the incubator. In addition, for the behavior analysis to control for positional effects, fish from each of the eight exposure conditions were loaded in order into each of the 6 wells of the first row, with the remaining two being placed in the first two positions of the second row; this pattern was subsequently repeated across replicate plates.	
Domain 3: Exposure Characterization	Metric 7: Experimental System/Test Media Preparation	High	Test solution preparation was described in detail on page 6 and 7. A passive dosing design using O-rings was used due to the low water solubility of the test substance. The exposure system was adequately described on page 7. After the 24 h exposure, live larvae from each exposure vessel were transferred into small plastic weight boats for the behavioral analysis.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were run consistently across treatment groups.	
	Metric 9: Measurement of Test Substance Concentration	High	Measurements of the test substance water concentrations were carried out using an LC-MS system. Measurements were taken in the stock solutions at time zero and at the completion of exposure (24 hours).	
	Metric 10: Exposure Duration and Frequency	High	The exposure duration was 24 hours followed by a behavioral assessment that lasted 28 minutes. This was an adequate exposure time to capture dose-response effects and an adequate behavior assay duration to capture changes in movement patterns.	
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Study Citation:	Bencic, D. C., Flick, R. W., Bell, M. E., Henderson, W. M., Huang, W., Purucker, S. T., Glinski, D. A., Blackwell, B. R., Christen, C. H., Stacy, E. H., Biales, A. D. (2024). A multiomics study following acute exposures to phthalates in larval fathead minnows (<i>Pimephales promelas</i>) – The potential application of omics data in risk evaluations under TSCA (internal use only).			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Larvae			
Health Outcome:	Behavioral			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	11581733			
Domain		Metric	Rating	Comments
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	The number of exposure groups was adequate (11 treatment groups). Specific for the behavior assay, authors collected movement data from the control, control + O-ring, and the 6 highest treatment groups without significant mortality. The authors did not show movement data across treatment groups; instead they used the data to calculate the behavior point of departure.
	Metric 12:	Testing at or Below Solubility Limit	High	Due to the low solubility of the test substance, a passive dosing design using a chemical-saturated high purity silicone O-ring was used, along with diluting stock solutions in methanol.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	FHM larvae were obtained from an on-site breeding culture at AWBERC in Cincinnati, OH. Test organisms were described on pages 4 and 5.
	Metric 14:	Acclimatization and Pretreatment Conditions	Medium	Authors did not specifically mention acclimating test organisms prior to the exposure start, but culture conditions for all larvae were similar to exposure conditions (e.g., 25 degree C incubator, source of water). Moreover, for the behavior assay, fish were acclimated for 10 min in the dark.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	There were 13 larvae placed in each treatment beaker, and there were 3 replicates of each of the 11 treatment groups for the chemical exposure. For the behavioral analyses, 3 larvae were used per replicate.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Test conditions were adequate. Exposures were conducted in accordance with the approved AWBERC Animal Care and Use Protocol. Routine water chemistries were within normal ranges, and both temperature and pH levels exhibited little change throughout the experiment.
	Metric 17:	Outcome Assessment Methodology	High	Following the 24-hour exposure, mortality was recorded in each beaker and dead larvae were removed. High concentrations with substantial (> 10%) mortality were excluded from further analysis. Live larvae from each exposure vessel were transferred into well plates for the behavioral analysis. The behavior analysis was described in detail on page 8 and 13.
	Metric 18:	Consistency of Outcome Assessment	High	The outcome was assessed consistently across treatment groups.
Domain 6: Confounding / Variable Control				

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Study Citation:	Bencic, D. C., Flick, R. W., Bell, M. E., Henderson, W. M., Huang, W., Purucker, S. T., Glinski, D. A., Blackwell, B. R., Christen, C. H., Stacy, E. H., Biales, A. D. (2024). A multiomics study following acute exposures to phthalates in larval fathead minnows (<i>Pimephales promelas</i>) – The potential application of omics data in risk evaluations under TSCA (internal use only).			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Larvae			
Health Outcome:	Behavioral			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	11581733			
Domain	Metric	Rating	Comments	
	Metric 19: Confounding Variables in Test Design and Procedures	High	All study treatment groups were treated equally throughout the experiment. For the behavior assay, fish across treatment groups were handled in the same manner (e.g., transfer from beakers to well-plates, acclimation period in well-plates). While authors did not specifically mention acclimating test organisms prior to the chemical exposure start, culture conditions and exposure conditions (e.g., minus absence/presence chemical) were similar and appropriate pre-exposure and during exposure. Routine water chemistries were within normal ranges, and both temperature and pH levels exhibited little change throughout the experiment.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information given about differences among treatment group organisms that could influence the outcome assessment.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	The behavioral data was analyzed using the ZebraLab software. The bPOD calculation was described in detail on page 14.	
	Metric 22: Reporting of Data	Medium	bPOD outcomes were described in the text of the results section, and Table 6 gives a bPOD values for DBP. However, behavior data (movement patterns) across treatment groups were not included in the report.	
	Metric 23: Explanation of Unexpected Outcomes	Low	Measures of variability were not given with the behavioral data.	
Additional Comments:	The primary goal of this study was to investigate the potential application of omics data in risk evaluation. This evaluation is for the calculation of the behavior point of departure as part of the behavior analysis following exposure to DBP.			

Overall Quality Determination**High**

Study Citation:	Bencic, D. C., Flick, R. W., Bell, M. E., Henderson, W. M., Huang, W., Purucker, S. T., Glinski, D. A., Blackwell, B. R., Christen, C. H., Stacy, E. H., Biales, A. D. (2024). A multiomics study following acute exposures to phthalates in larval fathead minnows (<i>Pimephales promelas</i>) – The potential application of omics data in risk evaluations under TSCA (internal use only).			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Larvae			
Health Outcome:	Mechanistic-Cell signaling/function			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	11581733			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The chemical name and CASRN were reported.
	Metric 2:	Test Substance Source	Low	The test substance was purchased from MilliporeSigma (Burlington, MA), but no information was given on analytical verification.
	Metric 3:	Test Substance Purity	High	The purity was reported as greater than or equal to 98%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	A control and a control plus O-ring group were tested.
	Metric 5:	Negative Control Response	Medium	The effect measured is gene expression (transcriptomic analysis). There is no gene expression profile established for control/unexposed organisms in this developmental stage for this species, particularly gene expression data from the entire fish. Also, given the nature of the collection of fertilized eggs and an outbred population, gene expression even among unexposed fish is expected to vary. From a sequencing perspective, there were no unexpected results.
	Metric 6:	Randomized Allocation	Medium	Larvae were randomly distributed to exposure beakers, and the beakers were randomly arranged on trays in the incubator.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	Test solution preparation was described in detail on page 6 and 7. A passive dosing design using O-rings was used due to the low water solubility of the test substance. The exposure system was adequately described on page 7. The RNA analysis was described on pages 8-10.
	Metric 8:	Consistency of Exposure	High	Exposures were run consistently across treatment groups.
	Metric 9:	Administration Measurement of Test Substance Concentration	High	Measurements of the test substance water concentrations were carried out using an LC-MS system. Measurements were taken in the stock solutions at time zero and at the completion of exposure (24 hours).
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration was 24 hours and was an appropriate time length to observe transcriptomic changes.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups (11 treatment groups) and spacing was adequate to obtain the transcriptomic changes and derive a transcriptomic point of departure.
	Metric 12:	Testing at or Below Solubility Limit	High	Due to the low solubility of the test substance, a passive dosing design using a chemical-saturated high purity silicone O-ring was used, along with diluting stock solutions in methanol.
Domain 4: Test Organism				
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Study Citation:	Bencic, D. C., Flick, R. W., Bell, M. E., Henderson, W. M., Huang, W., Purucker, S. T., Glinski, D. A., Blackwell, B. R., Christen, C. H., Stacy, E. H., Biales, A. D. (2024). A multiomics study following acute exposures to phthalates in larval fathead minnows (<i>Pimephales promelas</i>) – The potential application of omics data in risk evaluations under TSCA (internal use only).			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Larvae			
Health Outcome:	Mechanistic-Cell signaling/function			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	11581733			
Domain	Metric	Rating	Comments	
	Metric 13: Test Organism Characteristics	High	FHM larvae were obtained from an on-site breeding culture at AWBERC in Cincinnati, OH. Test organisms were described on pages 4 and 5.	
	Metric 14: Acclimatization and Pretreatment Conditions	Medium	Authors didn’t specifically mention acclimating test organisms prior to the exposure start, but culture conditions for the larvae were similar to exposure conditions (25 degree C incubator, source of water).	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 13 larvae placed in each treatment beaker, and there were 3 replicates of each of the 11 treatment groups for the exposure. For the transcriptomics analysis, there were 4 larvae per replicate. Each individual well contained one larva.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Test conditions were adequate. Exposures were conducted in accordance with the approved AWBERC Animal Care and Use Protocol. Routine water chemistries were within normal ranges, and both temperature and pH levels exhibited little change throughout the experiment.	
	Metric 17: Outcome Assessment Methodology	High	Following the 24-hour exposure, mortality was recorded in each beaker and dead larvae were removed. High concentrations with substantial (> 10%) mortality were excluded from further analysis. Live larvae from each exposure were used for RNA analysis. The sample processing, RNA isolation, RNA sequencing, and tPOD calculation were described in detail on pages 8-10.	
	Metric 18: Consistency of Outcome Assessment	High	The outcome was assessed consistently across treatment groups.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	All study treatment groups were treated equally throughout experiment. While authors did not specifically mention acclimating test organisms prior to the exposure start, culture conditions and exposure conditions (e.g., minus absence/presence chemical) were similar and appropriate pre-exposure and during exposure. Routine water chemistries were within normal ranges, and both temperature and pH levels exhibited little change throughout the experiment.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information given about differences among treatment group organisms that could influence the outcome assessment.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	tPOD calculation was described on pages 10 and 11 and seemed appropriate to assess results.	
	Metric 22: Reporting of Data	High	Table 4 shows tPOD data and some results are described in the ”PODs” section of the results.	

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Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Larvae		
Health Outcome:	Mechanistic-Cell signaling/function		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	11581733		
Domain	Metric	Rating	Comments
	Metric 23: Explanation of Unexpected Outcomes	High	Measures of variability were given with the tPOD data.
Additional Comments:	The primary goal of this study was to investigate the potential application of omics data in risk evaluation. This data evaluation is for the calculation of the transcriptomics-based point of departure following exposure DBP.		
Overall Quality Determination		High	

Study Citation:	Bencic, D. C., Flick, R. W., Bell, M. E., Henderson, W. M., Huang, W., Purucker, S. T., Glinski, D. A., Blackwell, B. R., Christen, C. H., Stacy, E. H., Biales, A. D. (2024). A multiomics study following acute exposures to phthalates in larval fathead minnows (<i>Pimephales promelas</i>) – The potential application of omics data in risk evaluations under TSCA (internal use only).			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Larvae			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	11581733			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The chemical name and CASRN were reported.
	Metric 2:	Test Substance Source	Low	The test substance was purchased from MilliporeSigma (Burlington, MA), but no information was given on analytical verification.
	Metric 3:	Test Substance Purity	High	The purity was reported as greater than or equal to 98%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	A control and a control plus O-ring group were tested.
	Metric 5:	Negative Control Response	High	There were no mortalities in the control test groups.
	Metric 6:	Randomized Allocation	Medium	Larvae were randomly distributed to exposure beakers, and the beakers were randomly arranged on trays in the incubator.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	Test solution preparation was described in detail on page 6 and 7. A passive dosing design using O-rings was used due to the low water solubility of the test substance. The exposure system was adequately described on page 7.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were run consistently across treatment groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Measurements of the test substance water concentrations were carried out using an LC-MS system. Measurements were taken in the stock solutions at time zero and at the completion of exposure (24 hours).
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration was 24 hours and was an appropriate time length to observe mortality effects.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups was adequate for the experimental design (11 treatment groups), and spacing was appropriate as a NOEC was determined at the end of the study (i.e., lowest concentration was low enough).
	Metric 12:	Testing at or Below Solubility Limit	High	Due to the low solubility of the test substance, a passive dosing design using a chemical-saturated high purity silicone O-ring was used, along with diluting stock solutions in methanol.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	FHM larvae were obtained from an on-site breeding culture at AWBERC in Cincinnati, OH. Test organisms were described on pages 4 and 5.
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Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Larvae			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	11581733			
Domain	Metric	Rating	Comments	
	Metric 14:	Acclimatization and Pretreatment Conditions	Medium	Authors did not specifically mention acclimating test organisms prior to the exposure start, but culture conditions for all larvae were similar to exposure conditions (e.g., 25 degree C incubator, source of water +/- phthalate).
	Metric 15:	Number of Organisms and Replicates per Group	Medium	There were 13 larvae placed in each treatment beaker, and there were 3 replicates of each of the 11 treatment groups.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Test conditions were adequate. Exposures were conducted in accordance with the approved AWBERC Animal Care and Use Protocol. Routine water chemistries were within normal ranges, and both temperature and pH levels exhibited little change throughout the experiment.
	Metric 17:	Outcome Assessment Methodology	High	Following the 24-hour exposure, mortality was recorded in each beaker and dead larvae were removed. High concentrations with substantial (> 10%) mortality were excluded from further analysis.
	Metric 18:	Consistency of Outcome Assessment	High	The outcome was assessed consistently across treatment groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	All study treatment groups were treated equally throughout the experiment. While authors did not specifically mention acclimating test organisms prior to the exposure start, culture conditions and exposure conditions (e.g., minus absence/presence chemical) were similar and appropriate pre-exposure and during exposure. Routine water chemistries were within normal ranges and both temperature and pH levels exhibited little change throughout the experiment.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information given about differences among treatment group organisms that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	LC50 calculation is described in detail in the supplemental document. Also, NOEC derivation was briefly described on page 13 of the report, and Table 2 reported the raw data and percent mortality.
	Metric 22:	Reporting of Data	High	Table 2 (2B) showed the mortality data across all treatments for all 3 replicates.
	Metric 23:	Explanation of Unexpected Outcomes	Low	Measures of variability were not given with the mortality data.
Additional Comments:	The primary goal of this study was to investigate the potential application of omics data in risk evaluation. This specific evaluation is for the mortality assessment with DBP.			

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Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Larvae		
Health Outcome:	Mortality		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	11581733		
Domain	Metric	Rating	Comments
Overall Quality Determination		High	

Study Citation:	Bencic, D. C., Flick, R. W., Bell, M. E., Henderson, W. M., Huang, W., Purucker, S. T., Glinski, D. A., Blackwell, B. R., Christen, C. H., Stacy, E. H., Biales, A. D. (2024). A multiomics study following acute exposures to phthalates in larval fathead minnows (<i>Pimephales promelas</i>) – The potential application of omics data in risk evaluations under TSCA (internal use only).			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Larvae			
Health Outcome:	Mechanistic-Cell signaling/function			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	11581733			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The chemical name and CASRN were reported.
	Metric 2:	Test Substance Source	Low	The test substance was purchased from MilliporeSigma (Burlington, MA), but no information was given on analytical verification.
	Metric 3:	Test Substance Purity	High	The purity was reported as greater than or equal to 98%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	A control and a control plus O-ring group were tested.
	Metric 5:	Negative Control Response	High	The effect measured is metabolomics. There is no metabolomic profile established for control/unexposed organisms in this developmental stage for this species, particularly a metabolomics profile from the entire fish. That said, to best evaluate ‘normality’ of the control response, the metabolite profiles of the fathead minnow larvae with and without o-rings (i.e. control vs. vehicle control) were compared and no discernable differences in their profiles were observed leading to the conclusion of ‘no unexpected observed measurements in controls’. Also, given the nature of the collection of fertilized eggs and an outbred population, a metabolomic profile even among unexposed fish is expected to vary. From a spectra analysis perspective, there were no unexpected results.
	Metric 6:	Randomized Allocation	Medium	Larvae were randomly distributed to exposure beakers, and the beakers were randomly arranged on trays in the incubator.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	Test solution preparation was described in detail on page 6 and 7. A passive dosing design using O-rings was used due to the low water solubility of the test substance. The exposure system was adequately described on page 7. The metabolomics analysis procedure was further described on page 11.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were run consistently across treatment groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Measurements of the test substance water concentrations were carried out using an LC-MS system. Measurements were taken in the stock solutions at time zero and at the completion of exposure (24 hours).
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration was 24 hours and was an appropriate time length to observe metabolomic results.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups seemed adequate (11 treatment groups).
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Study Citation:	Bencic, D. C., Flick, R. W., Bell, M. E., Henderson, W. M., Huang, W., Purucker, S. T., Glinski, D. A., Blackwell, B. R., Christen, C. H., Stacy, E. H., Biales, A. D. (2024). A multiomics study following acute exposures to phthalates in larval fathead minnows (<i>Pimephales promelas</i>) – The potential application of omics data in risk evaluations under TSCA (internal use only).			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Larvae			
Health Outcome:	Mechanistic-Cell signaling/function			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	11581733			
Domain	Metric	Rating	Comments	
	Metric 12:	Testing at or Below Solubility Limit	High	Due to the low solubility of the test substance, a passive dosing design using a chemical-saturated high purity silicone O-ring was used, along with diluting stock solutions in methanol.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	FHM larvae were obtained from an on-site breeding culture at AWBERC in Cincinnati, OH. Test organisms were described on pages 4 and 5.
	Metric 14:	Acclimatization and Pretreatment Conditions	Medium	Authors didn't specifically mention acclimating test organisms prior to the exposure start, but culture conditions for the larvae were similar to exposure conditions (e.g., 25 degree C incubator, source of water).
	Metric 15:	Number of Organisms and Replicates per Group	Medium	There were 13 larvae placed in each treatment beaker, and there were 3 replicates of each of the 11 treatment groups for the exposure. For the metabolomics analysis, there were 3 larvae per replicate. Three larvae from each set of the replicate treatments were transferred to individual wells of a 1.0 mL 96-well deep well plate.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Test conditions were adequate. Exposures were conducted in accordance with the approved AWBERC Animal Care and Use Protocol. Routine water chemistries were within normal ranges, and both temperature and pH levels exhibited little change throughout the experiment.
	Metric 17:	Outcome Assessment Methodology	High	Following the 24-hour exposure, mortality was recorded in each beaker and dead larvae were removed. High concentrations with substantial (> 10%) mortality were excluded from further analysis. Live larvae from each exposure were used for metabolomics analysis. The metabolite extraction, metabolite derivatization, GC-qToF/MS analysis, and mPOD calculation were described in detail on pages 10-12.
	Metric 18:	Consistency of Outcome Assessment	High	The outcome was assessed consistently across treatment groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	All study treatment groups were treated equally throughout the experiment. While authors did not specifically mention acclimating test organisms prior to the exposure start, culture conditions and exposure conditions (e.g., minus absence/presence chemical) were similar and appropriate pre-exposure and during exposure. Routine water chemistries were within normal ranges, and both temperature and pH levels exhibited little change throughout the experiment.
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Study Citation:	Bencic, D. C., Flick, R. W., Bell, M. E., Henderson, W. M., Huang, W., Purucker, S. T., Glinski, D. A., Blackwell, B. R., Christen, C. H., Stacy, E. H., Biales, A. D. (2024). A multiomics study following acute exposures to phthalates in larval fathead minnows (<i>Pimephales promelas</i>) – The potential application of omics data in risk evaluations under TSCA (internal use only).			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Larvae			
Health Outcome:	Mechanistic-Cell signaling/function			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	11581733			
Domain	Metric	Rating	Comments	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information given about differences among treatment group organisms that could influence the outcome assessment.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Metabolomics data analysis and mPOD calculation was described on pages 11 and 12 and seemed appropriate to assess results.	
	Metric 22: Reporting of Data	High	Table 5 shows mPOD data and some results are described in the "PODs" section of the results.	
	Metric 23: Explanation of Unexpected Outcomes	High	Measures of variability were given with the mPOD data.	
Additional Comments:	The primary goal of this study was to investigate the potential application of omics data in risk evaluation. This data evaluation is for the calculation of the metabolomics-based point of departure following exposure to DBP.			

Overall Quality Determination**High**

Study Citation:	Bionomics,, EG&G (1983). Acute toxicity of fourteen phthalate esters to fathead minnows.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1316188			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The test substance was identified definitively by accepted name [di-n-butyl phthalate; DBP]
	Metric 2:	Test Substance Source	Low	The study states "The fourteen phthalate esters, all clear colorless liquids, labeled 1A through 1N, were received on 18 December 1981 in 1-liter amber glass bottles."In the appendix on analytical methods, the study states that the test chemicals were "supplied by the Chemical Manufacturers Association, Washington. D.C."However, specific details about the test substance source (manufacturer, batch/lot #, etc..) were not reported and test substance identities were not analytically verified.
	Metric 3:	Test Substance Purity	Low	Purities of the test substances were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Appropriate, concurrent negative control groups were used."Duplicate control jars containing the same dilution water and maintained under the same conditions as the exposure jars, but containing no test material, were established."
	Metric 5:	Negative Control Response	High	No mortalities were observed in the control groups, and the study notes that "The pH values of the test solutions remained comparable to the controls throughout the exposure period (Table "3a). Dissolved oxygen concentrations of the test solutions were comparable to the controls at the testis initiation; however, DO concentrations were reduced with increasing [DBP] concentrations at the end of the exposure."
	Metric 6:	Randomized Allocation	Medium	"Ten fathead minnows (population descriptions in Table 1) were randomly distributed to each test jar after the test solutions had been prepared."
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	Experimental system and test media preparation were described in adequate detail. Tests were conducted in 19.6L glass jars, filled with 15L of dilution water and appropriate amount of test substance to achieve desired concentration. The temperature was controlled. "Dilution water used was soft water reconstituted from deionized water" (characteristics given). Test solutions were not aerated. Water temperature, pH, DO, hardness, alkalinity, and specific conductance were all monitored. No measures were taken to prevent loss of test substance over the course of the exposure, and in fact the authors reported that "an appreciable loss of phthalate ester from solution occurred during each exposure. The concentrations of phthalate ester present in solution at the end of the exposures ranged from <4 to 68% of the 0-hour concentration."
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Study Citation:	Bionomics,, EG&G (1983). Acute toxicity of fourteen phthalate esters to fathead minnows.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1316188			
Domain	Metric	Rating	Comments	
	Metric 8: Consistency of Exposure Administration	Low	Since no measures were used to maintain a consistent exposure concentration over the course of the experiment, there is no way to know how confidently two different exposure groups can be compared. The 96hr concentration shows the minimum possible exposure the test organisms were subjected to, but since no intermediate measurements were reported, the rate at which the test substance was lost, and how consistently the loss occurred between groups is unknown.	
	Metric 9: Measurement of Test Substance Concentration	Medium	Test substance concentrations were only measured at the beginning and end of the study (0-hr and 96-hr). Analytical methods were appropriate, and detailed in an appendix – "An aliquot of the concentrate was analyzed by gas-liquidchromatography with electron capture detection."	
	Metric 10: Exposure Duration and Frequency	High	Exposure duration is in accordance with the cited methodology [Methods for Acute Toxicity Tests with Fish, Macroinvertebrates, and Amphibians" (U.S. EPA, 1975).]	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	Five different exposure concentrations under the limit of water solubility for DBP were used (40, 24, 14, 8.7 and 5.2 uL/L; nominal concentration).	
	Metric 12: Testing at or Below Solubility Limit	High	No exposure concentrations were above the limit of solubility; as such, no solvents were necessary.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Medium	Complete details regarding test organism characteristics are lacking. Only the source ("fathead minnows were obtained from cultures maintained at EG&G, Bionomics, Wareham, Massachusetts") and the mean length and mean weight of the test organisms in each fish population lot were reported.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All test organisms underwent the same pretreatment process, including an at least 14-day pretreatment period and a 48-hr acclimatization period immediately before the tests (described in detail on page 2 of the study).	
	Metric 15: Number of Organisms and Replicates per Group	Low	Ten fish were included in each treatment group. It appears two replicates were conducted for each exposure concentration.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Test conditions were adequate – well detailed on pages 2-4 of the study (16hr light /8hr dark photoperiod; dry food fed ad libitum during the pretreatment period; 22+/-1 °c temp; measured water parameters stated).	
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Study Citation:	Bionomics,, EG&G (1983). Acute toxicity of fourteen phthalate esters to fathead minnows.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1316188			
Domain	Metric	Rating	Comments	
	Metric 17:	Outcome Assessment Methodology	Uninformative	The authors report the percentage mortality for each study group at 24hr, 48hr, 72hr and 96hr. Since repeated concentration measurements were not taken, the accuracy of a NOEC / LC50 derived from this data is limited. A NOEC can be derived from the lowest reported concentration (96hr), but the actual exposure experienced by the test organisms could range anywhere between there and the highest reported (0hr) concentration. The authors state "LC50 values were calculated using 0-hour analytical results since a significant loss of phthalate ester from solution occurred during many tests. Estimating LC50 values on 0-hour analytical results allows for more equitable comparisons between tests." Since the actual concentration the test organisms were exposed to is unknown, the reported LC50 is not accurate to a specific concentration, but to a widely variable range of potential concentrations lower than the reported LC50.
	Metric 18:	Consistency of Outcome Assessment	High	Outcome assessment was done consistently across study groups."The computer program utilized (Stephan, 1978, personal communication) estimated LC50 values using one of three statistical methods in the following order of preference: moving average angle analysis, probit analysis, binomial probability. The method selected was determined by the characteristics of the data base (i.e. presence or absence of test concentrations causing mortality of 100% of the animals in the test population, test concentrations causing mortality of a partial number of animals in the population, etc.). The computer program scanned the data base, identified the most appropriate statistical method and performed the analysis."
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	Many variables were measured, and appear to be consistent across study and control groups."Good quality control was maintained throughout the analyses as indicated by the QA spiked sample analytical results. The pH values and dissolved oxygen concentrations remained comparable to the respective controls during exposures to solutions of phthalate esters IE through IN."
	Metric 20:	Outcomes Unrelated to Exposure	High	No attrition of test organisms unrelated to exposure was reported.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	N/A	Statistical analysis for LC50 calculation was selected and performed by a computer program (in order of preference: moving average angle analysis, probit analysis, or binomial probability).
	Metric 22:	Reporting of Data	High	Data is reported for all study groups in the appendix.
	Metric 23:	Explanation of Unexpected Outcomes	High	No unexpected outcomes occurred.

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Study Citation:	Bionomics,, EG&G (1983). Acute toxicity of fourteen phthalate esters to fathead minnows.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route,	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Media, Path:	
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	1316188

Domain	Metric	Rating	Comments
Additional Comments:	Authors note that substantial loss of test material occurred between 0-hr and 96-hr point, but used the 0-hr concentration to calculate the LC50. Tests organisms were actually exposed to an unknown, lower concentration than the 0-hr reported concentration, so this results in an inflated LC50 (the true LC50 would be lower than the reported value).		

Overall Quality Determination**Uninformative**

Study Citation:	Defoe, D. L., Holcombe, G. W., Hammermeister, D. E., Biesinger, K. E. (1990). Solubility and toxicity of eight phthalate esters to four aquatic organisms. Environmental Toxicology and Chemistry 9(5):623-636.		
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Juvenile		
Health Outcome:	Mortality		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	5774391		
Domain	Metric	Rating	Comments
Domain 1: Test Substance			
Metric 1:	Test Substance Identity	Low	The full chemical name is provided in addition to the acronym but no specific CAS number or additional information is provided.
Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified.
Metric 3:	Test Substance Purity	Medium	One test substance (DBoP, di-n-butyl ortho) was commercially purchased and of high purity (>99%). The chemical was also synthesized for tests and purity not analytically verified. Iso and tere forms were also synthesized.
Domain 2: Test Design			
Metric 4:	Negative Controls	High	Control groups (no chemical) were evaluated in concert with treatment groups. No solvents were used to necessitate a solvent control.
Metric 5:	Negative Control Response	High	There were no concerns or anomalies associated with control groups.
Metric 6:	Randomized Allocation	Medium	Fish were randomly distributed among treatments and randomly subsampled for measurements.
Domain 3: Exposure Characterization			
Metric 7:	Experimental System/Test Media Preparation	High	Sufficient detail is provided regarding the experimental design as well as development of stock solutions. Water physiochemical characteristics were regularly measured and reported. DBP concentrations were measured and reported.
Metric 8:	Consistency of Exposure	High	Details of exposure are provided and are consistent among study groups.
Metric 9:	Administration	High	Exposure concentrations were measured using appropriate methods.
Metric 10:	Measurement of Test Substance Concentration	High	Exposure was appropriate and followed standard ASTM protocols.
Metric 11:	Exposure Duration and Frequency	High	The range of concentrations allowed for calculation of an LC50.
Metric 12:	Number of Exposure Groups/Spacing of Exposure Levels	High	
Metric 12:	Testing at or Below Solubility Limit	Medium	Some test concentrations were at or above solubility though verification of concentrations was provided and care taken to ensure minimal degradation or loss of test substance during experiments.
Domain 4: Test Organism			
Metric 13:	Test Organism Characteristics	High	Test organisms were obtained from a reliable source and test organism details were provided.
Metric 14:	Acclimatization and Pretreatment Conditions	Low	Minimal details on pretreatment were provided.

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Study Citation:	Defoe, D. L., Holcombe, G. W., Hammermeister, D. E., Biesinger, K. E. (1990). Solubility and toxicity of eight phthalate esters to four aquatic organisms. Environmental Toxicology and Chemistry 9(5):623-636.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Juvenile
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	5774391

Domain	Metric	Rating	Comments
	Metric 15: Number of Organisms and Replicates per Group	Low	Test organisms follow standard ASTM protocol and are reported as fish loading (g/L).
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Study conditions (physiochemical characteristics, feeding details) are well documented.
	Metric 17: Outcome Assessment Methodology	High	The outcome of interest (LC50) was appropriate.
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no variations or inconsistencies reported across study groups and environmental conditions are provided.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There is no information to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were appropriate.
	Metric 22: Reporting of Data	Medium	Minimal results were reported, though LC50 values are provided.
	Metric 23: Explanation of Unexpected Outcomes	High	Authors reported no unexpected outcomes and reported variance. Authors provided discussion on different results measured in other papers.
Additional Comments: A 96h acute toxicity test for DBP (ortho, iso, tere) reported LC50 values. Behavior was also assessed. DBP was toxic to minnows.			

Overall Quality Determination**High**

Study Citation:	Mccarthy, J. F., Whitmore, D. K. (1985). Chronic toxicity of di-n-butyl and di-n-octyl phthalate to daphnia-magna and the fathead minnow. Environmental Toxicology and Chemistry 4(2):167-179.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Embryo			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1336024			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	The chemical purity was reported as 99.5 %.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	Low	The biological response of the negative control group was not reported for the preliminary test.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.	
	Metric 8: Consistency of Exposure Administration	Medium	Daily renewals occurred, but few details were provided.	
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured and are similar to nominal concentrations.	
	Metric 10: Exposure Duration and Frequency	Medium	The duration of exposure was reported but was shorter than recommended.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	Pretreatment conditions were the same for control and exposed organisms, but details were limited.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 30-50 embryos per treatment with two replicates.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health.	
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Study Citation:	Mccarthy, J. F., Whitmore, D. K. (1985). Chronic toxicity of di-n-butyl and di-n-octyl phthalate to daphnia-magna and the fathead minnow. Environmental Toxicology and Chemistry 4(2):167-179.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Embryo			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1336024			
Domain		Metric	Rating	Comments
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	Results for this criterion should be related to the solvent control. There was statistically significant mortality in the negative control group that may impact results for reproductive effects. The study authors attempted to salvage the study by relating everything to surviving D. magna. However, this introduces further uncertainty regarding the actual effects of DBP on D. magna. "Survival of D. magna exposed to DBP exceeded 80% in all concentrations except 1.8 and 3.2 mg/L and in the carrier-free control. The reason for the poor survival (and poor reproduction) of the control group is not clear; the same problem was evident in the DOP experiment (see below). By the end of the experiment (day 16), 70% of the D. magna were alive at 1.8 mg/L and 18% were alive at 3.2 mg/L DBP (Fig. 1 and Table 3)."
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23:	Explanation of Unexpected Outcomes	Low	There was unexpectedly low survival in the negative control.
Additional Comments:	This form is for fathead minnow mortality in the preliminary range finding test. Results were reported in the text as an LC50 value for 96h of 2.02mg/L for DBP. Confidence intervals (95%) were reported as well.			
Overall Quality Determination			Medium	

Study Citation:	Viscient., Smithers (2018). Di-n-butyl phthalate - short-term reproduction assay with fathead minnow (<i>Pimephales promelas</i>) following OPPTS 890.1350 and OECD 229 guidelines.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	10064185			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The chemical was identified by name and CAS#.	
	Metric 2: Test Substance Source	High	The source of the test substance was reported and the lot number was identified.	
	Metric 3: Test Substance Purity	High	Chemical purity was reported as 99.3%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The experimental system and methods for preparation of test media were minimally described.	
	Metric 8: Consistency of Exposure Administration	Medium	Few details were provided about the flow-through system.	
	Metric 9: Measurement of Test Substance Concentration	Medium	Exposure concentrations were measured using appropriate analytical technologies and methods but were about half of nominals.	
	Metric 10: Exposure Duration and Frequency	Medium	The duration of exposure was suitable for a preliminary study.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	Adults were tested, which is suitable for a preliminary test but not standard for an acute test.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.	
	Metric 15: Number of Organisms and Replicates per Group	Low	The number of test organisms and replicates were reported. The numbers were low and replicates were not used.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Medium	Environmental conditions of the test system were most likely conducive to maintenance of organism health but actual measured condition values for control and exposed vessels were not reported	

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Study Citation:	Viscient., Smithers (2018). Di-n-butyl phthalate - short-term reproduction assay with fathead minnow (<i>Pimephales promelas</i>) following OPPTS 890.1350 and OECD 229 guidelines.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Adult
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	10064185

Domain	Metric	Rating	Comments
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Sufficient data were provided to conduct an independent statistical analysis.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This form is for the preliminary exposure, which begins on P. 146 in Appendix 6.

Overall Quality Determination

Medium

Study Citation:	Bionomics,, EG&G (1984). Acute toxicity of thirteen phthalate esters to fathead minnows (<i>Pimephales promelas</i>) under flow-through conditions.		
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Mortality		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	1316189		
Domain	Metric	Rating	Comments
Domain 1: Test Substance			
	Metric 1: Test Substance Identity	Low	The test substance was identified only by nomenclature. No other information (CASRN, structure etc.) was provided.
	Metric 2: Test Substance Source	Low	The test substance was received from the General Electric Company, Hudson Falls, New York, on 11 and 18 December 1981. The test substance identity was NOT analytically verified by the performing laboratory.
	Metric 3: Test Substance Purity	High	Authors reported the test substance as "100% active ingredient" (doubtful that it is literally one hundred percent but can be taken to mean very pure as received from manufacturer).
Domain 2: Test Design			
	Metric 4: Negative Controls	High	Negative controls were used.
	Metric 5: Negative Control Response	High	There was no unacceptable mortality in controls.
	Metric 6: Randomized Allocation	Medium	"The test was initiated when ten fathead minnows were randomly distributed to each aquarium "
Domain 3: Exposure Characterization			
	Metric 7: Experimental System/Test Media Preparation	Medium	The flow-through regime was described in detail. Stock solutions were prepared daily & maintained throughout the experiment. Observations of droplets of undissolved test substance and cloudy test solution were reported at high concentrations (Table 16).
	Metric 8: Consistency of Exposure	High	Exposures were administered consistently across study groups.
	Metric 9: Administration Measurement of Test Substance Concentration	High	Test substance concentrations were measured pre-test, 0h, 96h, and 144h.
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type and/or outcome(s) of interest (96-h acute toxicity study for fathead minnows, extended to 144h to observe additional mortality as per study design).
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	Numbers and spacing of exposure groups were adequate.
	Metric 12: Testing at or Below Solubility Limit	High	The highest dose chosen was at the solubility limit of the chemical as "communicated verbally to EG&G Bionomics from the Syracuse Research Corporation (1982)." Some droplets of undissolved phthalate were seen at high concentrations (Table 16). See Appendix I for details of the solubilizing apparatus used to disperse phthalates in the exposure water.
Domain 4: Test Organism			

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Study Citation:	Bionomics,, EG&G (1984). Acute toxicity of thirteen phthalate esters to fathead minnows (<i>Pimephales promelas</i>) under flow-through conditions.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1316189			
Domain		Metric	Rating	Comments
	Metric 13:	Test Organism Characteristics	Medium	Fathead minnows were from an in-house culture. Length and wet weight measurements were given in Table 1.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Acclimation was performed in holding tanks for "a minimum of 14 days" for both control & exposed fish.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	There were ten fish per group with two replicates.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Test conditions were described in detail & adequate for maintaining the health of P. promelas.
	Metric 17:	Outcome Assessment Methodology	High	Fish were observed for mortality every 24 hours.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information to suggest differences among groups in attrition or other health outcomes.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but described in limited detail. (Computer program cited as a personal communication using one of three statistical methods for estimating the LC50, but no details of mathematical operations were given).
	Metric 22:	Reporting of Data	High	Data for all outcomes was presented.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments: Definitive LC 50 values (24, 48, 72, 96, 120 and 144 hours) reported for DBP.				

Overall Quality Determination**High**

Study Citation:	Mccarthy, J. F., Whitmore, D. K. (1985). Chronic toxicity of di-n-butyl and di-n-octyl phthalate to daphnia-magna and the fathead minnow. Environmental Toxicology and Chemistry 4(2):167-179.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Embryo			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1336024			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	Chemical was identified by name only	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity reported as 99.5 %	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group	
	Metric 5: Negative Control Response	Medium	The biological response of the negative control group was reported and reasonable for assessed outcomes although the solvent control had poor survival and hatch	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations	
	Metric 8: Consistency of Exposure Administration	Medium	Daily renewals but few details were provided	
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured and are similar to nominal concentrations	
	Metric 10: Exposure Duration and Frequency	Medium	The duration of exposure was reported but was shorter than recommended	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source	
	Metric 14: Acclimatization and Pretreatment Conditions	High	Pretreatment conditions were the same for control and exposed organisms, details were limited	
	Metric 15: Number of Organisms and Replicates per Group	Medium	30-50 embryos per treatment with two replicates	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health	
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Study Citation:	Mccarthy, J. F., Whitmore, D. K. (1985). Chronic toxicity of di-n-butyl and di-n-octyl phthalate to daphnia-magna and the fathead minnow. Environmental Toxicology and Chemistry 4(2):167-179.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Embryo
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	1336024

Domain	Metric	Rating	Comments
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18: Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	Results for this criterion should be related to the solvent control. There was statistically-significant mortality in the negative control group that may impact results for reproductive effects. The study authors attempted to salvage the study by relating everything to surviving D. magna. However, this introduces further uncertainty regarding the actual effects of DBP on D. magna. "Survival of D. magna exposed to DBP exceeded 80% in all concentrations except 1.8 and 3.2 mg/L and in the carrier-free control. The reason for the poor survival (and poor reproduction) of the control group is not clear; the same problem was evident in the DOP experiment (see below). By the end of the experiment (day 16), 70% of the D. magna were alive at 1.8 mg/L and 18% were alive at 3.2 mg/L DBP (Fig. 1 and Table 3)."
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23: Explanation of Unexpected Outcomes	Low	Unexpectedly low survival in negative control

Additional Comments: This form is for fathead minnow mortality in the early life-stage test.

Overall Quality Determination

Medium

Study Citation:	Viscient., Smithers (2018). Di-n-butyl phthalate - short-term reproduction assay with fathead minnow (<i>Pimephales promelas</i>) following OPPTS 890.1350 and OECD 229 guidelines.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Adult			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	10064185			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	High	The source of the test substance was reported and the lot number was identified
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.3%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The test organisms were adequately described and were obtained from an unnamed lab source
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health

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Study Citation:	Viscient,, Smithers (2018). Di-n-butyl phthalate - short-term reproduction assay with fathead minnow (<i>Pimephales promelas</i>) following OPPTS 890.1350 and OECD 229 guidelines.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Adult
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Reproductive/Teratogenic
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	10064185

Domain	Metric	Rating	Comments
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18: Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
	Metric 20: Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: None

Overall Quality Determination**High**

Study Citation:	Viscient., Smithers (2018). Di-n-butyl phthalate - short-term reproduction assay with fathead minnow (<i>Pimephales promelas</i>) following OPPTS 890.1350 and OECD 229 guidelines.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Adult			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	10064185			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The chemical was identified by name and CAS#.
	Metric 2:	Test Substance Source	High	The source of the test substance was reported and the lot number was identified.
	Metric 3:	Test Substance Purity	High	Chemical purity was reported as 99.3%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable.
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8:	Consistency of Exposure	High	Exposures were administered consistently across study groups.
	Metric 9:	Administration Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The test organisms were adequately described and were obtained from an unnamed lab source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The number of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
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Study Citation:	Viscient,, Smithers (2018). Di-n-butyl phthalate - short-term reproduction assay with fathead minnow (<i>Pimephales promelas</i>) following OPPTS 890.1350 and OECD 229 guidelines.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Adult
Health Outcome:	Reproductive/Teratogenic
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	10064185

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	High	There were no differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: None

Overall Quality Determination**High**

Study Citation:	Viscient., Smithers (2018). Di-n-butyl phthalate - short-term reproduction assay with fathead minnow (<i>Pimephales promelas</i>) following OPPTS 890.1350 and OECD 229 guidelines.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Adult			
Health Outcome:	Behavioral			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	10064185			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	High	The source of the test substance was reported and the lot number was identified
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.3%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure	High	exposures were administered consistently across study groups
	Metric 9:	Administration Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The test organisms were adequately described and were obtained from an unnamed lab source
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
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Study Citation:	Viscient,, Smithers (2018). Di-n-butyl phthalate - short-term reproduction assay with fathead minnow (<i>Pimephales promelas</i>) following OPPTS 890.1350 and OECD 229 guidelines.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Adult			
Health Outcome:	Behavioral			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	10064185			
Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes
Additional Comments:	This report accounts for behavioral outcomes reported in the definitive test. This includes respiration rate is included in this as it was reported as a behavioral outcome.			

Overall Quality Determination**High**

Study Citation:	Viscient., Smithers (2018). Di-n-butyl phthalate - short-term reproduction assay with fathead minnow (<i>Pimephales promelas</i>) following OPPTS 890.1350 and OECD 229 guidelines.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Adult			
Health Outcome:	Endocrine			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	10064185			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	High	The source of the test substance was reported and the lot number was identified
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.3%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure	High	exposures were administered consistently across study groups
	Metric 9:	Administration Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The test organisms were adequately described and were obtained from an unnamed lab source
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
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Study Citation:	Viscient,, Smithers (2018). Di-n-butyl phthalate - short-term reproduction assay with fathead minnow (<i>Pimephales promelas</i>) following OPPTS 890.1350 and OECD 229 guidelines.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Adult
Health Outcome:	Endocrine
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	10064185

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
	Metric 20: Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: This form accounts for the thyroid histology results reported in the paper.

Overall Quality Determination

High

Study Citation:	Viscient., Smithers (2018). Di-n-butyl phthalate - short-term reproduction assay with fathead minnow (<i>Pimephales promelas</i>) following OPPTS 890.1350 and OECD 229 guidelines.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	10064185			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	High	The source of the test substance was reported and the lot number was identified
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.3%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure	High	exposures were administered consistently across study groups
	Metric 9:	Administration Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The test organisms were adequately described and were obtained from an unnamed lab source
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
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Study Citation:	Viscient,, Smithers (2018). Di-n-butyl phthalate - short-term reproduction assay with fathead minnow (<i>Pimephales promelas</i>) following OPPTS 890.1350 and OECD 229 guidelines.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	10064185			
Domain	Metric	Rating	Comments	
	Metric 18: Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions	
	Metric 20: Outcomes Unrelated to Exposure	High	there were no differences among groups	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical methods were adequately described	
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group	
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes	
Additional Comments:	None			

Overall Quality Determination**High**

Study Citation:	Viscient., Smithers (2018). Di-n-butyl phthalate - short-term reproduction assay with fathead minnow (<i>Pimephales promelas</i>) following OPPTS 890.1350 and OECD 229 guidelines.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Adult			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	10064185			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	High	The source of the test substance was reported and the lot number was identified
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.3%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure	High	exposures were administered consistently across study groups
	Metric 9:	Administration Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The test organisms were adequately described and were obtained from an unnamed lab source
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
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Study Citation:	Viscient,, Smithers (2018). Di-n-butyl phthalate - short-term reproduction assay with fathead minnow (<i>Pimephales promelas</i>) following OPPTS 890.1350 and OECD 229 guidelines.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Pimephales promelas</i> ; Adult			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	10064185			
Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes
Additional Comments: None				

Overall Quality Determination**High**

Study Citation:	Jee, J. H., Koo, J. G., Keum, Y. H., Park, K. H., Choi, S. H., Kang, J. C. (2009). Effects of dibutyl phthalate and di-ethylhexyl phthalate on acetylcholinesterase activity in bagrid catfish, <i>Pseudobagrus fulvidraco</i> (Richardson). Journal of Applied Ichthyology 25(6):771-775.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water, Food/Diet; Dietary			
Taxa, Species, Age:	Vertebrate; Fish; <i>Pseudobagrus fulvidraco</i> ; Adult			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1335887			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The chemical was identified by name.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	The chemical grade was reported as analytical.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was adequate.	
	Metric 6: Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were adequate for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure is via diet.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms used were appropriate for evaluation of the specific outcomes of interest.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions.	
	Metric 15: Number of Organisms and Replicates per Group	Low	The initial number of organisms was adequate. Replicates were not used.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	

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Study Citation:	Jee, J. H., Koo, J. G., Keum, Y. H., Park, K. H., Choi, S. H., Kang, J. C. (2009). Effects of dibutyl phthalate and di-ethylhexyl phthalate on acetylcholinesterase activity in bagrid catfish, <i>Pseudobagrus fulvidraco</i> (Richardson). Journal of Applied Ichthyology 25(6):771-775.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, Food/Diet; Dietary
Taxa, Species, Age:	Vertebrate; Fish; <i>Pseudobagrus fulvidraco</i> ; Adult
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	1335887

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	High	There were no differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This evaluation is for AChE activity.

Overall Quality Determination

High

Study Citation:	Jee, J. H., Koo, J. G., Keum, Y. H., Park, K. H., Choi, S. H., Kang, J. C. (2009). Effects of dibutyl phthalate and di-ethylhexyl phthalate on acetylcholinesterase activity in bagrid catfish, <i>Pseudobagrus fulvidraco</i> (Richardson). Journal of Applied Ichthyology 25(6):771-775.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water, Food/Diet; Dietary			
Taxa, Species, Age:	Vertebrate; Fish; <i>Pseudobagrus fulvidraco</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1335887			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	Chemical identified by name	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory	
	Metric 3: Test Substance Purity	High	Chemical grade reported as analytical	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was adequate	
	Metric 6: Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study group	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were adequate for a dose response	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure is via diet	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were appropriate for evaluation of the specific outcomes of interest	
	Metric 14: Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions	
	Metric 15: Number of Organisms and Replicates per Group	Low	Initial number was adequate, replicates were not used	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest	
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups	
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Study Citation:	Jee, J. H., Koo, J. G., Keum, Y. H., Park, K. H., Choi, S. H., Kang, J. C. (2009). Effects of dibutyl phthalate and di-ethylhexyl phthalate on acetylcholinesterase activity in bagrid catfish, <i>Pseudobagrus fulvidraco</i> (Richardson). Journal of Applied Ichthyology 25(6):771-775.		
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water, Food/Diet; Dietary		
Taxa, Species, Age:	Vertebrate; Fish; <i>Pseudobagrus fulvidraco</i> ; Adult		
Health Outcome:	Mortality		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	1335887		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
Metric 20:	Outcomes Unrelated to Exposure	High	There were no differences among groups
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	Low	Statistical analysis may have been performed but not described adequately
Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes
Additional Comments: None			
Overall Quality Determination		High	

Study Citation:	Jee, J. H., Koo, J. G., Keum, Y. H., Park, K. H., Choi, S. H., Kang, J. C. (2009). Effects of dibutyl phthalate and di-ethylhexyl phthalate on acetylcholinesterase activity in bagrid catfish, <i>Pseudobagrus fulvidraco</i> (Richardson). Journal of Applied Ichthyology 25(6):771-775.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water, Food/Diet; Dietary			
Taxa, Species, Age:	Vertebrate; Fish; <i>Pseudobagrus fulvidraco</i> ; Adult			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1335887			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	Chemical identified by name	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory	
	Metric 3: Test Substance Purity	High	Chemical grade reported as analytical	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was adequate	
	Metric 6: Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study group	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were adequate for a dose response	
	Metric 12: Testing at or Below Solubility Limit	N/A	exposure is via diet	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were appropriate for evaluation of the specific outcomes of interest	
	Metric 14: Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions	
	Metric 15: Number of Organisms and Replicates per Group	Low	Initial number was adequate, replicates were not used	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest	
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups	
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Study Citation:	Jee, J. H., Koo, J. G., Keum, Y. H., Park, K. H., Choi, S. H., Kang, J. C. (2009). Effects of dibutyl phthalate and di-ethylhexyl phthalate on acetylcholinesterase activity in bagrid catfish, <i>Pseudobagrus fulvidraco</i> (Richardson). Journal of Applied Ichthyology 25(6):771-775.		
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water, Food/Diet; Dietary		
Taxa, Species, Age:	Vertebrate; Fish; <i>Pseudobagrus fulvidraco</i> ; Adult		
Health Outcome:	Development/Growth		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	1335887		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
Metric 20:	Outcomes Unrelated to Exposure	High	There were no differences among groups
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	High	Statistical methods were adequately described
Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes
Additional Comments: None			
Overall Quality Determination		High	

Study Citation:	Ohtani, H., Miura, I., Ichikawa, Y. (2000). Effects of dibutyl phthalate as an environmental endocrine disruptor on gonadal sex differentiation of genetic males of the frog <i>Rana rugosa</i> . Environmental Health Perspectives 108(12):1189-1193.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Rana rugosa</i> ; Larvae			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	676307			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	High	The test substance was identified as being from Sigma.	
	Metric 3: Test Substance Purity	Low	Purity or grade of the test substance was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable for reported outcomes.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations. The exposure containers were "2L enameled containers."	
	Metric 8: Consistency of Exposure Administration	Low	The study provided few details on exposure administration.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source (bred in-lab from wild caught organisms).	
	Metric 14: Acclimatization and Pretreatment Conditions	Medium	All pretreatment conditions appeared to be the same for control and exposed organisms, but few details were provided.	
	Metric 15: Number of Organisms and Replicates per Group	Low	Fifty tadpoles per group were used, but no replicates were reported.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.	

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Study Citation:	Ohtani, H., Miura, I., Ichikawa, Y. (2000). Effects of dibutyl phthalate as an environmental endocrine disruptor on gonadal sex differentiation of genetic males of the frog <i>Rana rugosa</i> . Environmental Health Perspectives 108(12):1189-1193.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Rana rugosa</i> ; Larvae
Health Outcome:	Reproductive/Teratogenic
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	676307

Domain	Metric	Rating	Comments
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcomes of interest.
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described (Chi-sq test).
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: None

Overall Quality Determination**Medium**

Study Citation:	Adams, W. J., Biddinger, G. R., Robillard, K. A., Gorsuch, J. W. (1995). A summary of the acute toxicity of 14 phthalate esters to representative aquatic organisms. Environmental Toxicology and Chemistry 14(9):1569-1574.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Salmo mykiss</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1321996; Linked HERO ID(s): 1321996, 1316224			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The test substance nomenclature was reported without a CASRN.
	Metric 2:	Test Substance Source	Low	The test substance was provided by a manufacturer from commercially available batches. The manufacture's name and batch number were not provided. No analytical data was reported.
	Metric 3:	Test Substance Purity	High	
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	A negative control was reported.
	Metric 5:	Negative Control Response	High	The control response was acceptable.
	Metric 6:	Randomized Allocation	Low	An allocation method was not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The experimental system was well described. However, headspace or measures to prevent volatilization were not reported.
	Metric 8:	Consistency of Exposure Administration	High	Exposure administration was consistent across groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Sample extracts were analyzed by gas chromatography at the start and end of the test.
	Metric 10:	Exposure Duration and Frequency	High	The duration and frequency of exposure were appropriate for the test.
	Metric 11:	Number of Exposure Groups/Spacing of Exposure Levels	High	Exposure levels were appropriate. A range finding test was performed.
	Metric 12:	Testing at or Below Solubility Limit	High	The test was performed at or below the water solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Organisms were acclimated appropriately.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Each test was performed using duplicate test concentrations with ten organisms per test vessel.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions were appropriate for the test.
	Metric 17:	Outcome Assessment Methodology	High	The intended outcomes were reported.

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Study Citation:	Adams, W. J., Biddinger, G. R., Robillard, K. A., Gorsuch, J. W. (1995). A summary of the acute toxicity of 14 phthalate esters to representative aquatic organisms. Environmental Toxicology and Chemistry 14(9):1569-1574.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Salmo mykiss</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1321996; Linked HERO ID(s): 1321996, 1316224			
Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	High	Outcome assessment was consistent across groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	Environmental conditions were consistent across groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There were no reported differences between groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were performed and described.
	Metric 22:	Reporting of Data	Medium	Only treatment endpoints were reported.
	Metric 23:	Explanation of Unexpected Outcomes	High	No unexpected outcomes were reported.
Additional Comments: None				

Overall Quality Determination**High**

Study Citation:	Tollefsen, K. E., Meys, J. F., Frydenlund, J., Stenersen, J. (2002). Environmental estrogens interact with and modulate the properties of plasma sex steroid-binding proteins in juvenile Atlantic salmon (<i>Salmo salar</i>). Marine Environmental Research 54(3-5):697-701.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Salmo salar</i> ; Juvenile			
Health Outcome:	Mechanistic-Cell signaling/function-Receptor binding/ regulation of receptor activity-Endocrine toxicity-Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332592			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The test substance was identified by name only.	
	Metric 2: Test Substance Source	Low	The source of the DBP was not reported.	
	Metric 3: Test Substance Purity	Low	The purity of the DBP was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	The study authors reported using a negative control in which the vehicle 2-propanol only was used.	
	Metric 5: Negative Control Response	High	The negative control response was presented in Fig. 2 and was appropriate for the outcome of interest.	
	Metric 6: Randomized Allocation	Low	It was not reported how the Atlantic salmon were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The test system was described as flow-through, but little other details were provided on the test system. The preparation of the DBP concentrations was not reported. It was not reported whether the exposure vessels were made of glass or plastic.	
	Metric 8: Consistency of Exposure Administration	Low	Reporting omissions prevented evaluation of the consistency of the administration. Test chambers were not reported, nor were test volumes. Environmental conditions were not reported either. Ovrevik, Stenersen, Nilssen, Tollefsen (2002) was cited for methods.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if the test concentrations were measured at any point in the study.	
	Metric 10: Exposure Duration and Frequency	Medium	The exposure duration was reported to be two weeks. This appeared adequate to observe a response.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Medium	There were only three exposure groups, which is lower than is typical, but the spacing appeared adequate to observe a response.	
	Metric 12: Testing at or Below Solubility Limit	High	The test concentrations were reported in Fig. 2, and they were below the water solubility limit. It was reported the vehicle 2-propanol was used as well.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the Atlantic salmon was not reported. They were reported to be juveniles, but length and weight were not reported.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the fish were acclimated to test conditions prior to the study.	
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Study Citation:	Tollefsen, K. E., Meys, J. F., Frydenlund, J., Stenersen, J. (2002). Environmental estrogens interact with and modulate the properties of plasma sex steroid-binding proteins in juvenile Atlantic salmon (<i>Salmo salar</i>). Marine Environmental Research 54(3-5):697-701.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Salmo salar</i> ; Juvenile			
Health Outcome:	Mechanistic-Cell signaling/function-Receptor binding/ regulation of receptor activity-Endocrine toxicity-Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332592			
Domain	Metric	Rating	Comments	
	Metric 15: Number of Organisms and Replicates per Group	Low	The number of organisms per test chamber, and the number of replicates were not reported. Six to eight individual fish were used in each exposure group.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Little information was provided on the environmental conditions and the loading rate of the fish. It was not reported if the organisms were fed during the exposure.	
	Metric 17: Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported. Øvrevik, Stenersen, Nilssen, and Tollefsen (2001) and Tollefsen (2002) were cited for the outcome assessment methodology.	
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited. Øvrevik, Stenersen, Nilssen, and Tollefsen (2001) and Tollefsen (2002) were cited for the outcome assessment methodology.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	It was not reported if the fish were acclimated, nor were the environmental conditions reported.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Figure 2 reports that significance compared to the control group was analyzed using a one-way ANOVA test followed by a Dunnett's post test.	
	Metric 22: Reporting of Data	High	Exposure and control responses were reported in Fig 2 along with SEM. Fig 1 reported the displacement of [3H]estradiol from the high affinity estradiol binding site in diluted salmon plasma.	
	Metric 23: Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in the figures.	
Additional Comments:	This evaluation was on the effect of DBP on sex steroid binding proteins in Atlantic salmon and how this chemical modulates the endocrine system through the interaction with these proteins. Mechanistic outcomes for cell signaling/function, endocrine, receptor/binding, and reproduction were chosen as the outcomes of interest.			
Overall Quality Determination		Low		

Study Citation:	Battelle, (2018). 21-d amphibian metamorphosis assay (AMA) of dibutyl phthalate with African clawed frog, xenopus laevis.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Larvae			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	10064183			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	Chemical was identified by name and CAS#	
	Metric 2: Test Substance Source	High	The source and lot number of the test substance was reported, and a Certificate of Analysis was provided.	
	Metric 3: Test Substance Purity	High	Chemical purity reported as >=99%	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors used an appropriate concurrent negative control group for the range-finding test.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable for the range-finding test.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups for the range-finding test.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The report provided only limited details on the measures taken to appropriately prepare test concentrations for the range-finding test. It is unclear if vessel material was consistent between the range-finding test and the definitive test.	
	Metric 8: Consistency of Exposure Administration	Low	The study provided few details on exposure administration for the range-finding test.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured or measurements were not reported for the range-finding test.	
	Metric 10: Exposure Duration and Frequency	Medium	The duration of exposure was suitable for a range-finder	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response for a range-finding test.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Medium	The test organisms used in the range-finder were not adequately described but assumed to be same source as definitive test	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized	
	Metric 15: Number of Organisms and Replicates per Group	Low	10 organisms were tested per study group in the range-finder, but replication was not reported.	
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Study Citation:	Battelle, (2018). 21-d amphibian metamorphosis assay (AMA) of dibutyl phthalate with African clawed frog, xenopus laevis.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Larvae			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	10064183			
Domain	Metric	Rating	Comments	
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment for the range-finder were not reported.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions for the range-finder
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups for the range-finder
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Sufficient data were provided to conduct an independent statistical analysis.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group for the range-finder
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes
Additional Comments:	This form is evaluating the range-finding test conducted to determine the concentration of 1-BP that causes lethality and morbidity in <i>Xenopus laevis</i> .			
Overall Quality Determination		Medium		

Study Citation:	Battelle, (2018). 21-d amphibian metamorphosis assay (AMA) of dibutyl phthalate with African clawed frog, xenopus laevis.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Larvae			
Health Outcome:	Behavioral			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	10064183			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The chemical was identified by name and CAS#.	
	Metric 2: Test Substance Source	High	The source and lot number of the test substance was reported, and a Certificate of Analysis was provided.	
	Metric 3: Test Substance Purity	High	The chemical purity was reported as >=99%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors used an appropriate concurrent negative control group for the range-finding test.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable for the range-finding test.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups for the range-finding test.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The report provided only limited details on the measures taken to appropriately prepare test concentrations for the range-finding test. It is unclear if vessel material was consistent between the range-finding test and the definitive test.	
	Metric 8: Consistency of Exposure Administration	Low	The study provided few details on exposure administration for the range-finding test.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured or measurements were not reported for the range-finding test.	
	Metric 10: Exposure Duration and Frequency	Medium	The duration of exposure was suitable for a range-finding test.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response for a range-finding test.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Medium	The test organisms used in the range-finder were not adequately described but assumed to be from the same source as the definitive test.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.	
	Metric 15: Number of Organisms and Replicates per Group	Low	Ten organisms were tested per study group in the range-finder, but replication was not reported.	
Domain 5: Outcome Assessment				
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Study Citation:	Battelle, (2018). 21-d amphibian metamorphosis assay (AMA) of dibutyl phthalate with African clawed frog, xenopus laevis.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Larvae			
Health Outcome:	Behavioral			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	10064183			
Domain	Metric	Rating	Comments	
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment for the range-finder were not reported.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions for the range-finder.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups for the range-finder.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Sufficient data were provided to conduct an independent statistical analysis.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group for the range-finder.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments:	This form is for the behavioral outcome reported for the preliminary test.			
Overall Quality Determination		Medium		

Study Citation:	Gardner, S. T., Wood, A. T., Lester, R., Onkst, P. E., Burnham, N., Perygin, D. H., Rayburn, J. (2016). Assessing differences in toxicity and teratogenicity of three phthalates, Diethyl phthalate, Di-n-propyl phthalate, and Di-n-butyl phthalate, using <i>Xenopus laevis</i> embryos. Journal of Toxicology and Environmental Health, Part A: Current Issues 79(2):71-82.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Embryo			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3070743			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The test substance was synthesized by a source other than the manufacturer.	
	Metric 3: Test Substance Purity	Low	Purity or grade of the test substance was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	Low	The biological response of the negative control group was not reported.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	Seven concentrations with a suitable range were used for the test.	
	Metric 12: Testing at or Below Solubility Limit	High	The solvent concentration was appropriate.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	Two replicates of twenty embryos was suitable.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate. pH was monitored but not reported.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	

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Study Citation:	Gardner, S. T., Wood, A. T., Lester, R., Onkst, P. E., Burnham, N., Perygin, D. H., Rayburn, J. (2016). Assessing differences in toxicity and teratogenicity of three phthalates, Diethyl phthalate, Di-n-propyl phthalate, and Di-n-butyl phthalate, using <i>Xenopus laevis</i> embryos. Journal of Toxicology and Environmental Health, Part A: Current Issues 79(2):71-82.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Embryo			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3070743			
Domain	Metric	Rating	Comments	
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.	
	Metric 22: Reporting of Data	High	Data for exposure-related findings were shown for each treatment and control group in graphs.	
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.	
Additional Comments:	Malformations, length			
Overall Quality Determination		Medium		

Study Citation:	Gardner, S. T., Wood, A. T., Lester, R., Onkst, P. E., Burnham, N., Perygin, D. H., Rayburn, J. (2016). Assessing differences in toxicity and teratogenicity of three phthalates, Diethyl phthalate, Di-n-propyl phthalate, and Di-n-butyl phthalate, using <i>Xenopus laevis</i> embryos. Journal of Toxicology and Environmental Health, Part A: Current Issues 79(2):71-82.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Embryo			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3070743			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	Chemical was identified by name only	
	Metric 2: Test Substance Source	High	The test substance was synthesized by a source other than the manufacturer	
	Metric 3: Test Substance Purity	Low	Purity or grade of test substance was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group	
	Metric 5: Negative Control Response	Low	The biological response of the negative control groups was not reported.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail	
	Metric 8: Consistency of Exposure Administration	High	exposures were administered consistently across study groups	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	7 concentrations with a suitable range	
	Metric 12: Testing at or Below Solubility Limit	High	The solvent concentration was appropriate	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source	
	Metric 14: Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms	
	Metric 15: Number of Organisms and Replicates per Group	Medium	Two replicates of twenty embryos was suitable	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate, pH was monitored but not reported	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest	
	Metric 18: Consistency of Outcome Assessment	Medium	Details regarding the execution of the study protocol for outcome assessment were limited but sufficient	

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Study Citation:	Gardner, S. T., Wood, A. T., Lester, R., Onkst, P. E., Burnham, N., Perygin, D. H., Rayburn, J. (2016). Assessing differences in toxicity and teratogenicity of three phthalates, Diethyl phthalate, Di-n-propyl phthalate, and Di-n-butyl phthalate, using <i>Xenopus laevis</i> embryos. Journal of Toxicology and Environmental Health, Part A: Current Issues 79(2):71-82.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Embryo
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	3070743

Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Medium	The study did not provide enough information to allow a comparison of environmental conditions. However, this unlikely would have had substantial impacts on results.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described
	Metric 22: Reporting of Data	Low	Data for exposure-related findings were shown for each treatment and control group in graphs
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: Other phthalates evaluated were diethyl (DEP), di-n-propyl (DnPP), and di-n-butyl (DBP) phthalate. Only DBP is included for purposes of an EPA HPS.

Overall Quality Determination

Medium

Study Citation:	Lee, S. K., Owens, G. A., Veeramachaneni, D. N. (2005). Exposure to low concentrations of di-n-butyl phthalate during embryogenesis reduces survival and impairs development of <i>Xenopus laevis</i> frogs. Journal of Toxicology and Environmental Health, Part A: Current Issues 68(10):763-772.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Embryo			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	673293			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The DBP was identified by CAS number.	
	Metric 2: Test Substance Source	Low	The DBP was reported to be from Sigma Aldrich and a lot number was provided, but it was not reported if it was analytically verified.	
	Metric 3: Test Substance Purity	High	The purity was reported to be 99.8%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a DMSO solvent control as well as a negative control using FETAX solution.	
	Metric 5: Negative Control Response	High	The solvent control response and the negative control response are reported in Table1.	
	Metric 6: Randomized Allocation	Medium	Embryos were randomly assigned to treatment and control groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	It was reported that stock solutions were prepared every 48h by diluting DBP into DMSO in amber glass containers. The solutions were then vortexed and refrigerated until use. Final concentrations were made every 24h by adding the stock solution to the FETAX solution in appropriate amounts.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered in 100mm pyrex petri dishes with 20mL of solution and 50 embryos in each test chamber. Tests were administered for 96h until stage 46.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if exposure concentrations were measured at any point in the study.	
	Metric 10: Exposure Duration and Frequency	High	The exposure duration was appropriate for the study and produced a dose response.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	There were 6 exposure concentrations as well as a negative control and a solvent control. Spacing of exposure concentrations was adequate for a response.	
	Metric 12: Testing at or Below Solubility Limit	High	0.01% DMSO was used as a solvent in the appropriate amount for this study. A solvent control was used as well.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	Adult broodstock was reported to be from Nasco in Fort Atkinson, WI. The adults were bred and embryos ranging from stages 8-11 were collected from them.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the embryos were acclimated to test conditions in any way,	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 50 embryos per test chamber, and all treatments had a total of 300 embryos. The negative control had 600 embryos. All tests were completed in duplicate.	

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Study Citation:	Lee, S. K., Owens, G. A., Veeramachaneni, D. N. (2005). Exposure to low concentrations of di-n-butyl phthalate during embryogenesis reduces survival and impairs development of <i>Xenopus laevis</i> frogs. Journal of Toxicology and Environmental Health, Part A: Current Issues 68(10):763-772.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Embryo
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	673293

Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	The embryos were kept at 24C for the duration of the study. They did not need to be fed. Adult frogs were fed pellets and were kept at 20-24C in 10mMNaCl. The photoperiod was 12L:12D.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—development/growth of embryos as they developed into tadpoles (stage 46).
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. After the exposure, tadpoles were euthanized and refrigerated. They were then examined under a dissecting scope for abnormalities and stage. Length was determined by photographing each tadpole.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	It was not reported if any acclimation occurred.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical analysis was discussed in the "Statistical Analyses" section of the article.
	Metric 22: Reporting of Data	High	Data for the negative control, the solvent control, and all exposure levels was reported in Table 1. Figure 1 contained data for the control and the exposure concentrations.
	Metric 23: Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Mean SE for body length was reported in Table 1.
Additional Comments: This portion of the evaluation was on the effect of DBP on the growth and development of <i>X. laevis</i> embryos into tadpoles. Development/growth was chosen as the outcome according to the Eco outcomes document as the development as well as deformities were recorded as endpoints.			

Overall Quality Determination**High**

Study Citation:	Lee, S. K., Owens, G. A., Veeramachaneni, D. N. (2005). Exposure to low concentrations of di-n-butyl phthalate during embryogenesis reduces survivability and impairs development of <i>Xenopus laevis</i> frogs. Journal of Toxicology and Environmental Health, Part A: Current Issues 68(10):763-772.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Embryo			
Health Outcome:	Behavioral			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	673293			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The DBP was identified by CAS number.	
	Metric 2: Test Substance Source	Low	The DBP was reported to be from Sigma Aldrich and a lot number was provided, but it was not reported if it was analytically verified.	
	Metric 3: Test Substance Purity	High	The purity was reported to be 99.8%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a DMSO solvent control as well as a negative control using FETAX solution.	
	Metric 5: Negative Control Response	Low	The behavior of the control group was not reported.	
	Metric 6: Randomized Allocation	Medium	Embryos were randomly assigned to treatment and control groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	It was reported that stock solutions were prepared every 48h by diluting DBP into DMSO in amber glass containers. The solutions were then vortexed and refrigerated until use. Final concentrations were made every 24h by adding the stock solution to the FETAX solution in appropriate amounts.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered in 100mm pyrex petri dishes with 20mL of solution and 50 embryos in each test chamber. Tests were administered for 96h until stage 46.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if exposure concentrations were measured at any point in the study.	
	Metric 10: Exposure Duration and Frequency	High	The exposure duration was appropriate for the study and produced a dose response.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	There were six exposure concentrations as well as a negative control and a solvent control. Spacing of exposure concentrations was adequate for a response.	
	Metric 12: Testing at or Below Solubility Limit	High	DMSO (0.01%) was used as a solvent in the appropriate amount for this study. A solvent control was used as well.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	Adult broodstock was reported to be from Nasco in Fort Atkinson, WI. The adults were bred and embryos ranging from stages 8-11 were collected from them.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the embryos were acclimated to test conditions in any way.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 50 embryos per test chamber, and all treatments had a total of 300 embryos. The negative control had 600 embryos. All tests were completed in duplicate.	
Domain 5: Outcome Assessment				
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Study Citation:	Lee, S. K., Owens, G. A., Veeramachaneni, D. N. (2005). Exposure to low concentrations of di-n-butyl phthalate during embryogenesis reduces survivability and impairs development of <i>Xenopus laevis</i> frogs. Journal of Toxicology and Environmental Health, Part A: Current Issues 68(10):763-772.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Embryo
Health Outcome:	Behavioral
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	673293

Domain	Metric	Rating	Comments
	Metric 16: Adequacy of Test Conditions	High	The embryos were kept at 24C for the duration of the study. They did not need to be fed. Adult frogs were fed pellets and were kept at 20-24C in 10mMNaCl. The photoperiod was 12L:12D.
	Metric 17: Outcome Assessment Methodology	Low	Behavior assessment methodology was not reported.
	Metric 18: Consistency of Outcome Assessment	Low	It was not reported how behavior was assessed during the exposure.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	It was not reported if any acclimation occurred.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical analysis was discussed in the "Statistical Analyses" section of the article.
	Metric 22: Reporting of Data	Medium	Behavior results were briefly described in the results section and a little bit in the discussion section, but no behavior data was shown in tables or figures.
	Metric 23: Explanation of Unexpected Outcomes	Medium	Study authors did not report any unexpected outcomes. No variability was given for the behavioral results.
Additional Comments: This portion of the evaluation was on the effect of DBP on the behavior of <i>X. laevis</i> embryos.			

Overall Quality Determination**Medium**

Study Citation:	Lee, S. K., Owens, G. A., Veeramachaneni, D. N. (2005). Exposure to low concentrations of di-n-butyl phthalate during embryogenesis reduces survival and impairs development of <i>Xenopus laevis</i> frogs. Journal of Toxicology and Environmental Health, Part A: Current Issues 68(10):763-772.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Embryo			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	673293			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The DBP was identified by CAS number.	
	Metric 2: Test Substance Source	Low	The DBP was reported to be from Sigma Aldrich and a lot number was provided, but it was not reported if it was analytically verified.	
	Metric 3: Test Substance Purity	High	The purity was reported to be 99.8%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a DMSO solvent control as well as a negative control using FETAX solution.	
	Metric 5: Negative Control Response	High	The solvent control response and the negative control response are reported in Table1.	
	Metric 6: Randomized Allocation	Medium	Embryos were randomly assigned to treatment and control groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	It was reported that stock solutions were prepared every 48h by diluting DBP into DMSO in amber glass containers. The solutions were then vortexed and refrigerated until use. Final concentrations were made every 24h by adding the stock solution to the FETAX solution in appropriate amounts.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered in 100mm pyrex petri dishes with 20mL of solution and 50 embryos in each test chamber. Tests were administered for 96h until stage 46.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if exposure concentrations were measured at any point in the study.	
	Metric 10: Exposure Duration and Frequency	High	The exposure duration was appropriate for the study and produced a dose response.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	There were 6 exposure concentrations as well as a negative control and a solvent control. Spacing of exposure concentrations was adequate for a response.	
	Metric 12: Testing at or Below Solubility Limit	High	0.01% DMSO was used as a solvent in the appropriate amount for this study. A solvent control was used as well.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	Adult broodstock was reported to be from Nasco in Fort Atkinson, WI. The adults were bred and embryos ranging from stages 8-11 were collected from them.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the embryos were acclimated to test conditions in any way,	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 50 embryos per test chamber, and all treatments had a total of 300 embryos. The negative control had 600 embryos. All tests were completed in duplicate.	
Domain 5: Outcome Assessment				
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Study Citation:	Lee, S. K., Owens, G. A., Veeramachaneni, D. N. (2005). Exposure to low concentrations of di-n-butyl phthalate during embryogenesis reduces survival and impairs development of <i>Xenopus laevis</i> frogs. Journal of Toxicology and Environmental Health, Part A: Current Issues 68(10):763-772.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Embryo			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	673293			
Domain	Metric	Rating	Comments	
	Metric 16: Adequacy of Test Conditions	High	The embryos were kept at 24C for the duration of the study. They did not need to be fed. Adult frogs were fed pellets and were kept at 20-24C in 10mMNaCl. The photoperiod was 12L:12D.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest–mortality. LC50 values were determined, and percent mortality was recorded.	
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. Dead embryos were removed and counted every 24h when solutions were changed.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	It was not reported if any acclimation occurred.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical analysis was discussed in the "Statistical Analyses" section of the article.	
	Metric 22: Reporting of Data	High	Data for the negative control, the solvent control, and all exposure levels was reported in Table 1.	
	Metric 23: Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.	
Additional Comments:	This portion of the evaluation was on the effect of DBP on mortality of <i>X. laevis</i> embryos. Mortality was monitored every 24h and was therefore chosen as an outcome for the study.			
Overall Quality Determination		High		

Study Citation:	Xu, Y., Gye, M. C. (2018). Developmental toxicity of dibutyl phthalate and citrate ester plasticizers in <i>Xenopus laevis</i> embryos. Chemosphere 204:523-534.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Embryo			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	4829262			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The test substance was identified by name only.	
	Metric 2: Test Substance Source	Low	The source was reported to be Sigma Aldrich in Seoul, Korea, but it was not reported if it was analytically verified.	
	Metric 3: Test Substance Purity	High	The purity was reported to be 99%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of a concurrent negative control.	
	Metric 5: Negative Control Response	High	The negative control response was reported in Table 2 and Figures 1 and 2 and was adequate for the outcome of interest.	
	Metric 6: Randomized Allocation	Low	It was not reported how the embryos were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The system was a static renewal system with renewals every 24h. DMSO was used to prepare the stock solutions, and serial dilutions were used to prepare the test concentrations with FETAX used as the diluent.	
	Metric 8: Consistency of Exposure Administration	Medium	Well plates were reported to be use, but it was unclear what size they were. There were four plates with 40 embryos for each treatment. Treatments were for 48h.	
	Metric 9: Measurement of Test Substance Concentration	High	Measured concentrations were reported in parenthesis in ppm in the "materials and methods" section.	
	Metric 10: Exposure Duration and Frequency	High	The exposure duration was reported to be 96h, which was adequate for the outcomes of interest.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	There were six exposure groups and the spacing was appropriate to observe a response.	
	Metric 12: Testing at or Below Solubility Limit	High	DMSO was used as a vehicle solvent at an appropriate level.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The <i>Xenopus laevis</i> were bred at Hanyang University Aquarium. Embryos were used for this study.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	Organisms were bred at the performing laboratory, and lab conditions were appropriate.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were four wells that contained 40 embryos for each treatment.	
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Study Citation:	Xu, Y., Gye, M. C. (2018). Developmental toxicity of dibutyl phthalate and citrate ester plasticizers in <i>Xenopus laevis</i> embryos. Chemosphere 204:523-534.		
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Embryo		
Health Outcome:	Development/Growth		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	4829262		
Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Embryos were kept in well plates with 10 embryos each at 23C. FETAX solution was used as the embryo rearing solution and diluent. The solution was renewed daily for the duration of the test.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—embryo growth and malformations.
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. MZ8: Leica, Heerbrugg, Switzerland was cited for malformations and Nieuwkoop and Faber for staging.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were described in detail in the "statistical analysis" section.
	Metric 22: Reporting of Data	High	Data for the control and the exposure responses were reported in Table 2 and in Figures 1 and 2.
	Metric 23: Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability.
Additional Comments:	This portion of the evaluation was on the effect of DBP on <i>X. laevis</i> embryo length and malformation. Development and growth was selected as the outcome of interest. MZ8: Leica, Heerbrugg, Switzerland was cited for malformations, and Nieuwkoop and Faber was cited for developmental staging.		
Overall Quality Determination		High	

Study Citation:	Xu, Y., Gye, M. C. (2018). Developmental toxicity of dibutyl phthalate and citrate ester plasticizers in <i>Xenopus laevis</i> embryos. Chemosphere 204:523-534.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Embryo			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Genotox (including DNA repair)-Oxidative stress (including redox biology)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	4829262			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The test substance was identified by name only.	
	Metric 2: Test Substance Source	Low	The source was reported to be Sigma Aldrich in Seoul, Korea, but it was not reported if it was analytically verified.	
	Metric 3: Test Substance Purity	High	The purity was reported to be 99%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of a concurrent negative control.	
	Metric 5: Negative Control Response	High	The negative control response was reported in Figures 3-5 and was adequate for the outcomes of interest.	
	Metric 6: Randomized Allocation	Low	It was not reported how the embryos were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The system was a static renewal system with renewals every 24h. DMSO was used to prepare the stock solutions, and serial dilutions were used to prepare the test concentrations with FETAX used as the diluent.	
	Metric 8: Consistency of Exposure Administration	Medium	Well plates were reported to be use, but it was unclear what size they were. There were 4 plates with 40 embryos for each treatment. Treatments were for 48h.	
	Metric 9: Measurement of Test Substance Concentration	High	Measured concentrations were reported in parenthesis in ppm in the "materials and methods" section.	
	Metric 10: Exposure Duration and Frequency	High	The exposure duration was reported to be 96h, which was adequate for the outcomes of interest.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	There were 6 exposure groups and the spacing was appropriate to observe a response.	
	Metric 12: Testing at or Below Solubility Limit	High	DMSO was used as a vehicle solvent at an appropriate level.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The <i>Xenopus laevis</i> were bred at Hanyang University Aquarium. Embryos were used for this study.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	Organisms were bred at the performing laboratory, and lab conditions were appropriate.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 4 wells that contained 40 embryos for each treatment.	
Domain 5: Outcome Assessment				
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Study Citation:	Xu, Y., Gye, M. C. (2018). Developmental toxicity of dibutyl phthalate and citrate ester plasticizers in <i>Xenopus laevis</i> embryos. Chemosphere 204:523-534.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Embryo			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Genotox (including DNA repair)-Oxidative stress (including redox biology)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	4829262			
Domain	Metric	Rating	Comments	
	Metric 16: Adequacy of Test Conditions	High	Embryos were kept in well plates with 10 embryos each at 23C. FETAX solution was used as the embryo rearing solution and diluent. The solution was renewed daily for the duration of the test.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest–DNA fragmentation, oxidative stress reaction, mRNA levels.	
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical methods were described in detail in the "statistical analysis" section.	
	Metric 22: Reporting of Data	High	Data for the control and the exposure responses were reported in Figures 3-5 and were appropriate for the outcomes of interest.	
	Metric 23: Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in the figures.	
Additional Comments:	This portion of the evaluation was on the effect of DBP on <i>X. laevis</i> embryos. Mechanistic outcomes for biomarkers, genotox, and oxidative stress were chosen as the outcomes of interest.			

Overall Quality Determination**High**

Study Citation:	Xu, Y., Gye, M. C. (2018). Developmental toxicity of dibutyl phthalate and citrate ester plasticizers in <i>Xenopus laevis</i> embryos. Chemosphere 204:523-534.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Embryo			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	4829262			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The test substance was identified by name only.	
	Metric 2: Test Substance Source	Low	The source was reported to be Sigma Aldrich in Seoul, Korea, but it was not reported if it was analytically verified.	
	Metric 3: Test Substance Purity	High	The purity was reported to be 99%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of a concurrent negative control.	
	Metric 5: Negative Control Response	High	The negative control response was reported in Table 1 and was adequate for the outcome of interest.	
	Metric 6: Randomized Allocation	Low	It was not reported how the embryos were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The system was a static renewal system with renewals every 24h. DMSO was used to prepare the stock solutions, and serial dilutions were used to prepare the test concentrations with FETAX used as the diluent.	
	Metric 8: Consistency of Exposure Administration	Medium	Well plates were reported to be use, but it was unclear what size they were. There were 4 plates with 40 embryos for each treatment. Treatments were for 48h.	
	Metric 9: Measurement of Test Substance Concentration	High	Measured concentrations were reported in parenthesis in ppm in the "materials and methods" section.	
	Metric 10: Exposure Duration and Frequency	High	The exposure duration was reported to be 96h, which was adequate for the outcomes of interest.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	There were 6 exposure groups and the spacing was appropriate to observe a response.	
	Metric 12: Testing at or Below Solubility Limit	High	DMSO was used as a vehicle solvent at an appropriate level.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The <i>Xenopus laevis</i> were bred at Hanyang University Aquarium. Embryos were used for this study.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	Organisms were bred at the performing laboratory, and lab conditions were appropriate.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 4 wells that contained 40 embryos for each treatment.	
Domain 5: Outcome Assessment				
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Study Citation:	Xu, Y., Gye, M. C. (2018). Developmental toxicity of dibutyl phthalate and citrate ester plasticizers in <i>Xenopus laevis</i> embryos. Chemosphere 204:523-534.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Embryo			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	4829262			
Domain		Metric	Rating	Comments
	Metric 16:	Adequacy of Test Conditions	High	Embryos were kept in well plates with 10 embryos each at 23C. FETAX solution was used as the embryo rearing solution and diluent. The solution was renewed daily for the duration of the test.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest–mortality in the form of LC50 values.
	Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. Embryos were assessed every 24h and any dead embryos were removed.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were described in detail in the "statistical analysis" section.
	Metric 22:	Reporting of Data	High	Data for the control and the exposure responses were reported in Table 1.
	Metric 23:	Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability. 95% confidence intervals were not provided.
Additional Comments:	This portion of the evaluation was on the effect of DBP on <i>X. laevis</i> embryos. LC50 values were reported, so mortality was selected as the outcome of interest.			
Overall Quality Determination			High	

Study Citation:	Battelle, (2018). 21-d amphibian metamorphosis assay (AMA) of dibutyl phthalate with African clawed frog, xenopus laevis.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Larvae			
Health Outcome:	Behavioral			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	10064183			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	Chemical was identified by name, CAS#, and structure.	
	Metric 2: Test Substance Source	High	The source and lot number of the test substance was reported, and a Certificate of Analysis was provided.	
	Metric 3: Test Substance Purity	High	Chemical purity reported as >=99%	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and acceptable.	
	Metric 6: Randomized Allocation	Medium	Organisms were randomly allocated to tanks, and tanks were randomly allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were suitable and described in adequate detail. Exposure water-contact components included glass, stainless steel, and Teflon.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups	
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source and were appropriate for use in this study.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Test conditions were conducive to maintenance of organism health	

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Study Citation:	Battelle, (2018). 21-d amphibian metamorphosis assay (AMA) of dibutyl phthalate with African clawed frog, xenopus laevis.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Larvae			
Health Outcome:	Behavioral			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	10064183			
Domain	Metric	Rating	Comments	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest	
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions	
	Metric 20: Outcomes Unrelated to Exposure	High	There were no differences among groups that could influence the outcome assessment.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical methods were adequately described and were appropriate.	
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group	
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes	
Additional Comments:	This form is for the behavioral outcomes reported in the text for the definitive test.			

Overall Quality Determination**High**

Study Citation:	Battelle, (2018). 21-d amphibian metamorphosis assay (AMA) of dibutyl phthalate with African clawed frog, xenopus laevis.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Larvae			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	10064183			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	Chemical was identified by name, CAS#, and structure.	
	Metric 2: Test Substance Source	High	The source and lot number of the test substance was reported, and a Certificate of Analysis was provided.	
	Metric 3: Test Substance Purity	High	Chemical purity reported as >=99%	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and acceptable.	
	Metric 6: Randomized Allocation	Medium	Organisms were randomly allocated to tanks, and tanks were randomly allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were suitable and described in adequate detail. Exposure water-contact components included glass, stainless steel, and Teflon.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups	
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source and were appropriate for use in this study.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Test conditions were conducive to maintenance of organism health	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest	
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Study Citation:	Battelle, (2018). 21-d amphibian metamorphosis assay (AMA) of dibutyl phthalate with African clawed frog, xenopus laevis.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Larvae			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	10064183			
Domain	Metric	Rating	Comments	
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions	
	Metric 20: Outcomes Unrelated to Exposure	High	There were no differences among groups that could influence the outcome assessment.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical methods were adequately described and were appropriate.	
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group	
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes	
Additional Comments:	None			

Overall Quality Determination**High**

Study Citation:	Battelle, (2018). 21-d amphibian metamorphosis assay (AMA) of dibutyl phthalate with African clawed frog, xenopus laevis.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Larvae			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	10064183			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	Chemical was identified by name, CAS#, and structure.	
	Metric 2: Test Substance Source	High	The source and lot number of the test substance was reported, and a Certificate of Analysis was provided.	
	Metric 3: Test Substance Purity	High	Chemical purity reported as >=99%	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable	
	Metric 6: Randomized Allocation	Medium	Organisms were randomly allocated to tanks, and tanks were randomly allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were suitable and described in adequate detail. Exposure water-contact components included glass, stainless steel, and Teflon.	
	Metric 8: Consistency of Exposure	High	exposures were administered consistently across study groups	
	Metric 9: Administration Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source and were appropriate for use in this study.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Test conditions were conducive to maintenance of organism health	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest	
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Study Citation:	Battelle, (2018). 21-d amphibian metamorphosis assay (AMA) of dibutyl phthalate with African clawed frog, xenopus laevis.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Larvae			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	10064183			
Domain	Metric	Rating	Comments	
	Metric 18: Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions	
	Metric 20: Outcomes Unrelated to Exposure	High	there were no differences among groups that could influence the outcome assessment.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical methods were adequately described and were appropriate.	
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group	
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes	
Additional Comments:	None			

Overall Quality Determination**High**

Study Citation:	Battelle, (2018). 21-d amphibian metamorphosis assay (AMA) of dibutyl phthalate with African clawed frog, <i>Xenopus laevis</i> .		
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Larvae		
Health Outcome:	Endocrine		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	10064183		
Domain	Metric	Rating	Comments
Domain 1: Test Substance			
	Metric 1: Test Substance Identity	High	The chemical was identified by name, CAS#, and structure.
	Metric 2: Test Substance Source	High	The source and lot number of the test substance was reported, and a Certificate of Analysis was provided.
	Metric 3: Test Substance Purity	High	Chemical purity was reported as $\geq 99\%$.
Domain 2: Test Design			
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable.
	Metric 6: Randomized Allocation	Medium	Organisms were randomly allocated to tanks, and tanks were randomly allocated to study groups.
Domain 3: Exposure Characterization			
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were suitable and described in adequate detail. Exposure water-contact components included glass, stainless steel, and teflon.
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods.
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type.
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit.
Domain 4: Test Organism			
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source, and they were appropriate for use in this study.
	Metric 14: Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions.
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
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Study Citation:	Battelle, (2018). 21-d amphibian metamorphosis assay (AMA) of dibutyl phthalate with African clawed frog, xenopus laevis.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Larvae			
Health Outcome:	Endocrine			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	10064183			
Domain	Metric	Rating	Comments	
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.	
	Metric 20: Outcomes Unrelated to Exposure	High	There were no differences among groups that could influence the outcome assessment.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical methods were adequately described and were appropriate.	
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.	
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.	
Additional Comments:	Thyroid gland histopathology was conducted in this study and evaluation included thyroid gland hypertrophy/atrophy, follicular cell hypertrophy, follicular cell hyperplasia, follicular lumen area, colloid quality and follicular cell height/shape.			

Overall Quality Determination**High**

Study Citation:	Lee, S. K., Veeramachaneni, R., D.N. (2005). Subchronic exposure to low concentrations of di-n-butyl phthalate disrupts spermatogenesis in <i>Xenopus laevis</i> frogs. <i>Toxicological Sciences</i> 84(2):394-407.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Larvae			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	128004			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The DBP was identified by CASRN.	
	Metric 2: Test Substance Source	Low	The source was reported to be Sigma Aldrich, and the lot number was reported, but it was not reported if the DBP was analytically verified.	
	Metric 3: Test Substance Purity	High	The DBP was reported to be 99.8% pure.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a negative control in the FETAX solution and a negative control containing 0.01% DMSO.	
	Metric 5: Negative Control Response	High	The biological response of the negative control was reported in Table 1.	
	Metric 6: Randomized Allocation	Medium	It was reported that tadpoles were randomly assigned to treatment groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	Stock solutions were prepared in 100mL amber glass bottles weekly by adding DBP directly to DMSO. Glass pipettes were used to add the proper quantity to the test tanks. It was then manually stirred. Renewals were conducted every M-W-F.	
	Metric 8: Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups. The same solvent was used in all test concentrations and in the solvent control. All test concentrations were run until complete metamorphosis of the control organisms.	
	Metric 9: Measurement of Test Substance Concentration	Low	Study authors did not report if the exposure concentrations were measured.	
	Metric 10: Exposure Duration and Frequency	High	The exposure duration was reported to be from stage 52 (3wks) to stage 66 (8wks). This is typical of amphibian studies.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	There were five exposure levels as well as a negative control and a solvent control. This was adequate to see a response.	
	Metric 12: Testing at or Below Solubility Limit	High	A solvent was used in this study, and the solvent control response was adequate, indicating the concentration was appropriate.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	Breeding adults were reported to be from Nasco in Fort Atkinson, WI. Adults were bred in the laboratory performing the study, and the tadpoles were reared there. The tadpole stages were appropriate for the study.	
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Study Citation:	Lee, S. K., Veeramachaneni, R., D.N. (2005). Subchronic exposure to low concentrations of di-n-butyl phthalate disrupts spermatogenesis in <i>Xenopus laevis</i> frogs. <i>Toxicological Sciences</i> 84(2):394-407.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Larvae
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	128004

Domain	Metric	Rating	Comments
	Metric 14: Acclimatization and Pretreatment Conditions	High	Pretreatment conditions were the same as treatment conditions. Temperature and photoperiod were the same for each.
	Metric 15: Number of Organisms and Replicates per Group	Low	The number of test organisms for each test concentration were reported, but the number of replicates for each test concentration was not reported.
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Tadpoles were housed at a loading rate of 2.5 tadpoles/L. They were kept at 20-24C with a 12:12 photoperiod. The organisms were fed daily after 96h in age throughout the study. A slurry was used for the tadpoles and pellets were used after metamorphosis.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—body weights/development/growth.
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	StatView was used for statistical analysis. ANOVA was used as well as Tukey-Kramer post hoc test.
	Metric 22: Reporting of Data	High	Data for exposure related findings including control results can be found in Table 1.
	Metric 23: Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in Table 1.
Additional Comments:	This portion of the evaluation was on the effect of DBP on body weight in <i>Xenopus laevis</i> . Body weights for each treatment were obtained, so the development/growth outcome was chosen.		

Overall Quality Determination**High**

Study Citation:	Lee, S. K., Veeramachaneni, R., D.N. (2005). Subchronic exposure to low concentrations of di-n-butyl phthalate disrupts spermatogenesis in <i>Xenopus laevis</i> frogs. <i>Toxicological Sciences</i> 84(2):394-407.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Larvae			
Health Outcome:	Mechanistic-Endocrine toxicity			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	128004			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The DBP was identified by CASRN.	
	Metric 2: Test Substance Source	Low	The source was reported to be Sigma Aldrich, and the lot number was reported, but it was not reported if the DBP was analytically verified.	
	Metric 3: Test Substance Purity	High	The DBP was reported to be 99.8% pure.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a negative control in the FETAX solution and a negative control containing 0.01% DMSO.	
	Metric 5: Negative Control Response	High	The biological response of the negative controls was reported in the text under results.	
	Metric 6: Randomized Allocation	Medium	It was reported that tadpoles were randomly assigned to treatment groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	Stock solutions were prepared in 100mL amber glass bottles weekly by adding DBP directly to DMSO. Glass pipettes were used to add the proper quantity to the test tanks. It was then manually stirred. Renewal were conducted every M-W-F.	
	Metric 8: Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups. The same solvent was used in all test concentrations and in the solvent control. All test concentrations were run until complete metamorphosis of the control organisms.	
	Metric 9: Measurement of Test Substance Concentration	Low	Study authors did not report if the exposure concentrations were measured.	
	Metric 10: Exposure Duration and Frequency	High	The exposure duration was reported to be from stage 52 (3wks) to stage 66 (8wks). This is typical of amphibian studies.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	There were 5 exposure levels as well as a negative control and a solvent control. This was adequate to see a response.	
	Metric 12: Testing at or Below Solubility Limit	High	A solvent was used in this study, and the solvent control response was adequate indicating the concentration was appropriate.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	Breeding adults were reported to be from Nasco in Fort Atkinson, WI. Adults were bred in the laboratory performing the study, and the tadpoles were reared there. The tadpole stages were appropriate for the study.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	Pretreatment conditions were the same as treatment conditions. Temperature and photoperiod were the same for each.	
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Study Citation:	Lee, S. K., Veeramachaneni, R., D.N. (2005). Subchronic exposure to low concentrations of di-n-butyl phthalate disrupts spermatogenesis in <i>Xenopus laevis</i> frogs. <i>Toxicological Sciences</i> 84(2):394-407.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Larvae
Health Outcome:	Mechanistic-Endocrine toxicity
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	128004

Domain	Metric	Rating	Comments
	Metric 15: Number of Organisms and Replicates per Group	Low	The number of test organisms for each test concentration were reported, but the number of replicates for each test concentration was not reported.
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Tadpoles were housed at a loading rate of 2.5 tadpoles/L. They were kept at 20-24C with a 12:12 photoperiod. The organisms were fed daily after 96h in age throughout the study. A slurry was used for the tadpoles and pellets were used after metamorphosis.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—plasma testosterone levels.
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	StatView was used for statistical analysis. ANOVA was used as well as Tukey-Kramer post hoc test.
	Metric 22: Reporting of Data	Low	Data for the exposure related findings was reported in the text.
	Metric 23: Explanation of Unexpected Outcomes	Low	Variability was not reported.
Additional Comments:	This portion of the evaluation was on the effect of DBP on testosterone levels in <i>Xenopus laevis</i> . Plasma testosterone levels were measured, so the mechanistic endocrine outcome was chosen.		

Overall Quality Determination**High**

Study Citation:	Lee, S. K., Veeramachaneni, R., D.N. (2005). Subchronic exposure to low concentrations of di-n-butyl phthalate disrupts spermatogenesis in <i>Xenopus laevis</i> frogs. <i>Toxicological Sciences</i> 84(2):394-407.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Larvae			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	128004			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The DBP was identified by CASRN.	
	Metric 2: Test Substance Source	Low	The source was reported to be Sigma Aldrich, and the lot number was reported, but it was not reported if the DBP was analytically verified.	
	Metric 3: Test Substance Purity	High	The DBP was reported to be 99.8% pure.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a negative control in the FETAX solution and a negative control containing 0.01% DMSO.	
	Metric 5: Negative Control Response	High	Mortality in the control groups were less than 10% throughout the exposure.	
	Metric 6: Randomized Allocation	Medium	It was reported that tadpoles were randomly assigned to treatment groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	Stock solutions were prepared in 100mL amber glass bottles weekly by adding DBP directly to DMSO. Glass pipettes were used to add the proper quantity to the test tanks. It was then manually stirred. Renewal were conducted every M-W-F.	
	Metric 8: Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups. The same solvent was used in all test concentrations and in the solvent control. All test concentrations were run until complete metamorphosis of the control organisms.	
	Metric 9: Measurement of Test Substance Concentration	Low	Study authors did not report if the exposure concentrations were measured.	
	Metric 10: Exposure Duration and Frequency	High	The exposure duration was reported to be from stage 52 (3wks) to stage 66 (8wks). This is typical of amphibian studies.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	There were 5 exposure levels as well as a negative control and a solvent control. This was adequate to see a response.	
	Metric 12: Testing at or Below Solubility Limit	High	A solvent was used in this study, and the solvent control response was adequate indicating the concentration was appropriate.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	Breeding adults were reported to be from Nasco in Fort Atkinson, WI. Adults were bred in the laboratory performing the study, and the tadpoles were reared there. The tadpole stages were appropriate for the study.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	Pretreatment conditions were the same as treatment conditions. Temperature and photoperiod were the same for each.	
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Study Citation:	Lee, S. K., Veeramachaneni, R., D.N. (2005). Subchronic exposure to low concentrations of di-n-butyl phthalate disrupts spermatogenesis in <i>Xenopus laevis</i> frogs. <i>Toxicological Sciences</i> 84(2):394-407.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Larvae
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	128004

Domain	Metric	Rating	Comments
	Metric 15: Number of Organisms and Replicates per Group	Low	The number of test organisms for each test concentration were reported, but the number of replicates for each test concentration was not reported.
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Tadpoles were housed at a loading rate of 2.5 tadpoles/L. They were kept at 20-24C with a 12:12 photoperiod. The organisms were fed daily after 96h in age throughout the study. A slurry was used for the tadpoles and pellets were used after metamorphosis.
	Metric 17: Outcome Assessment Methodology	Low	The mortality assessment was not described in the methods section.
	Metric 18: Consistency of Outcome Assessment	Low	How mortality was assessed was not reported.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	StatView was used for statistical analysis. ANOVA was used as well as Tukey-Kramer post hoc test.
	Metric 22: Reporting of Data	Medium	Mortality was just briefly described in text in the results section. Cumulative percent mortalities per treatment group were given there but no data was shown in any table or figure.
	Metric 23: Explanation of Unexpected Outcomes	Medium	Study authors did not report any unexpected outcomes. No variability was shown for mortality data.
Additional Comments: This evaluation is for the mortality assessment during the study.			

Overall Quality Determination**High**

Study Citation:	Lee, S. K., Veeramachaneni, R., D.N. (2005). Subchronic exposure to low concentrations of di-n-butyl phthalate disrupts spermatogenesis in <i>Xenopus laevis</i> frogs. <i>Toxicological Sciences</i> 84(2):394-407.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Larvae			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	128004			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The DBP was identified by CASRN.	
	Metric 2: Test Substance Source	Low	The source was reported to be Sigma Aldrich, and the lot number was reported, but it was not reported if the DBP was analytically verified.	
	Metric 3: Test Substance Purity	High	The DBP was reported to be 99.8% pure.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a negative control in the FETAX solution and a negative control containing 0.01% DMSO.	
	Metric 5: Negative Control Response	High	The biological response of the negative controls was reported in Figures 2,3, and 6, as well as in Tables 1 and 2.	
	Metric 6: Randomized Allocation	Medium	It was reported that tadpoles were randomly assigned to treatment groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	Stock solutions were prepared in 100mL amber glass bottles weekly by adding DBP directly to DMSO. Glass pipettes were used to add the proper quantity to the test tanks. It was then manually stirred. Renewals were conducted every M-W-F.	
	Metric 8: Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups. The same solvent was used in all test concentrations and in the solvent control. All test concentrations were run until complete metamorphosis of the control organisms.	
	Metric 9: Measurement of Test Substance Concentration	Low	Study authors did not report if the exposure concentrations were measured.	
	Metric 10: Exposure Duration and Frequency	High	The exposure duration was reported to be from stage 52 (3wks) to stage 66 (8wks). This is typical of amphibian studies.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	There were five exposure levels as well as a negative control and a solvent control. This was adequate to see a response.	
	Metric 12: Testing at or Below Solubility Limit	High	A solvent was used in this study, and the solvent control response was adequate indicating the concentration was appropriate.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	Breeding adults were reported to be from Nasco in Fort Atkinson, WI. Adults were bred in the laboratory performing the study, and the tadpoles were reared there. The tadpole stages were appropriate for the study.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	Pretreatment conditions were the same as treatment conditions. Temperature and photoperiod were the same for each.	
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Study Citation:	Lee, S. K., Veeramachaneni, R., D.N. (2005). Subchronic exposure to low concentrations of di-n-butyl phthalate disrupts spermatogenesis in <i>Xenopus laevis</i> frogs. <i>Toxicological Sciences</i> 84(2):394-407.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Larvae
Health Outcome:	Reproductive/Teratogenic
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	128004

Domain	Metric	Rating	Comments
	Metric 15: Number of Organisms and Replicates per Group	Low	The number of test organisms for each test concentration were reported, but the number of replicates for each test concentration was not reported.
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Tadpoles were housed at a loading rate of 2.5 tadpoles/L. They were kept at 20-24C with a 12:12 photoperiod. The organisms were fed daily after 96h in age throughout the study. A slurry was used for the tadpoles and pellets were used after metamorphosis.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—histopathological changes in reproductive organs.
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	StatView was used for statistical analysis. ANOVA was used as well as Tukey-Kramer post hoc test.
	Metric 22: Reporting of Data	High	Data for exposure related findings including control results can be found in Tables 1 and 2 as well as in Figures 2,3 and 6. Other figures provided exposure related results only.
	Metric 23: Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in the tables.
Additional Comments:	This portion of the evaluation was on the effect of DBP on spermatogenesis in <i>Xenopus laevis</i> . Histopathological analysis was performed to observe the effects on the reproductive system in male frogs. Reproduction was therefore selected as the outcome of interest.		

Overall Quality Determination**High**

Study Citation:	Shen, O., Wu, W., Du, G., Liu, R., Yu, L., Sun, H., Han, X., Jiang, Y., Shi, W., Hu, W., Song, L., Xia, Y., Wang, S., Wang, X. (2011). Thyroid disruption by Di-n-butyl phthalate (DBP) and mono-n-butyl phthalate (MBP) in <i>Xenopus laevis</i> . PLoS ONE 6(4):e19159.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Larvae			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	787926			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The DBP was identified by CAS number.	
	Metric 2: Test Substance Source	Low	The source was reported to be Tokyo Kasei, however, it was not reported if the DBP was analytically verified.	
	Metric 3: Test Substance Purity	High	The purity was reported to be >99%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	A negative solvent control was reported to be used in this study.	
	Metric 5: Negative Control Response	High	The negative control response was reported in Table 2, and was adequate for the outcomes of interest.	
	Metric 6: Randomized Allocation	Low	It was not reported how the tadpoles were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	Stock solution was made by adding 250mg/L of DBP into DMSO. The stock solution was then diluted to the proper test concentration by adding to dechlorinated water in the appropriate amount.	
	Metric 8: Consistency of Exposure Administration	High	All exposures were for 21 days with test solutions renewed every two days. All exposure concentrations had 20 tadpoles in 5L of test solution.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if the test concentrations were measured at any point in the study.	
	Metric 10: Exposure Duration and Frequency	High	The exposure duration was reported to be 21 days, which appeared adequate for a response.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Medium	There were only three exposure levels, but this appeared adequate for a dose response. The spacing was appropriate for the study.	
	Metric 12: Testing at or Below Solubility Limit	High	DMSO was used as a vehicle solvent in this study at a concentration of 0.005%.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	Spawning adults were reported to be from Nasco in the United States. Tadpoles were obtained from these spawners and were placed into the test system at stage 51 (14-16d).	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the tadpoles were acclimated for any length of time.	
	Metric 15: Number of Organisms and Replicates per Group	Low	There were 20 tadpoles per test chamber, but the number of replicates per concentration was not reported.	
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Study Citation:	Shen, O., Wu, W., Du, G., Liu, R., Yu, L., Sun, H., Han, X., Jiang, Y., Shi, W., Hu, W., Song, L., Xia, Y., Wang, S., Wang, X. (2011). Thyroid disruption by Di-n-butyl phthalate (DBP) and mono-n-butyl phthalate (MBP) in <i>Xenopus laevis</i> . PLoS ONE 6(4):e19159.		
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: Not-reported		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Larvae		
Health Outcome:	Development/Growth		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	787926		
Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
Metric 16:	Adequacy of Test Conditions	High	Tadpoles were fed Nasco Frog Brittle and were kept at 22 C with a 12L:12D photoperiod.
Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest–development/growth.
Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. Tadpoles were assessed daily for their developmental stages. On day 22, stage, body length, interocular distance, and body-to-tail length ratio.
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	High	There weren't any reported differences among the study groups in environmental conditions.
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	High	ANOVA and Duncan's multiple comparison test were used for statistical analysis.
Metric 22:	Reporting of Data	High	Data for the control response and the dose response can be found in Table 2 and are adequate for the outcomes of interest.
Metric 23:	Explanation of Unexpected Outcomes	High	The study authors did not report any unexpected outcomes. Variability is reported in Table 2.
Additional Comments:	This portion of the study is on the effect of DBP on the stage, length, intraocular distances, and the body-to-tail length ration. Development/growth was selected as the outcome of interest. Tadpoles were kept in aquariums until Stage 57. A time period was not given for this, thus the "not reported" study duration.Nieuwkoop PD, Faber J (1994)Normal Table of <i>Xenopus laevis</i> (Daudin): ASystematical and Chronological Survey of the Development from the FertilizedEgg till the End of Metamorphosis; Garland Publishing; New Yorkwas cited as the reference for staging the tadpoles in the methods.		

Overall Quality Determination**High**

Study Citation:	Shen, O., Wu, W., Du, G., Liu, R., Yu, L., Sun, H., Han, X., Jiang, Y., Shi, W., Hu, W., Song, L., Xia, Y., Wang, S., Wang, X. (2011). Thyroid disruption by Di-n-butyl phthalate (DBP) and mono-n-butyl phthalate (MBP) in <i>Xenopus laevis</i> . PLoS ONE 6(4):e19159.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Larvae			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Epigenetics-Endocrine toxicity			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	787926			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The DBP was identified by CAS number.	
	Metric 2: Test Substance Source	Low	The source was reported to be Tokyo Kasei, however, it was not reported if the DBP was analytically verified.	
	Metric 3: Test Substance Purity	High	The purity was reported to be >99%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	A negative solvent control was reported to be used in this study.	
	Metric 5: Negative Control Response	High	The negative control response was reported in Figures 2 and 3, and was adequate for the outcomes of interest.	
	Metric 6: Randomized Allocation	Low	It was not reported how the tadpoles were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	Stock solution was made by adding 250mg/L of DBP into DMSO. The stock solution was then diluted to the proper test concentration by adding to dechlorinated water in the appropriate amount.	
	Metric 8: Consistency of Exposure Administration	High	All exposures were for 21 days with test solutions renewed every 2 days. All exposure concentrations had 20 tadpoles in 5L of test solution.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if the test concentrations were measured at any point in the study.	
	Metric 10: Exposure Duration and Frequency	High	The exposure duration was reported to be 21 days, which appeared adequate for a response.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Medium	There were only 3 exposure levels, but this appeared adequate for a dose response. The spacing was appropriate for the study.	
	Metric 12: Testing at or Below Solubility Limit	High	DMSO was used as a vehicle solvent in this study at a concentrations of 0.005%	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	Spawning adults were reported to be from Nasco in the United States. Tadpoles were obtained from these spawners and were placed into the test system at stage 51 (14-16d).	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the tadpoles were acclimated for any length of time.	
	Metric 15: Number of Organisms and Replicates per Group	Low	There were 20 tadpoles per test chamber, but the number of replicates per concentration was not reported.	
Domain 5: Outcome Assessment				
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Study Citation:	Shen, O., Wu, W., Du, G., Liu, R., Yu, L., Sun, H., Han, X., Jiang, Y., Shi, W., Hu, W., Song, L., Xia, Y., Wang, S., Wang, X. (2011). Thyroid disruption by Di-n-butyl phthalate (DBP) and mono-n-butyl phthalate (MBP) in <i>Xenopus laevis</i> . PLoS ONE 6(4):e19159.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Larvae			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Epigenetics-Endocrine toxicity			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	787926			
Domain	Metric	Rating	Comments	
	Metric 16: Adequacy of Test Conditions	High	Tadpoles were fed Nasco Frog Brittle and were kept at 22 C with a 12L:12D photoperiod.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—gene expression after exposure.	
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the conditions for the study groups.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	ANOVA and Duncan’s multiple comparison test were used for statistical analysis. Chi-square was used for the portion of the study pertaining the methylation.	
	Metric 22: Reporting of Data	High	Data for the control response and the dose response can be found in Figures 2 and 3 and in Table 3.	
	Metric 23: Explanation of Unexpected Outcomes	High	The study authors did not report any unexpected outcomes. Variability is reported in the figures and tables.	
Additional Comments:	This portion of the study is on the effect of DBP on gene expression relating to the the thyroid and endocrine system as well as methylation. Mechanistic outcomes were selected for this reason. Tadpoles were kept in aquariums until Stage 57. A time period was not given for this, thus the ”not reported” study duration.Nieuwkoop PD, Faber J (1994)Normal Table of <i>Xenopus laevis</i> (Daudin): ASystematical and Chronological Survey of the Development from the FertilizedEgg till the End of Metamorphosis; Garland Publishing: New Yorkwas cited as the reference for staging the tadpoles in the methods.			

Overall Quality Determination**High**

Study Citation:	Cruciani, V., Iovine, C., Thomé, J. P., Joaquim-Justo, C. (2015). Impact of three phthalate esters on the sexual reproduction of the Monogonont rotifer, <i>Brachionus calyciflorus</i> . <i>Ecotoxicology</i> 25(1):192-200.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Other Invertebrate (e.g., sea urchins, ciliates, rotifers); <i>Brachionus calyciflorus</i> ; Pallas; Larvae			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3070931			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The test substance was identified only by the chemical name. No other information was provided.
	Metric 2:	Test Substance Source	Low	The test substance was obtained from Sigma Aldrich, Germany, but the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of the test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group (i.e. all conditions equal except chemical exposure). Both water and solvent controls were used.
	Metric 5:	Negative Control Response	High	The biological responses of the controls were reported and were adequate.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations and/or minimize loss of test substance before and during the exposure. Concentrations of the test substance were not measured during the study.
	Metric 8:	Consistency of Exposure Administration	Medium	Details of initial exposure administration were reported but details of exposure administration past 48 hours was not clearly reported. It was reported that at 48 hours, rotifers were transferred to new test media and at 48, 72 and 96 hours, tubes were emptied into a glass petri dish for counting males and females, but no details were given on how the experiment was continued until 96 hours.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type and/or outcomes of interest.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	There were four exposure groups for DBP and five replicates were run for each concentration.
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate (i.e., no effects on biological responses were observed in the solvent control and no interactions were expected between the solvent and test substance).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.

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Study Citation:	Cruciani, V., Iovine, C., Thomé, J. P., Joaquim-Justo, C. (2015). Impact of three phthalate esters on the sexual reproduction of the Monogonont rotifer, <i>Brachionus calyciflorus</i> . <i>Ecotoxicology</i> 25(1):192-200.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Other Invertebrate (e.g., sea urchins, ciliates, rotifers); <i>Brachionus calyciflorus</i> ; Pallas; Larvae			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3070931			
Domain	Metric	Rating	Comments	
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized and/or whether pre-treatment conditions were the same for control and exposed groups.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Organism housing, environmental conditions and food were conducive to maintenance of health. It was reported that the population growth rate at 48 h in the control was greater than 0.7, which is indicative of healthy conditions.
	Metric 17:	Outcome Assessment Methodology	Medium	The outcome assessment methodology addressed or reported the intended outcome(s) of interest. Methodology consisted of counting non-ovigerous females, amictic ovigerous females, mictic ovigerous females, fertilized females, fertilized eggs carried per fertilized female and detached fertilized eggs. In the paper and the cited reference (Preston et al. 2000), it was reported that test tube contents were emptied into a petri dish at 48 and 72 hours but there were no details given regarding the methods for continuing the experiment until 96 hours.
	Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. Outcomes were assessed at 48, 72 and 96 hours in treatment groups and controls.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study on outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were clearly described.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments:	DBP exposure negatively affected asexual reproduction in rotifers at the highest concentration tested (2mg/L) at 48 and 72 hours and sexual reproduction at 2mg/L after 96 hours.			

Overall Quality Determination**Medium**

Study Citation:	Zhao, L. L., Xi, Y. L., Huang, L., Zha, C. W. (2009). Effects of three phthalate esters on the life-table demography of freshwater rotifer <i>Brachionus calyciflorus</i> Pallas. <i>Aquatic Ecology</i> 43(2):395-402.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Brachionus calyciflorus</i> ; Juvenile			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1336226			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The chemical was identified by name.	
	Metric 2: Test Substance Source	High	The source of the test substance was Sigma Aldrich (Germany). The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity was reported as >=97%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was adequate.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were adequate for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	Low	Concentrations exceeded solubility, but with solvents they were at an appropriate level aided in dissolution.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Medium	Eggs were collected from Lake Jinghu and cultured in the lab. There are minor reservations regarding the source of test organisms that are unlikely to have a substantial impact on results.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.	
Domain 5: Outcome Assessment				
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Study Citation:	Zhao, L. L., Xi, Y. L., Huang, L., Zha, C. W. (2009). Effects of three phthalate esters on the life-table demography of freshwater rotifer <i>Brachionus calyciflorus</i> Pallas. <i>Aquatic Ecology</i> 43(2):395-402.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Brachionus calyciflorus</i> ; Juvenile
Health Outcome:	Reproductive/Teratogenic
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	1336226

Domain	Metric	Rating	Comments
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	High	There were no differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: None

Overall Quality Determination**Medium**

Study Citation:	Zhao, L. L., Xi, Y. L., Huang, L., Zha, C. W. (2009). Effects of three phthalate esters on the life-table demography of freshwater rotifer <i>Brachionus calyciflorus</i> Pallas. <i>Aquatic Ecology</i> 43(2):395-402.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Brachionus calyciflorus</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1336226			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	Chemical was identified by name	
	Metric 2: Test Substance Source	High	The source of the test substance was Sigma Aldrich (Germany). The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity reported as >=97%	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control groups	
	Metric 5: Negative Control Response	High	The biological response of the negative control groups was adequate	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were adequate for a dose response	
	Metric 12: Testing at or Below Solubility Limit	Low	Concentrations exceeded solubility but solvents at an appropriate level aided in dissolution	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Medium	There are minor reservations source of test organisms that are unlikely to have a substantial impact on results. Organisms were collected in the sediment of Lake Jinghu and cultured in the lab.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health	
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Study Citation:	Zhao, L. L., Xi, Y. L., Huang, L., Zha, C. W. (2009). Effects of three phthalate esters on the life-table demography of freshwater rotifer <i>Brachionus calyciflorus</i> Pallas. <i>Aquatic Ecology</i> 43(2):395-402.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Brachionus calyciflorus</i> ; Juvenile
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	1336226

Domain	Metric	Rating	Comments
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
	Metric 20: Outcomes Unrelated to Exposure	High	There were no differences among groups
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: None

Overall Quality Determination**Medium**

Study Citation:	Streufert, J. M., Jones, J. R., Sanders, H. O. (1980). Toxicity and biological effects of phthalate esters on midges (<i>Chironomus plumosus</i>). Transactions of the Missouri Academy of Science 14:33-40.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Chironomus plumosus</i> ; Larvae			
Health Outcome:	Immobilization			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	813673; Linked HERO ID(s): 813673, 1332972			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The DBP was identified by name only.
	Metric 2:	Test Substance Source	Low	The source of the DBP was reported to be Aldrich Company in St. Louis, MO. It was not reported if the DBP was analytically verified.
	Metric 3:	Test Substance Purity	Low	The purity and the grade of the DBP was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	The study reported the use of a concurrent negative control in which the solvent was used.
	Metric 5:	Negative Control Response	Low	The biological response of the negative control was not reported. Only EC50 values were reported.
	Metric 6:	Randomized Allocation	Low	It was not reported how the larvae were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	Little information was provided on the preparation of the test media.
	Metric 8:	Consistency of Exposure Administration	High	Study authors cited the Committee on Methods of Toxicity Tests with Aquatic Organisms, 1975 for the methods used in the acute toxicity tests.
	Metric 9:	Measurement of Test Substance Concentration	Medium	It was reported that test concentrations were measured at the start of the test, but the methods used were not reported.
	Metric 10:	Exposure Duration and Frequency	Medium	The study duration was reported to be 48h. 96h are typical for midge acute toxicity tests. The test duration did not appear to have a significant effect on the outcome.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Uninformative	The study authors did not report the number of exposure groups or the spacing of the groups for the acute tests.
	Metric 12:	Testing at or Below Solubility Limit	High	Study authors reported using an appropriate vehicle solvent, and it was kept under 0.1mL/L in all test concentrations and controls.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were obtained from an in-house culture and were the appropriate age for the study.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report if the organisms were acclimated or needed to be acclimated to test conditions.
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Study Citation:	Streufert, J. M., Jones, J. R., Sanders, H. O. (1980). Toxicity and biological effects of phthalate esters on midges (<i>Chironomus plumosus</i>). Transactions of the Missouri Academy of Science 14:33-40.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Chironomus plumosus</i> ; Larvae			
Health Outcome:	Immobilization			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	813673; Linked HERO ID(s): 813673, 1332972			
Domain	Metric	Rating	Comments	
	Metric 15: Number of Organisms and Replicates per Group	Low	The number of test organisms per test chamber and the number of replicates was not reported, though this may have been included in the citation for methodology.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Medium	All tests were conducted at 22C with a 16L:8D photoperiod. Well water was used in the test media. It was not reported if the organisms were fed or what the biomass per test chamber was.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest–Immobilization was reported in EC50 values.	
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol were not reported. Organisms were assessed for immobilization, but it was not clear how this was done.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Methods described by Litchfield and Wilcoxon 1949 were used to determine EC50 values.	
	Metric 22: Reporting of Data	Low	Only EC50 values were reported.	
	Metric 23: Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in Table 2.	
Additional Comments:	This portion of the evaluation was on the acute toxicity of DBP on <i>C. pulmosus</i> . The study reported assessing immobilization as the outcome, so that was selected as the outcome for the evaluation. The study received an unacceptable ranking due to the lack of reporting on exposure groups and spacing.			

Overall Quality Determination**Uninformative**

Study Citation:	Streufort, J. M. (1978). Some effects of two phthalic acid esters on the life cycle of the midge (<i>Chironomus plumosus</i>).			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Chironomus plumosus</i> ; Larvae			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332972; Linked HERO ID(s): 813673, 1332972			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	Nomenclature referenced on page 13/62. No CAS or structure listed.	
	Metric 2: Test Substance Source	Low	Source was listed from Monsanto but not analytically verified.	
	Metric 3: Test Substance Purity	Low	No purity reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	Low	Ethanol listed for solvent controls, however, authors report using ethanol concentrations at higher than recommended for acute toxicity testing (1.8 ml/L) to increase solubility of compounds.	
	Metric 5: Negative Control Response	Low	Control responses are not reported for acute toxicity bioassays.	
	Metric 6: Randomized Allocation	Low	No reporting on how animals were allocated to treatment concentrations.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The acute toxicity bioassays were conducted as static non-renewal, with morality assessed at 24 and 48 hours.	
	Metric 8: Consistency of Exposure Administration	High	exposure administration appeared consistent among treatments and control.	
	Metric 9: Measurement of Test Substance Concentration	Low	The acute bioassay concentrations were not analyzed and LC50 concentrations are reported as nominal.	
	Metric 10: Exposure Duration and Frequency	High	The duration (48 hr) is appropriate.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Low	Non of the treatment concentrations for the acute bioassays are reported. Range finding tests were not described.	
	Metric 12: Testing at or Below Solubility Limit	Low	The LC50 values are under the solubility reported in the Final Scope for DBP (11.2 mg/L). It is not certain if the range of concentrations were under the solubility since they were not reported.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The source of animals was reported as from the CNFRL in Columbia MO. The life stages were identified.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	The pretreatment conditions were listed and similar to the 48 hr acute toxicity tests.	
	Metric 15: Number of Organisms and Replicates per Group	Low	The authors listed 10 individuals per treatment concentrations but did not report the level of replication for each treatment.	
Domain 5: Outcome Assessment				
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Study Citation:	Streufort, J. M. (1978). Some effects of two phthalic acid esters on the life cycle of the midge (<i>Chironomus plumosus</i>).			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Chironomus plumosus</i> ; Larvae			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332972; Linked HERO ID(s): 813673, 1332972			
Domain	Metric	Rating	Comments	
	Metric 16:	Adequacy of Test Conditions	Medium	DO, temperature, and photoperiod were reported for the acute bioassays.
	Metric 17:	Outcome Assessment Methodology	High	The authors had a very detailed list of multiple criteria to determine mortality from the bioassays.
	Metric 18:	Consistency of Outcome Assessment	High	The outcomes appear to be reported consistently.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	Nothing was reported to indicate differences among treatment groups with environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	No information to suggest differences among groups related to health outcomes.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Authors used Litchfield and Wilcoxon method for LC50 estimation.
	Metric 22:	Reporting of Data	Medium	LC 50 and 95% Confidence intervals are reported, but results from each concentration are not available.
	Metric 23:	Explanation of Unexpected Outcomes	High	No unexpected outcomes were reported.
Additional Comments:	None			
Overall Quality Determination		Medium		

Study Citation:	Streufort, J. M. (1978). Some effects of two phthalic acid esters on the life cycle of the midge (<i>Chironomus plumosus</i>).			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Chironomus plumosus</i> ; Larvae			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332972; Linked HERO ID(s): 813673, 1332972			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	Nomenclature was referenced on page 13/62. No CAS or structure was listed.	
	Metric 2: Test Substance Source	Low	Source was listed from Monsanto but not analytically verified.	
	Metric 3: Test Substance Purity	Low	No purity was reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	Low	Ethanol was listed for solvent controls, however, authors reported using ethanol concentrations at higher than recommended for acute toxicity testing (1.8 ml/L) to increase solubility of compounds.	
	Metric 5: Negative Control Response	Low	Control responses are not reported for acute toxicity bioassays.	
	Metric 6: Randomized Allocation	Low	There was no reporting on how animals were allocated to treatment concentrations.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The acute toxicity bioassays were conducted as static non-renewal, with morality assessed at 24 and 48 hours.	
	Metric 8: Consistency of Exposure Administration	High	Exposure administration appeared consistent among treatments and control.	
	Metric 9: Measurement of Test Substance Concentration	Low	The acute bioassay concentrations were not analyzed and LC50 concentrations were reported as nominal.	
	Metric 10: Exposure Duration and Frequency	High	The duration (48 hr) is appropriate.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Low	None of the treatment concentrations for the acute bioassays are reported. Range finding tests were not described.	
	Metric 12: Testing at or Below Solubility Limit	Low	The LC50 values are under the solubility reported in the Final Scope for DBP (11.2 mg/L). It is not certain if the range of concentrations were under the solubility since they were not reported.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The source of animals was reported as from the CNFRL in Columbia MO. The life stages were identified.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	The pretreatment conditions were listed and similar to the 48 hr acute toxicity tests.	
	Metric 15: Number of Organisms and Replicates per Group	Low	The authors listed 10 individuals per treatment concentrations but did not report the level of replication for each treatment.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Medium	Dissolved oxygen, temperature, and photoperiod were reported for the acute bioassays.	
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Study Citation:	Streufort, J. M. (1978). Some effects of two phthalic acid esters on the life cycle of the midge (<i>Chironomus plumosus</i>).			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Chironomus plumosus</i> ; Larvae			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332972; Linked HERO ID(s): 813673, 1332972			
Domain	Metric	Rating	Comments	
	Metric 17:	Outcome Assessment Methodology	High	The authors had a very detailed list of multiple criteria to determine mortality from the bioassays.
	Metric 18:	Consistency of Outcome Assessment	High	The outcomes appear to be reported consistently.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	Nothing was reported to indicate differences among treatment groups with environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information to suggest differences among groups related to health outcomes.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Authors used Litchfield and Wilcoxon method for LC50 estimation.
	Metric 22:	Reporting of Data	Medium	LC 50 and 95% confidence intervals are reported, but results from each concentration are not available.
	Metric 23:	Explanation of Unexpected Outcomes	High	No unexpected outcomes were reported.
Additional Comments:	None			
Overall Quality Determination		Medium		

Study Citation:	Jr, Mayer, F., Sanders, H. O., Walsh, D. F. (1973). Toxicity, residue dynamics, and reproductive effects of phthalate esters in aquatic invertebrates. Environmental Research 6(1):84-90.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Chironomus plumosus</i> ; Larvae			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1334646			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The chemical was identified by name.
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	The purity and/or grade of the test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Uninformative	A concurrent negative control group was not included or reported.
	Metric 5:	Negative Control Response	Low	The biological response of the negative control groups was not reported.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.
	Metric 8:	Consistency of Exposure Administration	Low	Few details of exposure administration were reported, but exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	Only one treatment was reported.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test animals was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of test replicates was not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.
	Metric 17:	Outcome Assessment Methodology	Medium	The outcome assessment methodology was not clearly reported.

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Study Citation:	Jr, Mayer, F., Sanders, H. O., Walsh, D. F. (1973). Toxicity, residue dynamics, and reproductive effects of phthalate esters in aquatic invertebrates. Environmental Research 6(1):84-90.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Chironomus plumosus</i> ; Larvae			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1334646			
Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	Medium	Few details regarding the execution of the study protocol for outcome assessment were provided.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was not performed.
	Metric 22:	Reporting of Data	Medium	Data for exposure-related findings were presented for the sampling period.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments: None				

Overall Quality Determination**Uninformative**

Study Citation:	Streufort, J. M. (1978). Some effects of two phthalic acid esters on the life cycle of the midge (<i>Chironomus plumosus</i>).			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Chironomus plumosus</i> ; Larvae			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332972; Linked HERO ID(s): 813673, 1332972			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	Nomenclature was referenced on page 13/62. No CAS or structure were listed.	
	Metric 2: Test Substance Source	Low	Source was listed from Monsanto but not analytically verified.	
	Metric 3: Test Substance Purity	Low	No purity was reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Solvent controls (Ethanol) were used at 0.12 ml/L.	
	Metric 5: Negative Control Response	High	Control responses are reported for emergence from chronic exposures.	
	Metric 6: Randomized Allocation	Low	There was no reporting on how animals were allocated to treatment concentrations.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The chronic exposure were conducted with a flow-through system for continual renewal of the chemical. The setup and flow-rate was described well on page 32/62.	
	Metric 8: Consistency of Exposure Administration	High	Exposure administration appeared consistent among treatments and control.	
	Metric 9: Measurement of Test Substance Concentration	Medium	GC was used to verify the concentrations from the chronic exposure on page 25/62.	
	Metric 10: Exposure Duration and Frequency	High	The chronic exposures for midge emergence ranged from 20-40 days for hydrosol substrate and 20 - 35 days for sand substrate.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Medium	The measured concentrations are reported in tables within the results section beginning on page 35/62.	
	Metric 12: Testing at or Below Solubility Limit	Medium	The chronic exposures are all below the published solubility value published in the final scope for DBP (11.2 mg/l).	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The source of animals was reported as from the CNFRL in Columbia, MO. The life stages were identified.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	The pretreatment conditions were listed and similar to the chronic exposures.	
	Metric 15: Number of Organisms and Replicates per Group	Low	Authors began each treatment and control group with 100 1st instar larvae for the chronic emergence bioassay. The replication and housing groups for this work was not well described.	
Domain 5: Outcome Assessment				
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Study Citation:	Streufort, J. M. (1978). Some effects of two phthalic acid esters on the life cycle of the midge (<i>Chironomus plumosus</i>).			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Chironomus plumosus</i> ; Larvae			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332972; Linked HERO ID(s): 813673, 1332972			
Domain	Metric	Rating	Comments	
	Metric 16:	Adequacy of Test Conditions	Medium	Dissolved oxygen, temperature, and photoperiod were reported for the chronic exposures.
	Metric 17:	Outcome Assessment Methodology	Medium	No significant differences in emergence were observed for the chronic exposures.
	Metric 18:	Consistency of Outcome Assessment	High	The outcomes appear to be reported consistently.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	Nothing was reported to indicate differences among treatment groups with environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information to suggest differences among groups related to health outcomes.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	The percent data was arc sin square-root transformed and Least Significant Difference tests were performed.
	Metric 22:	Reporting of Data	Medium	Emergence is reported in total numbers for each concentration, compound, and day of exposure in tables presented in the results section.
	Metric 23:	Explanation of Unexpected Outcomes	High	No unexpected outcomes were reported.
Additional Comments:	None			
Overall Quality Determination		Medium		

Study Citation:	Call, D. J., Cox, D. A., Geiger, D. L., Genisot, K. I., Markee, T. P., Brooke, L. T., Polkinghorne, C. N., Vandeventer, F. A., Gorsuch, J. W., Robillard, K. A., Parkerton, T. F., Reiley, M. C., Ankley, G. T., Mount, D. R. (2001). An assessment of the toxicity of phthalate esters to freshwater benthos. 2. Sediment exposures. Environmental Toxicology and Chemistry 20(8):1805-1815.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Sediment; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Chironomus tentans</i> ; Larvae			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	679311			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
Metric 1:	Test Substance Identity	High	Chemical was identified as a "single isomer" and the identify, including CASRN referenced in an outside paper, Call et al 2001.	
Metric 2:	Test Substance Source	High	Source of chemical was defined in Call et al 2001 as AldrichChemical (Milwaukee, WI, USA).	
Metric 3:	Test Substance Purity	High	Purity was identified as >99% in Call et al 2001.	
Domain 2: Test Design				
Metric 4:	Negative Controls	High	Negative controls (sediment and silica sand) were used in this experiment.	
Metric 5:	Negative Control Response	High	Biological response of control groups was appropriate as shown in Table 4.	
Metric 6:	Randomized Allocation	Low	Random allocation not reported.	
Domain 3: Exposure Characterization				
Metric 7:	Experimental System/Test Media Preparation	High	Methods of sediment collection and preparation (including chemical addition) and addition of sediment to test beakers were described in detail.	
Metric 8:	Consistency of Exposure Administration	High	Exposure consistency reported and consistent among different DBP treatments and controls.	
Metric 9:	Measurement of Test Substance Concentration	High	Concentrations measured using HPLC as described in methods and cited reference (Call et al 2001).	
Metric 10:	Exposure Duration and Frequency	High	Duration (10 day exposure) was appropriate for experimental design.	
Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Exposure groups were acceptable and spanned 5 concentrations per test species in addition to control; nominal doses unclear however measured doses reported in sediment and pore water.	
Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure via sediment.	
Domain 4: Test Organism				
Metric 13:	Test Organism Characteristics	Low	Source of test organism not reported.	
Metric 14:	Acclimatization and Pretreatment Conditions	Low	Acclimation of test organisms prior to exposure not reported.	
Metric 15:	Number of Organisms and Replicates per Group	Medium	Tests with DBP utilized 3 replicates of five different concentrations with 10 organisms per beaker and three sediment control replicates with 10 test organisms per beaker and three silica sand control replicates with 10 test organisms per beaker.	

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Study Citation:	Call, D. J., Cox, D. A., Geiger, D. L., Genisot, K. I., Markee, T. P., Brooke, L. T., Polkinghorne, C. N., Vandeventer, F. A., Gorsuch, J. W., Robillard, K. A., Parkerton, T. F., Reiley, M. C., Ankley, G. T., Mount, D. R. (2001). An assessment of the toxicity of phthalate esters to freshwater benthos. 2. Sediment exposures. Environmental Toxicology and Chemistry 20(8):1805-1815.		
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Sediment; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Chironomus tentans</i> ; Larvae		
Health Outcome:	Mortality		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	679311		
Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions including feeding, intermittent water renewal system, lighting, and temperature were reported. Authors recorded temperature, dissolved oxygen, and pH on days 0 and 10 and conductivity, hardness, alkalinity, and ammonia on days 1 and 9. Sediment TOC conditions described in Table 2.
	Metric 17: Outcome Assessment Methodology	Medium	Survivor count determined after the 10 day exposure but not reported as percent mortality.
	Metric 18: Consistency of Outcome Assessment	High	Outcome assessment conducted at conclusion of 10 day exposure.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	Metric 20: Outcomes Unrelated to Exposure	High	Details regarding test organism attrition and outcomes unrelated to exposure were reported for each study group and there were no differences among groups that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Survival data from toxicity tests were summarized using the trimmed Spearman-Kärber method. Dry weight data were analyzed by one-way analysis of variance and Dunnett's procedure using a SigmaStat Program.
	Metric 22: Reporting of Data	High	Survival data reported in Table 4 and LC50 values shown in Figure 1 and Table 6.
	Metric 23: Explanation of Unexpected Outcomes	Medium	Variability not reported but results suggest no excessive variability within replicates.
Additional Comments: None			

Overall Quality Determination**High**

Study Citation:	Call, D. J., Cox, D. A., Geiger, D. L., Genisot, K. I., Markee, T. P., Brooke, L. T., Polkinghorne, C. N., Vandeventer, F. A., Gorsuch, J. W., Robillard, K. A., Parkerton, T. F., Reiley, M. C., Ankley, G. T., Mount, D. R. (2001). An assessment of the toxicity of phthalate esters to freshwater benthos. 2. Sediment exposures. Environmental Toxicology and Chemistry 20(8):1805-1815.		
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Sediment; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Chironomus tentans</i> ; Larvae		
Health Outcome:	Development/Growth		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	679311		
Domain	Metric	Rating	Comments
Domain 1: Test Substance			
	Metric 1: Test Substance Identity	High	The chemical was identified as a "single isomer" and the identify, including CASRN referenced in an outside paper, Call et al 2001.
	Metric 2: Test Substance Source	High	The source of the chemical was defined in Call et al 2001 as AldrichChemical (Milwaukee, WI, USA).
	Metric 3: Test Substance Purity	High	The purity was identified as >99% in Call et al 2001.
Domain 2: Test Design			
	Metric 4: Negative Controls	High	Negative controls (sediment and silica sand) were used in this experiment.
	Metric 5: Negative Control Response	High	Biological response of control groups was appropriate as shown in Table 4.
	Metric 6: Randomized Allocation	Low	Random allocation was not reported.
Domain 3: Exposure Characterization			
	Metric 7: Experimental System/Test Media Preparation	High	Methods of sediment collection and preparation (including chemical addition) and addition of sediment to test beakers were described in detail.
	Metric 8: Consistency of Exposure Administration	High	Exposure consistency was reported and consistent among different DBP treatments and controls.
	Metric 9: Measurement of Test Substance Concentration	High	Concentrations were measured using HPLC as described in the methods and cited reference (Call et al 2001).
	Metric 10: Exposure Duration and Frequency	High	The duration (10-day exposure) was appropriate for the experimental design.
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Medium	Exposure groups were acceptable and spanned five concentrations per test species in addition to the control. Nominal doses were unclear, however measured doses were reported in sediment and pore water.
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via sediment.
Domain 4: Test Organism			
	Metric 13: Test Organism Characteristics	Low	The source of test organisms was not reported.
	Metric 14: Acclimatization and Pretreatment Conditions	Low	Acclimation of test organisms prior to exposure was not reported.
	Metric 15: Number of Organisms and Replicates per Group	Medium	Tests with DBP utilized three replicates of five different concentrations with 10 organisms per beaker; three sediment control replicates with 10 test organisms per beaker; and three silica sand control replicates with 10 test organisms per beaker.
Domain 5: Outcome Assessment			
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Study Citation:	Call, D. J., Cox, D. A., Geiger, D. L., Genisot, K. I., Markee, T. P., Brooke, L. T., Polkinghorne, C. N., Vandeventer, F. A., Gorsuch, J. W., Robillard, K. A., Parkerton, T. F., Reiley, M. C., Ankley, G. T., Mount, D. R. (2001). An assessment of the toxicity of phthalate esters to freshwater benthos. 2. Sediment exposures. Environmental Toxicology and Chemistry 20(8):1805-1815.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Sediment; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Chironomus tentans</i> ; Larvae			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	679311			
Domain	Metric	Rating	Comments	
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions including feeding, intermittent water renewal system, lighting, and temperature were reported. Authors recorded temperature, dissolved oxygen, and pH on days 0 and 10 and conductivity, hardness, alkalinity, and ammonia on days 1 and 9. Sediment TOC conditions were described in Table 2.	
	Metric 17: Outcome Assessment Methodology	High	Sediment was sieved, survivors collected, dried, and weighed.	
	Metric 18: Consistency of Outcome Assessment	High	The outcome assessment was conducted at the conclusion of the 10-day exposure.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.	
	Metric 20: Outcomes Unrelated to Exposure	High	Details regarding test organism attrition and outcomes unrelated to exposure were reported for each study group, and there were no differences among groups that could influence the outcome assessment.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Survival data from toxicity tests were summarized using the trimmed Spearman–Kärber method. Dry weight data were analyzed by one-way analysis of variance and Dunnett’s procedure using a SigmaStat Program.	
	Metric 22: Reporting of Data	High	Treatment and control data were reported in Table 4. Results were represented as average dry weight per individual.	
	Metric 23: Explanation of Unexpected Outcomes	Medium	Variability was not reported, but results suggest no excessive variability within replicates.	
Additional Comments: None				
Overall Quality Determination		High		

Study Citation:	Call, D. J., Markee, T. P., Geiger, D. L., Brooke, L. T., Vandeventer, F. A., Cox, D. A., Genisot, K. I., Robillard, K. A., Gorsuch, J. W., Parkerton, T. F., Reiley, M. C., Ankley, G. T., Mount, D. R. (2001). An assessment of the toxicity of phthalate esters to freshwater benthos. 1. Aqueous exposures. Environmental Toxicology and Chemistry 20(8):1798-1804.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Chironomus tentans</i> ; Larvae			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	679312			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	Chemical was identified by name, no CASRN or structure.	
	Metric 2: Test Substance Source	High	The source of the phthalate was Aldrich Chemical. The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity reported as >98%	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was adequate	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail	
	Metric 8: Consistency of Exposure Administration	High	exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and exposure frequency were reported and appropriate for the study type	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were adequate for a dose response	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms,	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects	
Domain 5: Outcome Assessment				
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Study Citation:	Call, D. J., Markee, T. P., Geiger, D. L., Brooke, L. T., Vandeventer, F. A., Cox, D. A., Genisot, K. I., Robillard, K. A., Gorsuch, J. W., Parkerton, T. F., Reiley, M. C., Ankley, G. T., Mount, D. R. (2001). An assessment of the toxicity of phthalate esters to freshwater benthos. 1. Aqueous exposures. Environmental Toxicology and Chemistry 20(8):1798-1804.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Chironomus tentans</i> ; Larvae			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	679312			
Domain	Metric	Rating	Comments	
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest	
	Metric 18: Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions	
	Metric 20: Outcomes Unrelated to Exposure	High	there were no differences among groups that could influence the outcome assessment.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical methods were adequately described	
	Metric 22: Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group, but results were described in the text.	
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes,	
Additional Comments:	None			
Overall Quality Determination		High		

Study Citation:	Call, D. J., Markee, T. P., Geiger, D. L., Brooke, L. T., Vandeventer, F. A., Cox, D. A., Genisot, K. I., Robillard, K. A., Gorsuch, J. W., Parkerton, T. F., Reiley, M. C., Ankley, G. T., Mount, D. R. (2001). An assessment of the toxicity of phthalate esters to freshwater benthos. 1. Aqueous exposures. Environmental Toxicology and Chemistry 20(8):1798-1804.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Chironomus tentans</i> ; Larvae			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	679312			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The chemical was identified by name only. There was no CASRN or structure reported.	
	Metric 2: Test Substance Source	High	The source of the phthalate was Aldrich Chemical. The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity was reported as >98%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was adequate.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and exposure frequency were reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were adequate for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	
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Study Citation:	Call, D. J., Markee, T. P., Geiger, D. L., Brooke, L. T., Vandeventer, F. A., Cox, D. A., Genisot, K. I., Robillard, K. A., Gorsuch, J. W., Parkerton, T. F., Reiley, M. C., Ankley, G. T., Mount, D. R. (2001). An assessment of the toxicity of phthalate esters to freshwater benthos. 1. Aqueous exposures. Environmental Toxicology and Chemistry 20(8):1798-1804.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Chironomus tentans</i> ; Larvae			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	679312			
Domain	Metric	Rating	Comments	
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.	
	Metric 20: Outcomes Unrelated to Exposure	High	There were no differences among groups that could influence the outcome assessment.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.	
	Metric 22: Reporting of Data	Medium	Mean data for exposure-related findings were shown for each treatment and control group.	
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.	
Additional Comments:	The LC50 of DBP for Chironomus was reported as 2.64 mg/L.			

Overall Quality Determination**High**

Study Citation:	Lake Superior Research Institute, (1997). Sediment toxicity testing program for phthalate esters.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Chironomus tentans</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	7325945			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The chemical was identified by name, lot # and CAS#.	
	Metric 2: Test Substance Source	Low	The test substance identity was NOT analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity was reported as >99%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable.	
	Metric 6: Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.	
	Metric 8: Consistency of Exposure Administration	High	Details of exposure administration were reported, and exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 10 organisms per replicate with two replicates used.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	
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Study Citation:	Lake Superior Research Institute, (1997). Sediment toxicity testing program for phthalate esters.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Chironomus tentans</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	7325945			
Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	High	There were no differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described.
	Metric 22:	Reporting of Data	Medium	Data for exposure-related findings were not shown for each treatment and control group, but results were described in the tables.
	Metric 23:	Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.
Additional Comments: None				

Overall Quality Determination**High**

Study Citation:	Lake Superior Research Institute, (1997). Sediment toxicity testing program for phthalate esters.		
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Sediment; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Chironomus tentans</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Mortality		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	7325945		
Domain	Metric	Rating	Comments
Domain 1: Test Substance			
	Metric 1: Test Substance Identity	High	The chemical was identified by name, lot # and CAS#.
	Metric 2: Test Substance Source	Low	The test substance identity was NOT analytically verified by the performing laboratory.
	Metric 3: Test Substance Purity	High	Chemical purity was reported as >99%.
Domain 2: Test Design			
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable.
	Metric 6: Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups.
Domain 3: Exposure Characterization			
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8: Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods.
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit.
Domain 4: Test Organism			
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 10 organisms per replicate with two replicates used.
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
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Study Citation:	Lake Superior Research Institute, (1997). Sediment toxicity testing program for phthalate esters.		
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Sediment; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Chironomus tentans</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Mortality		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	7325945		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
Metric 20:	Outcomes Unrelated to Exposure	High	There were no differences among groups.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	High	Statistical methods were adequately described.
Metric 22:	Reporting of Data	High	Data for exposure-related findings were shown for each treatment and control group.
Metric 23:	Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.
Additional Comments:	None		

Overall Quality Determination**High**

Study Citation:	Lake Superior Research Institute, (1997). Sediment toxicity testing program for phthalate esters.		
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Sediment; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Chironomus tentans</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Mortality		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	7325945		
Domain	Metric	Rating	Comments
Domain 1: Test Substance			
	Metric 1: Test Substance Identity	High	The chemical was identified by name, lot # and CAS#.
	Metric 2: Test Substance Source	Low	The test substance identity was NOT analytically verified by the performing laboratory.
	Metric 3: Test Substance Purity	High	Chemical purity was reported as >99%.
Domain 2: Test Design			
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable.
	Metric 6: Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups.
Domain 3: Exposure Characterization			
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8: Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods.
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit.
Domain 4: Test Organism			
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 10 organisms per replicate with two replicates used.
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
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Study Citation:	Lake Superior Research Institute, (1997). Sediment toxicity testing program for phthalate esters.		
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Sediment; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Chironomus tentans</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Mortality		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	7325945		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
Metric 20:	Outcomes Unrelated to Exposure	High	There were no differences among groups.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	High	Statistical methods were adequately described.
Metric 22:	Reporting of Data	High	Data for exposure-related findings were shown for each treatment and control group.
Metric 23:	Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.
Additional Comments:	None		

Overall Quality Determination**High**

Study Citation:	Lake Superior Research Institute, (1997). Sediment toxicity testing program for phthalate esters.		
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Sediment; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Chironomus tentans</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Mortality		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	7325945		
Domain	Metric	Rating	Comments
Domain 1: Test Substance			
	Metric 1: Test Substance Identity	High	The chemical was identified by name, lot # and CAS#.
	Metric 2: Test Substance Source	Low	The test substance identity was NOT analytically verified by the performing laboratory.
	Metric 3: Test Substance Purity	High	Chemical purity was reported as >99%.
Domain 2: Test Design			
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable.
	Metric 6: Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups.
Domain 3: Exposure Characterization			
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8: Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods.
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit.
Domain 4: Test Organism			
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 10 organisms per replicate with two replicates used.
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
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Study Citation:	Lake Superior Research Institute, (1997). Sediment toxicity testing program for phthalate esters.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Sediment; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Chironomus tentans</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	7325945

Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
Metric 20:	Outcomes Unrelated to Exposure	High	There were no differences among groups.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	High	Statistical methods were adequately described.
Metric 22:	Reporting of Data	High	Data for exposure-related findings were shown for each treatment and control group.
Metric 23:	Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.

Additional Comments: None

Overall Quality Determination**High**

Study Citation:	Adams, W. J., Biddinger, G. R., Robillard, K. A., Gorsuch, J. W. (1995). A summary of the acute toxicity of 14 phthalate esters to representative aquatic organisms. Environmental Toxicology and Chemistry 14(9):1569-1574.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Immobilization			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1321996; Linked HERO ID(s): 1321996, 1316224			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The test substance nomenclature was reported without a CASRN.
	Metric 2:	Test Substance Source	Low	The source was reported as provided by a manufacturer from commercially available batches. The manufacture name and batch number were not provided. No analytical data were reported.
	Metric 3:	Test Substance Purity	High	
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	A negative control was reported.
	Metric 5:	Negative Control Response	High	The control response was acceptable.
	Metric 6:	Randomized Allocation	Low	An allocation method was not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The experimental system was well described. However, headspace or measures to prevent volatilization were not reported.
	Metric 8:	Consistency of Exposure Administration	High	The exposure administration was consistent across groups.
	Metric 9:	Measurement of Test Substance Concentration	Medium	Sample extracts were analyzed by gas chromatography at the start and end of the test. In static studies, final test concentrations frequently were 50% of the initial concentrations. Loss of the phthalate esters was thought to be principally due to adsorption to the test vessels.
	Metric 10:	Exposure Duration and Frequency	High	Duration and frequency of exposure were appropriate for the test.
	Metric 11:	Number of Exposure Groups/Spacing of Exposure Levels	High	Exposure levels were appropriate. A range finding test was performed.
	Metric 12:	Testing at or Below Solubility Limit	High	The test was performed at or below water solubility.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Appropriate acclimation for the test was reported.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Each test was performed using duplicate test concentrations with 10 organisms per test vessel.
Domain 5: Outcome Assessment				
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Study Citation:	Adams, W. J., Biddinger, G. R., Robillard, K. A., Gorsuch, J. W. (1995). A summary of the acute toxicity of 14 phthalate esters to representative aquatic organisms. Environmental Toxicology and Chemistry 14(9):1569-1574.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Immobilization			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1321996; Linked HERO ID(s): 1321996, 1316224			
Domain	Metric	Rating	Comments	
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions were appropriate for the test.	
	Metric 17: Outcome Assessment Methodology	High	Intended outcomes were reported.	
	Metric 18: Consistency of Outcome Assessment	High	Outcome assessment was consistent across groups.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	Environmental conditions were consistent across groups.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There were no reported differences between groups.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical methods were performed and described.	
	Metric 22: Reporting of Data	Medium	Only treatment endpoints were reported.	
	Metric 23: Explanation of Unexpected Outcomes	High	No unexpected outcomes were reported.	
Additional Comments:	None			
Overall Quality Determination		High		

Study Citation:	Bionomics., Springborn (1984). Acute toxicity of fourteen phthalate esters to <i>Daphnia magna</i> (final report).			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1316223			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Medium	The chemical is identified by name and CASRN, but no other verification is provided.
	Metric 2:	Test Substance Source	Low	Chemicals were provided by the General Electric Company in 1-L amber glass bottles. No analytical verification or additional information was provided.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of the test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group (i.e. all conditions equal except chemical exposure).
	Metric 5:	Negative Control Response	High	No mortality or adverse effects were reported in the controls.
	Metric 6:	Randomized Allocation	Medium	Test organisms were impartially distributed among test containers.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Uninformative	The experimental system and/or test media preparation methods were adequately reported but did not account for physical-chemical properties, specifically the low solubility in the test medium and rapid degradation rate. The authors measured the test concentrations throughout the test, but the rate of degradation was very high, resulting in a final test concentration below the limit of detection of the measurement device. As a result, it cannot be determined whether the lack of mortality observed in the study is the result of the toxicity of the test material, or the lack of test material in the exposure vessel. Furthermore, because the final concentration is reported as a non definitive value (<), a mean-measured concentration cannot be determined.
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were measured using appropriate analytical technologies and methods. Analytical technologies used were highly sensitive (GC-MS), but not sensitive enough to measure the concentration in the corroborative test.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type and/or outcome(s) of interest (acute <i>Daphnia</i> study of 48-hour duration).
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	Test concentrations were spaced with the intention of bracketing the limit of solubility, but the actual measured test concentrations were very low and insufficient to capture an endpoint.
	Metric 12:	Testing at or Below Solubility Limit	Uninformative	Exposure concentrations were seemingly above the water solubility limit in the initial test. The authors reported that the initial test featured a film that captured and killed a significant number of the daphnia. A solvent should have been used.

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Study Citation:	Bionomics., Springborn (1984). Acute toxicity of fourteen phthalate esters to <i>Daphnia magna</i> (final report).			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1316223			
Domain	Metric	Rating	Comments	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source (and sex if relevant) of the test animals was not reported.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized and/or whether pre-treatment conditions were the same for control and exposed groups. As excess mortality was not observed in the controls, this was not determined to adversely affect the outcome.	
	Metric 15: Number of Organisms and Replicates per Group	Low	Five daphnia/replicate is far lower than the guideline required 20 daphnia/replicate.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Organism housing, environmental conditions, food, water, and nutrients were conducive to maintenance of health, and biomass loading was appropriate.	
	Metric 17: Outcome Assessment Methodology	Uninformative	Significant deficiencies in the reported outcome assessment methodology were identified. The test was intended to quantify the mortality up to the limit of solubility of the chemical. This was not accomplished due to the extremely low solubility in the test medium. In the first definitive test, the authors report that a film of insoluble test material formed on the surface that entrapped some of the test organisms and caused mortality. In the follow-up, corroborative test, another method was used to reduce the occurrence of the film, but the test material completely degraded by the end of that test, rendering it impossible to draw a conclusion about the toxicity of the substance, as no mortality was observed.	
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.	
	Metric 20: Outcomes Unrelated to Exposure	Uninformative	A film of insoluble test material caused mortality in the test organisms.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	N/A	No mortality was observed so statistical analysis was not needed.	
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group and were adequate to determine values for the endpoint(s) of interest.	
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Study Citation:	Bionomics., Springborn (1984). Acute toxicity of fourteen phthalate esters to <i>Daphnia magna</i> (final report).			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1316223			
Domain	Metric	Rating	Comments	
	Metric 23: Explanation of Unexpected Outcomes	Low	The authors did not conduct a solubility test in the test medium that would have explained why the measured test concentrations were so low in comparison to the reported solubility of the test material elsewhere. In addition, there was significant, non-dose dependent mortality observed in the lower concentrations of the initial test that was not explained.	
Additional Comments:	This study result should be interpreted with caution. This test was comprised of two parts- a definitive and corroborative test. The definitive test was not valid because a film of insoluble test material formed on the surface of the test medium, causing excess mortality. In the corroborative test, the material completely degraded by the end of the test, and no mortality was observed.			

Overall Quality Determination**Uninformative**

Study Citation:	Huang, B., Li, D., Yang, Y. (2016). Joint toxicity of two phthalates with waterborne copper to <i>Daphnia magna</i> and <i>Photobacterium phosphoreum</i> . Bulletin of Environmental Contamination and Toxicology 97(3):380-386.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Juvenile			
Health Outcome:	Immobilization			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5750702			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The test substance was identified by chemical name and CASRN.
	Metric 2:	Test Substance Source	Low	The source was reported but the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Medium	It was mentioned that chemicals used were analytical grade.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was adequate.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	Test media preparation methods were reported but did not provide the measures taken to minimize loss of test substance before and during the exposure. Concentration of test substance was measured during the study.
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using HPLC. The percent recovery of spiked test substance was within acceptable range. Measured concentrations were similar to nominal concentrations.
	Metric 10:	Exposure Duration and Frequency	Medium	Minor limitations in exposure frequency and duration of exposure were identified (acute daphnid toxicity study of 24-hour duration as opposed to 48 hours).
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Seven exposure groups and a control were tested.
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate. No effects on biological responses were observed in the solvent control.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions and all pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	There were ten daphnids per test vessel and they were tested in triplicates.

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Study Citation:	Huang, B., Li, D., Yang, Y. (2016). Joint toxicity of two phthalates with waterborne copper to <i>Daphnia magna</i> and <i>Photobacterium phosphoreum</i> . Bulletin of Environmental Contamination and Toxicology 97(3):380-386.		
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Juvenile		
Health Outcome:	Immobilization		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	5750702		
Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
Metric 16:	Adequacy of Test Conditions	High	Organism housing, environmental conditions, food and biomass loading seem to be conducive to the maintenance of health. The authors followed the protocol outlined in the the National Standard Method of China (GB/T 13266-1991).
Metric 17:	Outcome Assessment Methodology	Low	The outcome (immobilization of daphnids) assessment methodology was not clearly reported.
Metric 18:	Consistency of Outcome Assessment	High	Immobilization was recorded after 24 hours of exposure in all treatment groups and the control.
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study on outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately. The methods for calculating effect ratio (y-axis of Fig 1) and EC 50 values were not provided.
Metric 22:	Reporting of Data	Medium	Data for exposure-related findings were presented for each treatment and control group via figures. The methods for calculating effect ratio (y-axis of Fig 1) were not provided. EC 50 values were given without confidence intervals in the text .
Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments: Data analysis methods were not provided. EC 50 values were given without confidence intervals.			
Overall Quality Determination		High	

Study Citation:	Huang, G. L., Sun, H. W., Song, Z. H. (1999). Interactions between dibutyl phthalate and aquatic organisms. Bulletin of Environmental Contamination and Toxicology 63(6):759-765.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Larvae			
Health Outcome:	Nutritional & Metabolic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5551982			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The chemical was identified by name only.
	Metric 2:	Test Substance Source	Low	The source was not reported.
	Metric 3:	Test Substance Purity	Low	The purity and grade of the test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	A concurrent negative control group was reported.
	Metric 5:	Negative Control Response	High	A concurrent negative control group response was reported and suitable.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.
	Metric 8:	Consistency of Exposure Administration	Low	Only general methods of exposure administration were reported, so it was difficult to determine if the assessment was carried out consistently.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	Low	The exposure duration was not entirely clear. The duration was at least 10 hours.
	Metric 11:	Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable.
	Metric 12:	Testing at or Below Solubility Limit	Medium	A subset of the exposure concentrations exceeded the water solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups.
	Metric 15:	Number of Organisms and Replicates per Group	Low	There were five replicates with 10 daphnids each.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if they were adequate.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology was reported and adequate.

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Study Citation:	Huang, G. L., Sun, H. W., Song, Z. H. (1999). Interactions between dibutyl phthalate and aquatic organisms. Bulletin of Environmental Contamination and Toxicology 63(6):759-765.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Larvae
Health Outcome:	Nutritional & Metabolic
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	5551982

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were shown for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability.

Additional Comments: This evaluation is for digestive rate.

Overall Quality Determination

Low

Study Citation:	Huang, G. L., Sun, H. W., Song, Z. H. (1999). Interactions between dibutyl phthalate and aquatic organisms. Bulletin of Environmental Contamination and Toxicology 63(6):759-765.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Larvae			
Health Outcome:	Behavioral			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5551982			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The source was not reported.	
	Metric 3: Test Substance Purity	Low	The purity and grade of the test substance were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	A concurrent negative control group was reported.	
	Metric 5: Negative Control Response	High	A concurrent negative control group response was reported and suitable.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.	
	Metric 8: Consistency of Exposure Administration	Low	Only general methods of exposure administration were reported, so it was difficult to determine if the assessment was carried out consistently.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	Low	The exposure duration was not entirely clear. The duration was at least 10 hours.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable.	
	Metric 12: Testing at or Below Solubility Limit	Medium	A subset of the exposure concentrations exceeded the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups.	
	Metric 15: Number of Organisms and Replicates per Group	Low	There were five replicates with 10 daphnids each.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if they were adequate.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology was reported and adequate.	
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.	
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Study Citation:	Huang, G. L., Sun, H. W., Song, Z. H. (1999). Interactions between dibutyl phthalate and aquatic organisms. Bulletin of Environmental Contamination and Toxicology 63(6):759-765.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Larvae
Health Outcome:	Behavioral
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	5551982

Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were shown for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability.

Additional Comments: This evaluation form was for feeding behavior-filter rate.

Overall Quality Determination

Low

Study Citation:	Huang, G. L., Sun, H. W., Song, Z. H. (1999). Interactions between dibutyl phthalate and aquatic organisms. Bulletin of Environmental Contamination and Toxicology 63(6):759-765.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Larvae			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5551982			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The chemical was identified by name only.
	Metric 2:	Test Substance Source	Low	The source was not reported.
	Metric 3:	Test Substance Purity	Low	The purity and grade of the test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Uninformative	A concurrent negative control group was not included or reported.
	Metric 5:	Negative Control Response	Uninformative	A concurrent negative control group was not included, no response was reported.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.
	Metric 8:	Consistency of Exposure Administration	Low	Only general methods of exposure administration were reported, so it was difficult to determine if the assessment was carried out consistently.
	Metric 9:	Measurement of Test Substance Concentration	Uninformative	Exposure concentrations were not measured, and nominal values are highly uncertain due to the nature of the test substance.
	Metric 10:	Exposure Duration and Frequency	Medium	The exposure duration was shorter than a standard duration.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Uninformative	No information is provided on the number of exposure groups or the spacing of exposure levels.
	Metric 12:	Testing at or Below Solubility Limit	Low	It was unclear if exposure concentrations exceeded the water solubility limit as they were not reported.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	There are minor uncertainties about the source of test organisms.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The F0 test organisms were acclimatized to test conditions.
	Metric 15:	Number of Organisms and Replicates per Group	Low	There were three replicates, but the initial number of organisms was not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if they were adequate.
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported.
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Study Citation:	Huang, G. L., Sun, H. W., Song, Z. H. (1999). Interactions between dibutyl phthalate and aquatic organisms. Bulletin of Environmental Contamination and Toxicology 63(6):759-765.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Larvae			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5551982			
Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22:	Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group.
	Metric 23:	Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability.
Additional Comments:	Tests were repeated for different temperature, hardness, and humic acid concentrations. None of the results reported the use of controls, and all would be considered Unacceptable.			

Overall Quality Determination**Uninformative**

Study Citation:	Jonsson, S., Baun, A. (2003). Toxicity of mono- and diesters of o-phthalic esters to a crustacean, a green alga, and a bacterium. Environmental Toxicology and Chemistry 22(12):3037-3043.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Larvae			
Health Outcome:	Immobilization			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	789536			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The test substance was identified definitively (nomenclature, CASRN, and structure were reported).
	Metric 2:	Test Substance Source	High	DBP was purchased from Merck Eurolab (Stockholm, Sweden) but the test substance identity was NOT analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Percent purity was reported as >99%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	Low	The biological response of the control group was not clearly reported. It was stated that tests were repeated if the mortality in the control group exceeded 5%.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations and/or minimize loss of test substance before and during the exposure.
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were measured but not reported.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Uninformative	No information is provided on the exposure concentrations and the spacing of exposure levels.
	Metric 12:	Testing at or Below Solubility Limit	Low	Reporting omissions prevented determination of whether exposure concentrations exceeded the water solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of test animals were not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized and/or whether pre-treatment conditions were the same for control and exposed groups.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	There were four replicates per concentration and five animals were used in each replicate.

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Study Citation:	Jonsson, S., Baun, A. (2003). Toxicity of mono- and diesters of o-phthalic esters to a crustacean, a green alga, and a bacterium. Environmental Toxicology and Chemistry 22(12):3037-3043.		
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Larvae		
Health Outcome:	Immobilization		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	789536		
Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate and whether differences occurred between control and exposed populations.
Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported.
Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study on outcomes unrelated to the exposure.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	High	EC10 and EC50 values were calculated using probit analysis with maximum likelihood estimation.
Metric 22:	Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group.
Metric 23:	Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained. Authors noted surface entrapment of test animals in the highest tested concentration but these were excluded from the calculation of EC10 and EC50 values.
Additional Comments:	The exposure concentrations, spacing of exposure levels and control response were not reported. Measured concentrations were not reported. Mortality data were not provided for each of the treatment groups and control. Only 24 and 48 hour EC10 and EC50 values were reported.		
Overall Quality Determination		Uninformative	

Study Citation:	Mccarthy, J. F., Whitmore, D. K. (1985). Chronic toxicity of di-n-butyl and di-n-octyl phthalate to daphnia-magna and the fathead minnow. Environmental Toxicology and Chemistry 4(2):167-179.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1336024			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity was reported as 99.5 %.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	High	No mortalities were reported.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.	
	Metric 8: Consistency of Exposure Administration	Low	No details were provided.	
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured after Day 1 and reported in Table 2.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	Medium	Most exposure concentrations were below the water solubility limit. The high concentration was close.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source. "Test animals for both the acute immobilization tests (range finding test) and the reproduction test were collected when the animals were less than 24 h old."	
	Metric 14: Acclimatization and Pretreatment Conditions	Medium	Pretreatment conditions were the same for control and exposed organisms, but details were limited.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were five daphnids per treatment with two replicates.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Medium	Environmental conditions were not sufficiently reported to evaluate if adequate.	

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Study Citation:	Mccarthy, J. F., Whitmore, D. K. (1985). Chronic toxicity of di-n-butyl and di-n-octyl phthalate to daphnia-magna and the fathead minnow. Environmental Toxicology and Chemistry 4(2):167-179.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Juvenile
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	1336024

Domain	Metric	Rating	Comments
	Metric 17: Outcome Assessment Methodology	Medium	The outcome assessment methodology was reported in the text.
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group in text form.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments:	Range finding test:"In the acute mortality test (range-finding test), all D. magna were dead after 48 h of exposure to nominal concentrations of 7.5 and 10.0 mg/L DBP. At the lower doses of 3.0, 1.0 and 0.5 mg/L DBP and in controls, all animals survived, except for one individual at 3.0 mg/L. The LC50 (lethal concentration to 50% of the test population) is between 3.0 and 7.5 mg/L DBP. Although a probit analysis cannot be performed, because this procedure requires two responses that are between 0 and 100% mortality, a nonparametric analysis was developed for steep dose-response bioassays (Schmoyer, Beauchamp and McCarthy, manuscript in preparation). The LC50 was estimated using this method and was equal to 5.2 mg/L, with 95% confidence limits of 4.7 and 5.6 mg/L."		

Overall Quality Determination**Medium**

Study Citation:	Seyoum, A., Pradhan, A. (2019). Effect of phthalates on development, reproduction, fat metabolism and lifespan in <i>Daphnia magna</i> . Science of the Total Environment 654:969-977.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Embryo			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5043468			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	Chemical was identified by name and CAS#	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity reported as >99%	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group	
	Metric 5: Negative Control Response	Low	The biological response of the negative control groups was not reported	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations	
	Metric 8: Consistency of Exposure Administration	Medium	Reporting omissions are unlikely to have a substantial impact on results	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure (96 hr) was reported and appropriate for the study type	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Low	Only two exposure groups were reported	
	Metric 12: Testing at or Below Solubility Limit	High	The solvent concentration was appropriate	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The number of organisms and replicates were suitable	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Details of environmental conditions of test system were not reported	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome	
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Study Citation:	Seyoum, A., Pradhan, A. (2019). Effect of phthalates on development, reproduction, fat metabolism and lifespan in <i>Daphnia magna</i> . Science of the Total Environment 654:969-977.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Embryo			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5043468			
Domain	Metric	Rating	Comments	
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups	
	Metric 20: Outcomes Unrelated to Exposure	High	there were no differences among groups	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	Uninformative	Statistical analysis was not conducted.	
	Metric 22: Reporting of Data	Uninformative	Data presentation was inadequate	
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcome	
Additional Comments:	with ehippia			

Overall Quality Determination**Uninformative**

Study Citation:	Seyoum, A., Pradhan, A. (2019). Effect of phthalates on development, reproduction, fat metabolism and lifespan in <i>Daphnia magna</i> . Science of the Total Environment 654:969-977.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Juvenile			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5043468			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	Chemical was identified by name and CAS#	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity reported as >99%	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group	
	Metric 5: Negative Control Response	High	The biological response of the negative control groups was reported	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations	
	Metric 8: Consistency of Exposure Administration	Medium	Reporting omissions are unlikely to have a substantial impact on results	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Low	Only two exposure groups were reported	
	Metric 12: Testing at or Below Solubility Limit	High	The solvent concentration was appropriate	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The number of organisms and replicates were suitable	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Details of environmental conditions of test system were not reported	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome	
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited	
Domain 6: Confounding / Variable Control				
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Study Citation:	Seyoum, A., Pradhan, A. (2019). Effect of phthalates on development, reproduction, fat metabolism and lifespan in <i>Daphnia magna</i> . Science of the Total Environment 654:969-977.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Juvenile
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	5043468

Domain	Metric	Rating	Comments
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups
	Metric 20: Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Statistical analysis was performed but not described adequately
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcome

Additional Comments: 24 hr exposure, measured at 96 hr

Overall Quality Determination**Medium**

Study Citation:	Seyoum, A., Pradhan, A. (2019). Effect of phthalates on development, reproduction, fat metabolism and lifespan in <i>Daphnia magna</i> . Science of the Total Environment 654:969-977.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5043468			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	Chemical was identified by name and CAS#	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity reported as >99%	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group	
	Metric 5: Negative Control Response	Low	The biological response of the negative control groups was not reported	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations	
	Metric 8: Consistency of Exposure Administration	Medium	Reporting omissions are unlikely to have a substantial impact on results	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure (48 hr) was reported and appropriate for the study type	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Low	Only two exposure groups were reported	
	Metric 12: Testing at or Below Solubility Limit	High	The solvent concentration was appropriate	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The number of exposure groups and spacing of exposure levels were suitable	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Details of environmental conditions of test system were not reported	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome	
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited	
Domain 6: Confounding / Variable Control				
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Study Citation:	Seyoum, A., Pradhan, A. (2019). Effect of phthalates on development, reproduction, fat metabolism and lifespan in <i>Daphnia magna</i> . Science of the Total Environment 654:969-977.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5043468			
Domain	Metric		Rating	Comments
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups
	Metric 20:	Outcomes Unrelated to Exposure	High	There were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Uninformative	Statistical analysis was not conducted.
	Metric 22:	Reporting of Data	Uninformative	Data presentation was inadequate
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcome
Additional Comments: None				

Overall Quality Determination**Uninformative**

Study Citation:	Seyoum, A., Pradhan, A. (2019). Effect of phthalates on development, reproduction, fat metabolism and lifespan in <i>Daphnia magna</i> . Science of the Total Environment 654:969-977.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Juvenile			
Health Outcome:	Nutritional & Metabolic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5043468			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	Chemical was identified by name and CAS#	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity reported as >99%	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group	
	Metric 5: Negative Control Response	High	The biological response of the negative control groups was reported	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations	
	Metric 8: Consistency of Exposure Administration	Medium	Reporting omissions are unlikely to have a substantial impact on results	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Low	Only two exposure groups were reported	
	Metric 12: Testing at or Below Solubility Limit	High	The solvent concentration was appropriate	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The number of organisms and replicates were suitable	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Details of environmental conditions of test system were not reported	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome	
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited	
Domain 6: Confounding / Variable Control				
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Study Citation:	Seyoum, A., Pradhan, A. (2019). Effect of phthalates on development, reproduction, fat metabolism and lifespan in <i>Daphnia magna</i> . Science of the Total Environment 654:969-977.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Juvenile
Health Outcome:	Nutritional & Metabolic
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	5043468

Domain	Metric	Rating	Comments
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups
	Metric 20: Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Statistical analysis was performed but not described adequately
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcome

Additional Comments: lipid accumulation

Overall Quality Determination**Medium**

Study Citation:	Seyoum, A., Pradhan, A. (2019). Effect of phthalates on development, reproduction, fat metabolism and lifespan in <i>Daphnia magna</i> . Science of the Total Environment 654:969-977.			
Duration:	Overall Duration: > 21 days; Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Juvenile			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5043468			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The chemical was identified by name and CAS#.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity was reported as >99%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control groups was reported.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.	
	Metric 8: Consistency of Exposure Administration	Medium	Reporting omissions are unlikely to have a substantial impact on results.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Low	Only two exposure groups were reported.	
	Metric 12: Testing at or Below Solubility Limit	High	The solvent concentration was appropriate.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The number of organisms and replicates were suitable.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Details of environmental conditions of the test system were not reported.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome.	
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.	
Domain 6: Confounding / Variable Control				
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Study Citation:	Seyoum, A., Pradhan, A. (2019). Effect of phthalates on development, reproduction, fat metabolism and lifespan in <i>Daphnia magna</i> . Science of the Total Environment 654:969-977.
Duration:	Overall Duration: > 21 days; Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Juvenile
Health Outcome:	Reproductive/Teratogenic
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	5043468

Domain	Metric	Rating	Comments
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups.
	Metric 20: Outcomes Unrelated to Exposure	High	There were no differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This form is evaluating the progeny results.

Overall Quality Determination

Medium

Study Citation:	Shen, C., Wei, J., Wang, T., Wang, Y. (2019). Acute toxicity and responses of antioxidant systems to dibutyl phthalate in neonate and adult <i>Daphnia magna</i> . PeerJ 7(3):e6584.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Adult			
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5433053			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
Metric 1:	Test Substance Identity	High	The DBP was identified by CASRN.	
Metric 2:	Test Substance Source	High	The source of the DBP was reported to be Sinopharm Chemical Reagent Co. Ltd, and verified by GC-MS.	
Metric 3:	Test Substance Purity	Low	The purity/grade of the DBP was not reported.	
Domain 2: Test Design				
Metric 4:	Negative Controls	High	Study authors reported the use of a negative control without DBP and a solvent control.	
Metric 5:	Negative Control Response	High	The negative control responses were reported in Figures 1-6 and were appropriate for the outcomes of interest.	
Metric 6:	Randomized Allocation	Low	It was not reported how the Daphnids were allocated into study groups.	
Domain 3: Exposure Characterization				
Metric 7:	Experimental System/Test Media Preparation	Low	Limited details were provided on the preparation of the test substance. The test system was a static system.	
Metric 8:	Consistency of Exposure Administration	High	All tests were for 24 or 48h in 100mL of test solution with a 12L:12D photoperiod at 22C. Tests were conducted in dechlorinated water.	
Metric 9:	Measurement of Test Substance Concentration	High	Test concentrations were measured using GC-MS.	
Metric 10:	Exposure Duration and Frequency	High	The exposure duration was reported to be 48h with assessments conducted at 24 and 48h.	
Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	There were only two exposure levels reported, which is lower than is typical. Spacing was adequate to observe a response.	
Metric 12:	Testing at or Below Solubility Limit	High	Study authors reported the use of a vehicle solvent and had a solvent control with an appropriate amount of solvent.	
Domain 4: Test Organism				
Metric 13:	Test Organism Characteristics	High	The test organisms were reported to be from the Key Laboratory of Hydrobiology at Dalian Ocean University. The age of the <i>Daphnia</i> was appropriate for the study.	
Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the <i>Daphnia</i> were acclimated to test conditions.	
Metric 15:	Number of Organisms and Replicates per Group	Medium	There were 10 <i>Daphnia</i> per test chamber, and each concentration was tested with five replicates.	
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Study Citation:	Shen, C., Wei, J., Wang, T., Wang, Y. (2019). Acute toxicity and responses of antioxidant systems to dibutyl phthalate in neonate and adult <i>Daphnia magna</i> . PeerJ 7(3):e6584.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Adult
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	5433053

Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	The tests were conducted with dechlorinated water at 22C with a 12L:12D photoperiod. Prior to the study, the Daphnids were fed <i>Scenedesmus</i> sp. They were not fed for the duration of the test.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—changes in H2O2 concentrations, lipid peroxidation, T-AOC, SOD, CAT, and GST.
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions. It was not reported if the organisms were acclimated.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods included two-way ANOVA with Duncan's multiple range test.
	Metric 22: Reporting of Data	High	The biological control response and the exposure response were reported in Figures 1-6 and were appropriate for the outcomes of interest.
	Metric 23: Explanation of Unexpected Outcomes	High	The study authors did not report any unexpected outcomes. Variability was reported in the figures.
Additional Comments:	This portion of the evaluation was on the effect of DBP on H2O2, lipid peroxidation, T-AOC, SOD, CAT, and GST in <i>D. magna</i> adults. The mechanistic outcome of oxidative stress was selected as the outcome of interest. The supplemental data was not included in the Distiller download, but was provided via a link in the paper.		

Overall Quality Determination**High**

Study Citation:	Shen, C., Wei, J., Wang, T., Wang, Y. (2019). Acute toxicity and responses of antioxidant systems to dibutyl phthalate in neonate and adult <i>Daphnia magna</i> . PeerJ 7(3):e6584.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Juvenile
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	5433053

Domain	Metric	Rating	Comments
Domain 1: Test Substance			
	Metric 1: Test Substance Identity	High	The DBP was identified by CASRN.
	Metric 2: Test Substance Source	High	The source of the DBP was reported to be Sinopharm Chemical Reagent Co. Ltd, and verified by GC-MS.
	Metric 3: Test Substance Purity	Low	The purity/grade of the DBP was not reported.
Domain 2: Test Design			
	Metric 4: Negative Controls	High	Study authors reported the use of a negative control without DBP and a solvent control.
	Metric 5: Negative Control Response	High	The negative control responses were reported in Figures 1-6 and were appropriate for the outcomes of interest.
	Metric 6: Randomized Allocation	Low	It was not reported how the Daphnids were allocated into study groups.
Domain 3: Exposure Characterization			
	Metric 7: Experimental System/Test Media Preparation	Low	Limited details were provided on the preparation of the test substance. The test system was a static system.
	Metric 8: Consistency of Exposure Administration	High	All tests were for 24 or 48h in 100mL of test solution with a 12L:12D photoperiod at 22C. Tests were conducted in dechlorinated water.
	Metric 9: Measurement of Test Substance Concentration	High	Test concentrations were measured using GC-MS.
	Metric 10: Exposure Duration and Frequency	High	The exposure duration was reported to be 48h with assessments conducted at 24 and 48h.
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Low	There were only 2 exposure levels reported, which is lower than is typical. Spacing was adequate to observe a response.
	Metric 12: Testing at or Below Solubility Limit	High	Study authors reported the use of a vehicle solvent and had a solvent control with an appropriate amount of solvent.
Domain 4: Test Organism			
	Metric 13: Test Organism Characteristics	High	The test organisms were reported to be from the Key Laboratory of Hydrobiology at Dalian Ocean University. The age of the <i>Daphnia</i> was appropriate for the study.
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the <i>Daphnia</i> were acclimated to test conditions.
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 10 <i>Daphnia</i> per test chamber, and each concentrations was tested with 5 replicates.
Domain 5: Outcome Assessment			

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Study Citation:	Shen, C., Wei, J., Wang, T., Wang, Y. (2019). Acute toxicity and responses of antioxidant systems to dibutyl phthalate in neonate and adult <i>Daphnia magna</i> . PeerJ 7(3):e6584.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Juvenile			
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5433053			
Domain	Metric	Rating	Comments	
	Metric 16: Adequacy of Test Conditions	High	The tests were conducted with dechlorinated water at 22C with a 12L:12D photoperiod. Prior to the study, the Daphnids were fed <i>Scenedesmus</i> sp. They were not fed for the duration of the test.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—changes in H2O2 concentrations, lipid peroxidation, T-AOC, SOD, CAT, and GST.	
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions—it was not reported if the organisms were acclimated.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical methods included two-way ANOVA with Duncan’s multiple range test.	
	Metric 22: Reporting of Data	High	The biological control response and the exposure response were reported in Figures 1-6 and were appropriate for the outcomes of interest.	
	Metric 23: Explanation of Unexpected Outcomes	High	The study authors did not report any unexpected outcomes. Variability was reported in the figures.	
Additional Comments:	This portion of the evaluation was on the effect of DBP on H2O2, lipid peroxidation, T-AOC, SOD, CAT, and GST in <i>D. magna</i> neonates. The mechanistic outcome of oxidative stress was selected as the outcome of interest.The supplemental data was not included in the Distiller download, but was provided via a link in the paper.			
Overall Quality Determination		High		

Study Citation:	Shen, C., Wei, J., Wang, T., Wang, Y. (2019). Acute toxicity and responses of antioxidant systems to dibutyl phthalate in neonate and adult <i>Daphnia magna</i> . PeerJ 7(3):e6584.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5433053			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The DBP was identified by CASRN.	
	Metric 2: Test Substance Source	High	The source of the DBP was reported to be Sinopharm Chemical Reagent Co. Ltd, and verified by GC-MS.	
	Metric 3: Test Substance Purity	Low	The purity/grade of the DBP was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of a negative control without DBP and a solvent control.	
	Metric 5: Negative Control Response	High	The negative control response and the solvent control response were both reported in the supplementary data and were adequate.	
	Metric 6: Randomized Allocation	Low	It was not reported how the Daphnids were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	Limited details were provided on the preparation of the test substance. The test system was a static system.	
	Metric 8: Consistency of Exposure Administration	High	All tests were for 24 or 48h in 100mL of test solution with a 12L:12D photoperiod at 22C. Tests were conducted in dechlorinated water.	
	Metric 9: Measurement of Test Substance Concentration	High	Test concentrations were measured using GC-MS.	
	Metric 10: Exposure Duration and Frequency	High	The exposure duration was reported to be 48h with LC50 values calculated at 24 and 48h.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Medium	There were 5 exposure levels reported, but the highest concentrations did not have complete mortality.	
	Metric 12: Testing at or Below Solubility Limit	High	Study authors reported the use of a vehicle solvent and had a solvent control with an appropriate amount of solvent. The solvent control response was adequate for the outcome of interest.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were reported to be from the Key Laboratory of Hydrobiology at Dalian Ocean University. The age of the <i>Daphnia</i> was appropriate for the study.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the <i>Daphnia</i> were acclimated to test conditions.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 10 <i>Daphnia</i> per test chamber, and each concentrations was tested with 5 replicates.	
Domain 5: Outcome Assessment				
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Study Citation:	Shen, C., Wei, J., Wang, T., Wang, Y. (2019). Acute toxicity and responses of antioxidant systems to dibutyl phthalate in neonate and adult <i>Daphnia magna</i> . PeerJ 7(3):e6584.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5433053			
Domain		Metric	Rating	Comments
	Metric 16:	Adequacy of Test Conditions	High	The tests were conducted with dechlorinated water at 22C with a 12L:12D photoperiod. Prior to the study, the Daphnids were fed <i>Scenedesmus</i> sp. They were not fed for the duration of the test.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—LC50 values were reported.
	Metric 18:	Consistency of Outcome Assessment	Low	Little detail was provided on the outcome assessment protocol for the acute toxicity.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions—it was not reported if the organisms were acclimated.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	LC50 values and their 95% confidence issues were determined using Probit.
	Metric 22:	Reporting of Data	High	The biological control response and the exposure response were reported in the supplementary data. LC50 values were reported in Table 1.
	Metric 23:	Explanation of Unexpected Outcomes	High	The study authors did not report any unexpected outcomes. 95% confidence intervals were reported in Table 1.
Additional Comments:	This portion of the evaluation was on the acute toxicity of DBP on <i>D. magna</i> adults. Mortality was selected as the outcome of interest because LC50 values were reported.The supplemental data was not included in the Distiller download, but was provided via a link in the paper.			
Overall Quality Determination			High	

Study Citation:	Shen, C., Wei, J., Wang, T., Wang, Y. (2019). Acute toxicity and responses of antioxidant systems to dibutyl phthalate in neonate and adult <i>Daphnia magna</i> . PeerJ 7(3):e6584.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5433053			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The DBP was identified by CASRN.	
	Metric 2: Test Substance Source	High	The source of the DBP was reported to be Sinopharm Chemical Reagent Co. Ltd, and verified by GC-MS.	
	Metric 3: Test Substance Purity	Low	The purity/grade of the DBP was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of a negative control without DBP and a solvent control.	
	Metric 5: Negative Control Response	High	The negative control response and the solvent control response were both reported in the supplementary data and were adequate.	
	Metric 6: Randomized Allocation	Low	It was not reported how the Daphnids were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	Limited details were provided on the preparation of the test substance. The test system was a static system.	
	Metric 8: Consistency of Exposure Administration	High	All tests were for 24 or 48h in 100mL of test solution with a 12L:12D photoperiod at 22C. Tests were conducted in dechlorinated water.	
	Metric 9: Measurement of Test Substance Concentration	High	Test concentrations were measured using GC-MS.	
	Metric 10: Exposure Duration and Frequency	High	The exposure duration was reported to be 48h with LC50 values calculated at 24 and 48h.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Medium	There were 5 exposure levels reported, but the highest concentrations did not have complete mortality.	
	Metric 12: Testing at or Below Solubility Limit	High	Study authors reported the use of a vehicle solvent and had a solvent control with an appropriate amount of solvent. The solvent control response was adequate for the outcome of interest.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were reported to be from the Key Laboratory of Hydrobiology at Dalian Ocean University. The age of the <i>Daphnia</i> was appropriate for the study.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the <i>Daphnia</i> were acclimated to test conditions.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 10 <i>Daphnia</i> per test chamber, and each concentrations was tested with 5 replicates.	
Domain 5: Outcome Assessment				
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Study Citation:	Shen, C., Wei, J., Wang, T., Wang, Y. (2019). Acute toxicity and responses of antioxidant systems to dibutyl phthalate in neonate and adult <i>Daphnia magna</i> . PeerJ 7(3):e6584.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5433053			
Domain		Metric	Rating	Comments
	Metric 16:	Adequacy of Test Conditions	High	The tests were conducted with dechlorinated water at 22C with a 12L:12D photoperiod. Prior to the study, the Daphnids were fed <i>Scenedesmus</i> sp. They were not fed for the duration of the test.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—LC50 values were reported.
	Metric 18:	Consistency of Outcome Assessment	Low	Little detail was provided on the outcome assessment protocol for the acute toxicity.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions—it was not reported if the organisms were acclimated.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	LC50 values and their 95% confidence issues were determined using Probit.
	Metric 22:	Reporting of Data	High	The biological control response and the exposure response were reported in the supplementary data. LC50 values were reported in Table 1.
	Metric 23:	Explanation of Unexpected Outcomes	High	The study authors did not report any unexpected outcomes. 95% confidence intervals were reported in Table 1.
Additional Comments:	This portion of the evaluation was on the acute toxicity of DBP on <i>D. magna</i> neonates. Mortality was selected as the outcome of interest because LC50 values were reported.The supplemental data was not included in the Distiller download, but was provided via a link in the paper.			
Overall Quality Determination			High	

Study Citation:	Sugatt, R. H., O'Grady, D. P., Banerjee, S. (1984). Toxicity of organic mixtures saturated in water to <i>Daphnia magna</i> : Effect of compositional changes. Chemosphere 13(1):11-18.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2134926			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The DBP was identified by name only.
	Metric 2:	Test Substance Source	Low	The source of the DBP was not reported, nor was it reported to be analytically verified.
	Metric 3:	Test Substance Purity	Low	The purity was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of a concurrent negative control.
	Metric 5:	Negative Control Response	Low	The negative control response was not reported. Only LT50 values were reported.
	Metric 6:	Randomized Allocation	Low	It was not reported how the <i>D. magna</i> were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	Saturated aqueous solutions were prepared by adding an excess of each chemical to test tubes containing 20mL of water. Tubes were then capped and shaken vigorously at room temperature for at least 10 days. The saturated solutions were then withdrawn and centrifuged before being transferred to glass culture dishes. Glass covers were used to minimize loss of test substance, though this may not have been adequate to stop chemical loss and evaporation.
	Metric 8:	Consistency of Exposure Administration	Medium	The exposure appeared to be conducted consistently. However, loss of test substance could possibly reduce consistency.
	Metric 9:	Measurement of Test Substance Concentration	Low	It was not reported if the test substance was measured at any point in the study.
	Metric 10:	Exposure Duration and Frequency	High	The test duration was adequate to determine an LT50 value.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	There was only one exposure concentration for this study as the goal was to compare a singular saturated exposure to different mixture ratios.
	Metric 12:	Testing at or Below Solubility Limit	High	The singular chemical exposures in this study were conducted at saturation.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the <i>D. magna</i> was not reported. All but one of the studies used <24h old organisms. The study that did not use juveniles, used adults in order to compare toxicity between age groups.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the test organisms were acclimated prior to the start of the exposures.
	Metric 15:	Number of Organisms and Replicates per Group	Low	It was reported that there were 10-15 <i>D. magna</i> per test chamber. The number of replicates was not reported.

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Study Citation:	Sugatt, R. H., O'Grady, D. P., Banerjee, S. (1984). Toxicity of organic mixtures saturated in water to <i>Daphnia magna</i> : Effect of compositional changes. Chemosphere 13(1):11-18.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Juvenile
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	2134926

Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
Metric 16:	Adequacy of Test Conditions	Low	Dechlorinated municipal water was used in this study. The water quality characteristic were reported in the paper. The toxicity tests were conducted without feeding or aeration with a 16h light:8h dark photoperiod at room temperature. The culture conditions or holding conditions prior to the start of the study were not reported.
Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—mortality in terms of LT50 values.
Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. The number of dead organisms was determined by an absence of movement of any appendages as observed under 2X magnification.
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	Low	It was not reported if the test organisms were acclimated or what the culture/pretreatment conditions were.
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	High	LT50 values and 95% confidence intervals were determined using the method of Litchfield.
Metric 22:	Reporting of Data	Low	Only LT50 values were reported (Table 1). Control responses were not reported.
Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.
Additional Comments: This evaluation was for the mortality outcome reported DBP. Mortality outcomes were presented as LT50 values.			

Overall Quality Determination**Medium**

Study Citation:	Wei, J., Shen, Q., Ban, Y., Wang, Y., Shen, C., Wang, T., Zhao, W., Xie, X. (2018). Characterization of Acute and Chronic Toxicity of DBP to <i>Daphnia magna</i> . Bulletin of Environmental Contamination and Toxicology 101(2):214-221.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	4829279			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The chemical was identified by name only.
	Metric 2:	Test Substance Source	High	The test substance was obtained from S&T Ltd, Tianjin, China. The test substance was identified by GC-MS.
	Metric 3:	Test Substance Purity	High	
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using appropriate concurrent negative control groups.
	Metric 5:	Negative Control Response	High	Survival in experimental controls and vehicle controls was 100%.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations. Exposure was conducted in 100mL glass beakers.
	Metric 8:	Consistency of Exposure Administration	Medium	Nothing in the study suggested that exposures were not administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The test organisms were adequately described, but culture origin was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects (10 organisms per treatment per beaker and replicated five times).
Domain 5: Outcome Assessment				
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Study Citation:	Wei, J., Shen, Q., Ban, Y., Wang, Y., Shen, C., Wang, T., Zhao, W., Xie, X. (2018). Characterization of Acute and Chronic Toxicity of DBP to <i>Daphnia magna</i> . Bulletin of Environmental Contamination and Toxicology 101(2):214-221.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	4829279			
Domain	Metric	Rating	Comments	
	Metric 16:	Adequacy of Test Conditions	Medium	Environmental conditions of the test system were most likely conducive to maintenance of organism health but actual measured condition values for control and exposed vessels was not reported.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	High	There were no differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described.
	Metric 22:	Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group, but results were described in the text and tables.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments:	None			
Overall Quality Determination		High		

Study Citation:	Kashian, D. R., Dodson, S. I. (2002). Effects of common-use pesticides on developmental and reproductive processes in <i>Daphnia</i> . Toxicology and Industrial Health 18(5):225-235.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332818; Linked HERO ID(s): 1332818, 5489093			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The test substance was identified by name only.	
	Metric 2: Test Substance Source	Low	The source of the DBP was reported to be ChemSevice in West Chester, PA, USA, but it was not reported if it was analytically verified.	
	Metric 3: Test Substance Purity	Low	The purity was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of a concurrent negative control. Acetone was used in the control solvent.	
	Metric 5: Negative Control Response	Low	The biological response of the negative control was not reported.	
	Metric 6: Randomized Allocation	Medium	It was reported that the <i>D. magna</i> were randomly assigned to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	It was not reported how the test media was prepared. Test system was static renewal with media being refreshed every 3 days. No information was provided on the type of experimental vessel (glass or plastic) used.	
	Metric 8: Consistency of Exposure Administration	Medium	Exposures were administered for 6 days in 30mL of test solution with one adult organisms per test chamber. There were 40 replicates for each treatment.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if the test concentrations were measured at any point in the study.	
	Metric 10: Exposure Duration and Frequency	Low	The study duration was reported to be for 6 days. This is longer than is typical for acute studies, and much shorter than chronic studies.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Low	A range of test concentrations used (10-100ug/L) for DBP was provided, but actual test concentrations were not reported.	
	Metric 12: Testing at or Below Solubility Limit	High	An appropriate solvent and a solvent control were used in this study.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The study reported that adult <i>D. magna</i> were used in the study. Adult <i>D. magna</i> would not be as sensitive as neonates. This could affect the outcomes of the study.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the organisms were acclimated before the study.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There was one organism per test chamber and 40 replicates per treatment.	
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Study Citation:	Kashian, D. R., Dodson, S. I. (2002). Effects of common-use pesticides on developmental and reproductive processes in <i>Daphnia</i> . <i>Toxicology and Industrial Health</i> 18(5):225-235.		
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Adult		
Health Outcome:	Mortality		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	1332818; Linked HERO ID(s): 1332818, 5489093		
Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
Metric 16:	Adequacy of Test Conditions	Low	Organisms were kept at 21C with a 6L:18D photoperiod. This photoperiod is not typical of parthenogenic reproduction, though authors reported they normally see about 25% male production. It was reported they were fed, but the feeding regimen was not reported.
Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcomes of interest-survivorship.
Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment was limited and in cases not reported.
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	High	The G-test and the Student's T-test were used for statistical analysis depending on which outcome was being analyzed.
Metric 22:	Reporting of Data	Low	Data for each treatment level and control response were not reported. Only some of the data was reported.
Metric 23:	Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability.
Additional Comments:	This portion of the evaluation was on the decreased survivorship in <i>D. magna</i> after exposure to DBP. Mortality was selected as the outcome of interest.		
Overall Quality Determination		Low	

Study Citation:	Kashian, D. R., Dodson, S. I. (2002). Effects of common-use pesticides on developmental and reproductive processes in <i>Daphnia</i> . Toxicology and Industrial Health 18(5):225-235.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Adult			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332818; Linked HERO ID(s): 1332818, 5489093			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The test substance was identified by name only.	
	Metric 2: Test Substance Source	Low	The source of the DBP was reported to be ChemSevice in West Chester, PA, USA, but it was not reported if it was analytically verified.	
	Metric 3: Test Substance Purity	Low	The purity was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of a concurrent negative control. Acetone was used in the control solvent.	
	Metric 5: Negative Control Response	Low	The biological response of the negative control was not reported.	
	Metric 6: Randomized Allocation	Medium	It was reported that the <i>D. magna</i> were randomly assigned to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	It was not reported how the test media was prepared. Test system was static renewal with media being refreshed every three days. No information was provided on the type of experimental vessel (glass or plastic) used.	
	Metric 8: Consistency of Exposure Administration	Medium	Exposures were administered for six days in 30mL of test solution with one adult organism per test chamber. There were 40 replicates for each treatment.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if the test concentrations were measured at any point in the study.	
	Metric 10: Exposure Duration and Frequency	Low	The study duration was reported to be for six days. This is longer than is typical for acute studies, and much shorter than chronic studies.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Low	A range of test concentrations used (10-100ug/L) for DBP was provided, but actual test concentrations were not reported.	
	Metric 12: Testing at or Below Solubility Limit	High	An appropriate solvent and a solvent control were used in this study.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The study reported that adult <i>D. magna</i> were used in the study. Adult <i>D. magna</i> would not be as sensitive as neonates. This could affect the outcomes of the study.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the organisms were acclimated before the study.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There was one organism per test chamber and 40 replicates per treatment.	
Domain 5: Outcome Assessment				
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Study Citation:	Kashian, D. R., Dodson, S. I. (2002). Effects of common-use pesticides on developmental and reproductive processes in <i>Daphnia</i> . Toxicology and Industrial Health 18(5):225-235.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Adult			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332818; Linked HERO ID(s): 1332818, 5489093			
Domain	Metric	Rating	Comments	
	Metric 16: Adequacy of Test Conditions	Low	Organisms were kept at 21C with a 6L:18D photoperiod. This photoperiod is not typical of parthenogenic reproduction, though authors reported they normally see about 25% male production. It was reported they were fed, but the feeding regimen was not reported.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcomes of interest–fecundity.	
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited and in some cases not reported.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	The G-test and the Student’s T-test were used for statistical analysis depending on which outcome was being analyzed.	
	Metric 22: Reporting of Data	Low	Data for each treatment level and control response were not reported. Only some of the data was reported.	
	Metric 23: Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability.	
Additional Comments:	This portion of the evaluation was on the decreased fecundity in <i>D. magna</i> after exposure to DBP. Reproduction was selected as the outcome of interest.			
Overall Quality Determination		Low		

Study Citation:	Kashian, D. R., Dodson, S. I. (2002). Effects of common-use pesticides on developmental and reproductive processes in <i>Daphnia</i> . <i>Toxicology and Industrial Health</i> 18(5):225-235.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Adult			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332818; Linked HERO ID(s): 1332818, 5489093			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The test substance was identified by name only.
	Metric 2:	Test Substance Source	Low	The source of the DBP was reported to be ChemSevice in West Chester, PA, USA, but it was not reported if it was analytically verified.
	Metric 3:	Test Substance Purity	Low	The purity was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of a concurrent negative control. Acetone was used in the control solvent.
	Metric 5:	Negative Control Response	Low	The biological response of the negative control was not reported.
	Metric 6:	Randomized Allocation	Medium	It was reported that the <i>D. magna</i> were randomly assigned to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	It was not reported how the test media was prepared. Test system was static renewal with media being refreshed every 3 days. No information was provided on the type of experimental vessel (glass or plastic) used.
	Metric 8:	Consistency of Exposure Administration	Low	Exposures were administered for 6 days in 30mL of test solution with one adult organisms per test chamber. There were 40 replicates for each treatment.
	Metric 9:	Measurement of Test Substance Concentration	Low	It was not reported if the test concentrations were measured at any point in the study.
	Metric 10:	Exposure Duration and Frequency	Medium	The study duration was reported to be for 6 days. This is longer than is typical for acute studies, and much shorter than chronic studies.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	A range of test concentrations used (10-100ug/L) for DBP was provided, but actual test concentrations were not reported.
	Metric 12:	Testing at or Below Solubility Limit	High	An appropriate solvent and a solvent control were used in this study.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The study reported that adult <i>D. magna</i> were used in the study. Adult <i>D. magna</i> would not be as sensitive as neonates. This could affect the outcomes of the study.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the organisms were acclimated before the study.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	There was one organism per test chamber and 40 replicates per treatment.
Domain 5: Outcome Assessment				
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Study Citation:	Kashian, D. R., Dodson, S. I. (2002). Effects of common-use pesticides on developmental and reproductive processes in <i>Daphnia</i> . Toxicology and Industrial Health 18(5):225-235.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Adult			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332818; Linked HERO ID(s): 1332818, 5489093			
Domain	Metric	Rating	Comments	
	Metric 16:	Adequacy of Test Conditions	Low	Organisms were kept at 21C with a 6L:18D photoperiod. This photoperiod is not typical of parthenogenic reproduction, though authors reported they normally see about 25% male production. It was reported they were fed, but the feeding regimen was not reported.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcomes of interest—male production and decreased growth.
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment was limited and in cases not reported.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	The G-test and the Student’s T-test were used for statistical analysis depending on which outcome was being analyzed.
	Metric 22:	Reporting of Data	Low	Data for each treatment level and control response were not reported. Only some of the data was reported.
	Metric 23:	Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability.
Additional Comments:	This portion of the evaluation was on the decreased growth in terms of deformities and adult size, as well as the increased number of males in <i>D. magna</i> adults exposed to DBP.			
Overall Quality Determination		Low		

Study Citation:	Defoe, D. L., Holcombe, G. W., Hammermeister, D. E., Biesinger, K. E. (1990). Solubility and toxicity of eight phthalate esters to four aquatic organisms. Environmental Toxicology and Chemistry 9(5):623-636.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5774391			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The full chemical name is provided in addition to the acronym but no specific CAS number or additional information is provided.
	Metric 2:	Test Substance Source	Low	Test substance identity was not analytically verified.
	Metric 3:	Test Substance Purity	Medium	One test substance (DBoP, di-n-butyl ortho) was commercially purchased and of high purity (>99%). The chemical was also synthesized for tests, but the purity was not analytically verified. Iso and tere forms were also synthesized.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Control groups (no chemical) were evaluated in concert with treatment groups. No solvents were used to necessitate a solvent control.
	Metric 5:	Negative Control Response	High	There were no concerns or anomalies associated with control groups.
	Metric 6:	Randomized Allocation	Medium	Organisms were randomly distributed among treatments and randomly subsampled for measurements.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	Sufficient detail is provided regarding the experimental design as well as development of stock solutions. Water physiochemical characteristics were regularly measured and reported. DBP concentrations were measured and reported.
	Metric 8:	Consistency of Exposure	High	Details of exposure are provided and are consistent among study groups.
	Metric 9:	Administration Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate methods.
	Metric 10:	Exposure Duration and Frequency	High	Exposure was appropriate and followed standard ASTM protocols.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The range of concentrations allowed for calculation of an LC50.
	Metric 12:	Testing at or Below Solubility Limit	Medium	Some test concentrations were at or above solubility though verification of concentrations was provided and care taken to ensure minimal degradation or loss of test substance during experiments.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Test organisms were obtained from a reliable source and test organism details were provided.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	Minimal details on pretreatment were provided.
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Study Citation:	Defoe, D. L., Holcombe, G. W., Hammermeister, D. E., Biesinger, K. E. (1990). Solubility and toxicity of eight phthalate esters to four aquatic organisms. Environmental Toxicology and Chemistry 9(5):623-636.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5774391			
Domain	Metric	Rating	Comments	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The number of test organisms used follows standard ASTM protocol.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Study conditions (physiochemical characteristics, feeding details) are well documented.	
	Metric 17: Outcome Assessment Methodology	High	The outcome of interest (LC50) was appropriate.	
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across groups.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no variations or inconsistencies reported across study groups and environmental conditions are provided.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There is no information to suggest differences among groups.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical methods were appropriate.	
	Metric 22: Reporting of Data	High	Results for all treatments and outcomes were reported.	
	Metric 23: Explanation of Unexpected Outcomes	High	Authors reported no unexpected outcomes and reported variance. Author provided discussion on different results measured in other papers.	
Additional Comments:	21d chronic exposure of Daphnia to DBP had a significant effect on survival, and effects were dependent on form.			

Overall Quality Determination**High**

Study Citation:	Defoe, D. L., Holcombe, G. W., Hammermeister, D. E., Biesinger, K. E. (1990). Solubility and toxicity of eight phthalate esters to four aquatic organisms. Environmental Toxicology and Chemistry 9(5):623-636.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Adult			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5774391			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The full chemical name is provided in addition to the acronym but no specific CAS number or additional information is provided.
	Metric 2:	Test Substance Source	Low	Test substance identity was not analytically verified.
	Metric 3:	Test Substance Purity	Medium	One test substance (DBoP, di-n-butyl ortho) was commercially purchased and of high purity (>99%). Chemical was also synthesized for tests and purity not analytically verified. Iso and tere forms also synthesized.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Control groups (no chemical) were evaluated in concert with treatment groups. No solvents were used to necessitate a solvent control.
	Metric 5:	Negative Control Response	High	There were no concerns or anomalies associated with control groups.
	Metric 6:	Randomized Allocation	Medium	Organisms were randomly distributed among treatments and randomly subsampled for measurements.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	Sufficient detail is provided regarding the experimental design as well as development of stock solutions. Water physiochemical characteristics were regularly measured and reported. DBP concentrations were measured and reported.
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure are provided and are consistent among study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate methods.
	Metric 10:	Exposure Duration and Frequency	High	Exposure was appropriate and followed standard ASTM protocols.
	Metric 11:	Number of Exposure Groups/Spacing of Exposure Levels	High	Range of concentrations allowed for calculation of reproduction.
	Metric 12:	Testing at or Below Solubility Limit	Medium	Some test concentrations were at or above solubility though verification of concentrations was provided and care taken to ensure minimal degradation or loss of test substance during experiments.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Test organism was obtained from a reliable source and test organism details were provided.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	Minimal details on pretreatment were provided.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Test organisms follow standard ASTM protocol and are reported.
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Study Citation:	Defoe, D. L., Holcombe, G. W., Hammermeister, D. E., Biesinger, K. E. (1990). Solubility and toxicity of eight phthalate esters to four aquatic organisms. Environmental Toxicology and Chemistry 9(5):623-636.		
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Adult		
Health Outcome:	Reproductive/Teratogenic		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	5774391		
Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Study conditions (physiochemical characteristics, feeding details) are well documented.
	Metric 17: Outcome Assessment Methodology	High	The outcome of interest (reproduction) was appropriate.
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no variations or inconsistencies reported across study groups and environmental conditions are provided.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There is no information to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were appropriate.
	Metric 22: Reporting of Data	High	Results for all treatments and outcomes were reported.
	Metric 23: Explanation of Unexpected Outcomes	High	Authors reported no unexpected outcomes and reported variance. Author provided discussion on different results measured in other papers.
Additional Comments: 21d chronic exposure of Daphnia to DBP had a significant effect on reproduction.			
Overall Quality Determination		High	

Study Citation:	Huang, G. L., Sun, H. W., Song, Z. H. (1999). Interactions between dibutyl phthalate and aquatic organisms. Bulletin of Environmental Contamination and Toxicology 63(6):759-765.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Larvae			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5551982			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The chemical was identified by name only.
	Metric 2:	Test Substance Source	Low	The source was not reported.
	Metric 3:	Test Substance Purity	Low	The purity and grade of the test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	A concurrent negative control group was reported.
	Metric 5:	Negative Control Response	High	A concurrent negative control group response was reported and suitable.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.
	Metric 8:	Consistency of Exposure Administration	Low	Only general methods of exposure administration were reported, so it was difficult to determine if the assessment was carried out consistently.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration was suitable.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable.
	Metric 12:	Testing at or Below Solubility Limit	Medium	A subset of the exposure concentrations exceeded or nearly exceeded the water solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups.
	Metric 15:	Number of Organisms and Replicates per Group	Low	There were three replicates with one daphnid each.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if they were adequate.
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Study Citation:	Huang, G. L., Sun, H. W., Song, Z. H. (1999). Interactions between dibutyl phthalate and aquatic organisms. Bulletin of Environmental Contamination and Toxicology 63(6):759-765.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Larvae			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5551982			
Domain	Metric		Rating	Comments
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology was reported and adequate.
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were shown for each treatment and control group.
	Metric 23:	Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability.
Additional Comments: None				

Overall Quality Determination**Low**

Study Citation:	Huang, G. L., Sun, H. W., Song, Z. H. (1999). Interactions between dibutyl phthalate and aquatic organisms. Bulletin of Environmental Contamination and Toxicology 63(6):759-765.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Larvae			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5551982			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The source was not reported.	
	Metric 3: Test Substance Purity	Low	The purity and grade of the test substance were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	A concurrent negative control group was reported.	
	Metric 5: Negative Control Response	High	A concurrent negative control group response was reported and suitable.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.	
	Metric 8: Consistency of Exposure Administration	Low	Only general methods of exposure administration were reported, so it was difficult to determine if the assessment was carried out consistently.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The exposure duration was suitable.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable.	
	Metric 12: Testing at or Below Solubility Limit	Medium	A subset of the exposure concentrations exceeded or nearly exceeded the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups.	
	Metric 15: Number of Organisms and Replicates per Group	Low	There were three replicates with one daphnid each.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if they were adequate.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology was reported and adequate.	
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Study Citation:	Huang, G. L., Sun, H. W., Song, Z. H. (1999). Interactions between dibutyl phthalate and aquatic organisms. Bulletin of Environmental Contamination and Toxicology 63(6):759-765.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Larvae			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5551982			
Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were shown for each treatment and control group.
	Metric 23:	Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability.
Additional Comments: None				

Overall Quality Determination**Low**

Study Citation:	Jr, Mayer, F., Sanders, H. O., Walsh, D. F. (1973). Toxicity, residue dynamics, and reproductive effects of phthalate esters in aquatic invertebrates. Environmental Research 6(1):84-90.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Juvenile			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1334646			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The chemical was identified by name.
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Uninformative	A concurrent negative control group was not included or reported.
	Metric 5:	Negative Control Response	Low	The biological response of the negative control groups was not reported.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.
	Metric 8:	Consistency of Exposure Administration	Low	Few details of exposure administration were reported, but exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	Only one treatment was reported.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test animals was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of test replicates was not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.
	Metric 17:	Outcome Assessment Methodology	Medium	The outcome assessment methodology was not clearly reported.

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Study Citation:	Jr, Mayer, F., Sanders, H. O., Walsh, D. F. (1973). Toxicity, residue dynamics, and reproductive effects of phthalate esters in aquatic invertebrates. Environmental Research 6(1):84-90.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Juvenile			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1334646			
Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	Medium	Few details regarding the execution of the study protocol for outcome assessment were provided.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was not performed.
	Metric 22:	Reporting of Data	Medium	Data for exposure-related findings were presented for the sampling period.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments: None				

Overall Quality Determination**Uninformative**

Study Citation:	Mccarthy, J. F., Whitmore, D. K. (1985). Chronic toxicity of di-n-butyl and di-n-octyl phthalate to daphnia-magna and the fathead minnow. Environmental Toxicology and Chemistry 4(2):167-179.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Juvenile			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1336024			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity was reported as 99.5 %.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	Medium	The biological response of the negative control group was reported and reasonable for assessed outcomes although the non-solvent control had poor survival and fewer young.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.	
	Metric 8: Consistency of Exposure Administration	Medium	Daily renewals occurred, but few details were provided.	
	Metric 9: Measurement of Test Substance Concentration	Medium	Exposure concentrations were measured, but are considerably lower than nominal concentrations.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source. "Test animals for both the acute immobilization tests (range finding test) and the reproduction test were collected when the animals were less than 24 h old."	
	Metric 14: Acclimatization and Pretreatment Conditions	High	Pretreatment conditions were the same for control and exposed organisms, but details were limited.	
	Metric 15: Number of Organisms and Replicates per Group	Low	There were 40 daphnids per treatment, but no replicates used.	
Domain 5: Outcome Assessment				
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Study Citation:	Mccarthy, J. F., Whitmore, D. K. (1985). Chronic toxicity of di-n-butyl and di-n-octyl phthalate to daphnia-magna and the fathead minnow. Environmental Toxicology and Chemistry 4(2):167-179.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Juvenile			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1336024			
Domain	Metric	Rating	Comments	
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	Results for this criterion should be related to the solvent control. There was statistically-significant mortality in the negative control group that may impact results for reproductive effects. The study authors attempted to salvage the study by relating everything to surviving <i>D. magna</i> . However, this introduces further uncertainty regarding the actual effects of DBP on <i>D. magna</i> . "Survival of <i>D. magna</i> exposed to DBP exceeded 80% in all concentrations except 1.8 and 3.2 mg/L and in the carrier-free control. The reason for the poor survival (and poor reproduction) of the control group is not clear; the same problem was evident in the DOP experiment (see below). By the end of the experiment (day 16), 70% of the <i>D. magna</i> were alive at 1.8 mg/L and 18% were alive at 3.2 mg/L DBP (Fig. 1 and Table 3)."
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23:	Explanation of Unexpected Outcomes	Low	There was unexpectedly low survival in the clean control.
Additional Comments:	This form is for <i>D. magna</i> reproductive effects - total broods and days to primiparous instar. There were serious concerns regarding the survivability of negative control organisms in this study as well as number of young per adult. The study authors attempted to bypass the survivability issue by relating results to number of young per adult, but with such variability in controls it introduces uncertainty. The solvent control should be used as an alternative.			

Overall Quality Determination**Medium**

Study Citation:	Mccarthy, J. F., Whitmore, D. K. (1985). Chronic toxicity of di-n-butyl and di-n-octyl phthalate to daphnia-magna and the fathead minnow. Environmental Toxicology and Chemistry 4(2):167-179.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Juvenile			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1336024			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity was reported as 99.5 %.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	Medium	The biological response of the negative control group was reported and reasonable for assessed outcomes, although the non-solvent control had poor survival and fewer young.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations	
	Metric 8: Consistency of Exposure Administration	Medium	Daily renewals but few details were provided	
	Metric 9: Measurement of Test Substance Concentration	Medium	Exposure concentrations were measured but are considerably lower than nominal concentrations	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source. "Test animals for both the acute immobilization tests (range finding test) and the reproduction test were collected when the animals were less than 24 h old."	
	Metric 14: Acclimatization and Pretreatment Conditions	High	Pretreatment conditions were the same for control and exposed organisms, details were limited	
	Metric 15: Number of Organisms and Replicates per Group	Low	40 daphnids per treatment but no replicates	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health	

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Study Citation:	Mccarthy, J. F., Whitmore, D. K. (1985). Chronic toxicity of di-n-butyl and di-n-octyl phthalate to daphnia-magna and the fathead minnow. Environmental Toxicology and Chemistry 4(2):167-179.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Juvenile			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1336024			
Domain	Metric	Rating	Comments	
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest outcomes were assessed consistently across study groups
	Metric 18:	Consistency of Outcome Assessment	High	
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	Results for this criterion should be related to the solvent control. There was statistically-significant mortality in the negative control group that may impact results for reproductive effects. The study authors attempted to salvage the study by relating everything to surviving D. magna. However, this introduces further uncertainty regarding the actual effects of DBP on D. magna."Survival of D. magna exposed to DBP exceeded 80% in all concentrations except 1.8 and 3.2 mg/L and in the carrier-free control. The reason for the poor survival (and poor reproduction) of the control group is not clear; the same problem was evident in the DOP experiment (see below). By the end of the experiment (day 16), 70% of the D. magna were alive at 1.8 mg/L and 18% were alive at 3.2 mg/L DBP (Fig. 1 and Table 3)."
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	Low	Unexpectedly low survival in clean control
Additional Comments:	This form is to account for development of the Daphnids found in Table 4. There were serious concerns regarding the survivability of negative control organisms in this study as well as number of young per adult. The study authors attempted to bypass the survivability issue relate results to number of young per adult, but with such variability in controls introduces uncertainty. The solvent control should be used as an alternative.			

Overall Quality Determination**Medium**

Study Citation:	Mccarthy, J. F., Whitmore, D. K. (1985). Chronic toxicity of di-n-butyl and di-n-octyl phthalate to daphnia-magna and the fathead minnow. Environmental Toxicology and Chemistry 4(2):167-179.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1336024			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.5 %
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	Medium	The biological response of the negative control group was reported and reasonable for assessed outcomes although the non-solvent control had poor survival and fewer young
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Medium	Daily renewals but few details were provided
	Metric 9:	Measurement of Test Substance Concentration	Medium	Exposure concentrations were measured but are considerably lower than nominal concentrations
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source. "Test animals for both the acute immobilization tests (range finding test) and the reproduction test were collected when the animals were less than 24 h old."
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Pretreatment conditions were the same for control and exposed organisms, details were limited
	Metric 15:	Number of Organisms and Replicates per Group	Low	40 daphnids per treatment but no replicates
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health

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Study Citation:	Mccarthy, J. F., Whitmore, D. K. (1985). Chronic toxicity of di-n-butyl and di-n-octyl phthalate to daphnia-magna and the fathead minnow. Environmental Toxicology and Chemistry 4(2):167-179.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1336024			
Domain	Metric	Rating	Comments	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest outcomes were assessed consistently across study groups	
	Metric 18: Consistency of Outcome Assessment	High		
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	Results for this criterion should be related to the solvent control. There was statistically-significant mortality in the negative control group that may impact results for reproductive effects. The study authors attempted to salvage the study by relating everything to surviving D. magna. However, this introduces further uncertainty regarding the actual effects of DBP on D. magna. "Survival of D. magna exposed to DBP exceeded 80% in all concentrations except 1.8 and 3.2 mg/L and in the carrier-free control. The reason for the poor survival (and poor reproduction) of the control group is not clear; the same problem was evident in the DOP experiment (see below). By the end of the experiment (day 16), 70% of the D. magna were alive at 1.8 mg/L and 18% were alive at 3.2 mg/L DBP (Fig. 1 and Table 3)."	
	Metric 20: Outcomes Unrelated to Exposure	Medium		
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical methods were adequately described	
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group	
	Metric 23: Explanation of Unexpected Outcomes	Low	Unexpectedly low survival in clean control	
Additional Comments:	This form is to account for mortality in chronic Daphnia magna test with results found in Table 3. There were serious concerns regarding the survivability of negative control organisms in this study as well as number of young per adult. The study authors attempted to bypass the survivability issue relate results to number of young per adult, but with such variability in controls introduces uncertainty. The solvent control should be used as an alternative.			

Overall Quality Determination**Medium**

Study Citation:	Rhodes, J. E., Adams, W. J., Biddinger, G. R., Robillard, K. A., Gorsuch, J. W. (1995). Chronic toxicity of 14 phthalate esters to <i>Daphnia magna</i> and rainbow trout (<i>Oncorhynchus mykiss</i>). Environmental Toxicology and Chemistry 14(11):1967-1976.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	680120; Linked HERO ID(s): 1316195, 680120			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical identified by name
	Metric 2:	Test Substance Source	Low	The test substance identity was NOT analytically verified by the performing laboratory
	Metric 3:	Test Substance Purity	High	Purity reported as >95%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using a concurrent negative control
	Metric 5:	Negative Control Response	High	The biological responses of the negative control group was adequate
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were justified for a dose response by study authors
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Organism environmental conditions were conducive to maintenance of organism health
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome

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Study Citation:	Rhodes, J. E., Adams, W. J., Biddinger, G. R., Robillard, K. A., Gorsuch, J. W. (1995). Chronic toxicity of 14 phthalate esters to <i>Daphnia magna</i> and rainbow trout (<i>Oncorhynchus mykiss</i>). Environmental Toxicology and Chemistry 14(11):1967-1976.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Juvenile
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	680120; Linked HERO ID(s): 1316195, 680120

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
	Metric 20: Outcomes Unrelated to Exposure	High	There were no differences among groups that could influence the outcome assessment
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Statistical analysis was performed but not described adequately
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group and were adequate
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes
Additional Comments:	None		

Overall Quality Determination**High**

Study Citation:	Rhodes, J. E., Adams, W. J., Biddinger, G. R., Robillard, K. A., Gorsuch, J. W. (1995). Chronic toxicity of 14 phthalate esters to <i>Daphnia magna</i> and rainbow trout (<i>Oncorhynchus mykiss</i>). Environmental Toxicology and Chemistry 14(11):1967-1976.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Juvenile			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	680120; Linked HERO ID(s): 1316195, 680120			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The chemical was identified by name.	
	Metric 2: Test Substance Source	Low	The test substance identity was NOT analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Purity was reported as >95%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control.	
	Metric 5: Negative Control Response	High	The biological responses of the negative control group was adequate.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The experimental system and methods for preparation of test media were described in adequate detail.	
	Metric 8: Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were justified for a dose response by study authors.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The number of test organisms and replicates were reported and sufficient to characterize toxicological effects.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Organism housing and environmental conditions were conducive to maintenance of organism health.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome.	
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.	
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Study Citation:	Rhodes, J. E., Adams, W. J., Biddinger, G. R., Robillard, K. A., Gorsuch, J. W. (1995). Chronic toxicity of 14 phthalate esters to <i>Daphnia magna</i> and rainbow trout (<i>Oncorhynchus mykiss</i>). Environmental Toxicology and Chemistry 14(11):1967-1976.		
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Juvenile		
Health Outcome:	Reproductive/Teratogenic		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	680120; Linked HERO ID(s): 1316195, 680120		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	High	There were no differences among groups that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group and were adequate.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments: None			

Overall Quality Determination**High**

Study Citation:	Seyoum, A., Pradhan, A. (2019). Effect of phthalates on development, reproduction, fat metabolism and lifespan in <i>Daphnia magna</i> . Science of the Total Environment 654:969-977.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Juvenile			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5043468			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The chemical was identified by name and CAS#.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity was reported as >99%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control groups was reported.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.	
	Metric 8: Consistency of Exposure Administration	Medium	Reporting omissions are unlikely to have a substantial impact on results.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Low	Only two exposure groups were reported.	
	Metric 12: Testing at or Below Solubility Limit	High	The solvent concentration was appropriate.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The number of organisms and replicates were suitable.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Details of environmental conditions of the test system were not reported.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome.	
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Study Citation:	Seyoum, A., Pradhan, A. (2019). Effect of phthalates on development, reproduction, fat metabolism and lifespan in <i>Daphnia magna</i> . Science of the Total Environment 654:969-977.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Juvenile
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	5043468

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups.
	Metric 20: Outcomes Unrelated to Exposure	High	There were no differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: length

Overall Quality Determination**Medium**

Study Citation:	Wei, J., Shen, Q., Ban, Y., Wang, Y., Shen, C., Wang, T., Zhao, W., Xie, X. (2018). Characterization of Acute and Chronic Toxicity of DBP to <i>Daphnia magna</i> . Bulletin of Environmental Contamination and Toxicology 101(2):214-221.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Adult			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	4829279			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The chemical was identified by name only.
	Metric 2:	Test Substance Source	High	The test substance was obtained from S&T Ltd, Tianjin, China. The test substance was identified by GC-MS.
	Metric 3:	Test Substance Purity	High	
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using appropriate concurrent negative control groups.
	Metric 5:	Negative Control Response	High	Survival in experimental controls and vehicle controls was 100%.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations. The exposure was conducted with individual neonates in 20mL glass tubes.
	Metric 8:	Consistency of Exposure Administration	Medium	Nothing in the study suggested that exposures were not administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The test organisms were adequately described, but culture origin was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects (15 organisms per treatment, in individual 20mL glass tubes).
Domain 5: Outcome Assessment				
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Study Citation:	Wei, J., Shen, Q., Ban, Y., Wang, Y., Shen, C., Wang, T., Zhao, W., Xie, X. (2018). Characterization of Acute and Chronic Toxicity of DBP to <i>Daphnia magna</i> . Bulletin of Environmental Contamination and Toxicology 101(2):214-221.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Adult			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	4829279			
Domain	Metric	Rating	Comments	
	Metric 16:	Adequacy of Test Conditions	Medium	Environmental conditions of the test system were most likely conducive to maintenance of organism health but actual measured condition values for control and exposed vessels was not reported.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	High	There were no differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described.
	Metric 22:	Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group, but results were described in the text and tables.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments:	molting frequency, generational effects also reported			
Overall Quality Determination		High		

Study Citation:	Wei, J., Shen, Q., Ban, Y., Wang, Y., Shen, C., Wang, T., Zhao, W., Xie, X. (2018). Characterization of Acute and Chronic Toxicity of DBP to <i>Daphnia magna</i> . Bulletin of Environmental Contamination and Toxicology 101(2):214-221.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Adult			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	4829279			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	Chemical was identified by name only	
	Metric 2: Test Substance Source	High	The test substance was obtained from S&T Ltd, Tianjin, China. The test substance was identified by GC-MS.	
	Metric 3: Test Substance Purity	High	Chemical purity reported as >99%	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using appropriate concurrent negative control groups	
	Metric 5: Negative Control Response	High	survival in experimental controls and vehicle controls was 100%.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations. The exposure was conducted with individual neonates in 20mL glass tubes.	
	Metric 8: Consistency of Exposure Administration	Medium	Nothing in the study suggested that exposures were not administered consistently across study groups	
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response	
	Metric 12: Testing at or Below Solubility Limit	High	The solvent concentration was appropriate	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Medium	The test organisms were adequately described, culture origin was not reported	
	Metric 14: Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects (15 organisms per treatment, in individual 20mL glass tubes).	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Medium	Environmental conditions of test system were most likely conducive to maintenance of organism health but actual measured condition values for control and exposed vessels was not reported	

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Study Citation:	Wei, J., Shen, Q., Ban, Y., Wang, Y., Shen, C., Wang, T., Zhao, W., Xie, X. (2018). Characterization of Acute and Chronic Toxicity of DBP to <i>Daphnia magna</i> . Bulletin of Environmental Contamination and Toxicology 101(2):214-221.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Adult
Health Outcome:	Reproductive/Teratogenic
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	4829279

Domain	Metric	Rating	Comments
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18: Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20: Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described
	Metric 22: Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group, but results were described in the text and tables
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes
Additional Comments: generational effects also reported			

Overall Quality Determination**High**

Study Citation:	Bionomics,, Springborn (1984). Chronic toxicity of fourteen phthalate esters to <i>Daphnia magna</i> with cover letter dated 032585. :95.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Adult			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1316195; Linked HERO ID(s): 1316195, 680120			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The test substance was identified by name only (DBP), no other information was provided.	
	Metric 2: Test Substance Source	High	The test substance was from General Electric Company, Hudson Falls, New York	
	Metric 3: Test Substance Purity	Low	The purity was not provided	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	A negative control was used (not a solvent control)	
	Metric 5: Negative Control Response	Medium	Authors reported for DEHP that animals in the controls appeared to be trapped at the surface	
	Metric 6: Randomized Allocation	Medium	Organisms were allocated in an unbiased manner	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	Experimental system was a flow through system with an "enhanced mixing process" to add in the chemical, which is poorly soluble in water. A solvent was not used.	
	Metric 8: Consistency of Exposure Administration	High	Exposure was administered consistently across groups	
	Metric 9: Measurement of Test Substance Concentration	Low	Test concentrations were measured weekly, and there was significant decline in the concentrations of DBP over the course of the experiment.	
	Metric 10: Exposure Duration and Frequency	High	The exposure duration was appropriate - 21 days	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Medium	5 concentrations were tested. Actual concentrations were lower than nominal and declined over time despite the flow through design.	
	Metric 12: Testing at or Below Solubility Limit	Low	Some concentrations exceeded the water solubility limit. An Enhanced mixing technique was used to deliver the chemical in the flow through system, but the loss of the test substance over time indicates that the chemical was instable in water.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The source of the <i>Daphnia</i> was Springborn Bionomics	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	Acclimatization was not reported	
	Metric 15: Number of Organisms and Replicates per Group	Low	There is no description of how many replicates there were per treatments or how many animals there were per replicate. An old protocol is cited: "Protocol for conducting chronic toxicity tests with the water flea <i>Daphnia magna</i>)" developed at EG&G Bionomics {1982}.	
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Study Citation:	Bionomics., Springborn (1984). Chronic toxicity of fourteen phthalate esters to <i>Daphnia magna</i> with cover letter dated 032585. :95.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Adult			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1316195; Linked HERO ID(s): 1316195, 680120			
Domain	Metric	Rating	Comments	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions were recorded and were consistent.	
	Metric 17: Outcome Assessment Methodology	High	Reproduction was assessed as cumulative number of offspring.	
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across groups	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Medium	The water quality variables were comparable across treatments	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical tests were performed (not well described, authors cite an old method) but all data are presented in the tables for treatments and controls	
	Metric 22: Reporting of Data	High	All data are presented in the tables for treatments and controls	
	Metric 23: Explanation of Unexpected Outcomes	High	Outcomes were satisfactorily described	
Additional Comments:	None			
Overall Quality Determination		Medium		

Study Citation:	Bionomics,, Springborn (1984). Chronic toxicity of fourteen phthalate esters to <i>Daphnia magna</i> with cover letter dated 032585. :95.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1316195; Linked HERO ID(s): 1316195, 680120			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The test substance was identified by name only (DBP). No other information was given.	
	Metric 2: Test Substance Source	High	The test substance was from General Electric Company, Hudson Falls, New York.	
	Metric 3: Test Substance Purity	Low	The purity was not provided.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	A negative control was used (not a solvent control).	
	Metric 5: Negative Control Response	Medium	Authors reported for DEHP that animals in the controls appeared to be trapped at the surface.	
	Metric 6: Randomized Allocation	Medium	Organisms were allocated in an unbiased manner.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The experimental system was a flow through system with an "enhanced mixing process" to add in the chemical, which is poorly soluble in water. A solvent was not used.	
	Metric 8: Consistency of Exposure Administration	High	Exposure was administered consistently across groups.	
	Metric 9: Measurement of Test Substance Concentration	Low	Test concentrations were measured weekly, and there was significant decline in the concentrations of DBP over the course of the experiment.	
	Metric 10: Exposure Duration and Frequency	High	The exposure duration of 21 days was appropriate.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Five concentrations were tested. Actual concentrations were lower than nominal and declined over time despite the flow through design.	
	Metric 12: Testing at or Below Solubility Limit	Low	Some concentrations exceeded the water solubility limit. An Enhanced mixing technique was used to deliver the chemical in the flow through system, but the loss of the test substance over time indicates that the chemical was unstable in water.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The source of the <i>Daphnia</i> was Springborn Bionomics.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	Acclimatization was not reported.	
	Metric 15: Number of Organisms and Replicates per Group	Low	There is no description of how many replicates there were per treatments or how many animals there were per replicate. An old protocol is cited: "Protocol for conducting chronic toxicity tests with the water flea <i>Daphnia magna</i>)" developed at EG&G Bionomics {1982}.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions were recorded and were consistent.	
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Study Citation:	Bionomics,, Springborn (1984). Chronic toxicity of fourteen phthalate esters to <i>Daphnia magna</i> with cover letter dated 032585. :95.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Daphnia magna</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1316195; Linked HERO ID(s): 1316195, 680120			
Domain	Metric	Rating	Comments	
	Metric 17:	Outcome Assessment Methodology	High	Mortality was assessed weekly, and reproduction was assessed as cumulative number of offspring.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Medium	The water quality variables were comparable across treatments.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical tests were performed (not well described, authors cite an old method) but all data are presented in the tables for treatments and controls.
	Metric 22:	Reporting of Data	High	All data are presented in the tables for treatments and controls.
	Metric 23:	Explanation of Unexpected Outcomes	High	Outcomes were satisfactorily described.
Additional Comments:	None			
Overall Quality Determination		Medium		

Study Citation:	Jr, Mayer, F., Sanders, H. O., Walsh, D. F. (1973). Toxicity, residue dynamics, and reproductive effects of phthalate esters in aquatic invertebrates. Environmental Research 6(1):84-90.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Gammarus pseudolimnaeus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1334646			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The chemical was identified by name.
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the amount of details provided in the study being reviewed.
	Metric 5:	Negative Control Response	Low	The biological response of the negative control groups was not reported.
	Metric 6:	Randomized Allocation	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the amount of details provided in the study being reviewed.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	Concentrations of test substance were not measured during the study.
	Metric 8:	Consistency of Exposure Administration	Medium	Some details of exposure administration were reported and exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Uninformative	No information is provided on the number of exposure groups and spacing of exposure levels.
	Metric 12:	Testing at or Below Solubility Limit	Low	Reporting omissions prevented determination of whether exposure concentrations exceeded the water solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test animals was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of test replicates was not reported.
Domain 5: Outcome Assessment				
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Study Citation:	Jr, Mayer, F., Sanders, H. O., Walsh, D. F. (1973). Toxicity, residue dynamics, and reproductive effects of phthalate esters in aquatic invertebrates. Environmental Research 6(1):84-90.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Gammarus pseudolimnaeus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1334646			
Domain	Metric	Rating	Comments	
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported.
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22:	Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group.
	Metric 23:	Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability.
Additional Comments:	None			

Overall Quality Determination**Uninformative**

Study Citation:	Jr, Mayer, F., Sanders, H. O., Walsh, D. F. (1973). Toxicity, residue dynamics, and reproductive effects of phthalate esters in aquatic invertebrates. Environmental Research 6(1):84-90.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Gammarus pseudolimnaeus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1334646			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The chemical was identified by name.
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Uninformative	A concurrent negative control group was not included or reported.
	Metric 5:	Negative Control Response	Low	The biological response of the negative control groups was not reported.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.
	Metric 8:	Consistency of Exposure Administration	Low	Few details of exposure administration were reported and exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	Only one treatment was reported.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test animals was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of test replicates was not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.
	Metric 17:	Outcome Assessment Methodology	Medium	The outcome assessment methodology was not clearly reported.

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Study Citation:	Jr, Mayer, F., Sanders, H. O., Walsh, D. F. (1973). Toxicity, residue dynamics, and reproductive effects of phthalate esters in aquatic invertebrates. Environmental Research 6(1):84-90.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Gammarus pseudolimnaeus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1334646			
Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	Medium	Few details regarding the execution of the study protocol for outcome assessment were provided.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was not performed.
	Metric 22:	Reporting of Data	Medium	Data for exposure-related findings were presented for the sampling period.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments: None				

Overall Quality Determination**Uninformative**

Study Citation:	Thurén, A., Woin, P. (1991). Effects of phthalate esters on the locomotor activity of the freshwater amphipod <i>Gammarus pulex</i> . Bulletin of Environmental Contamination and Toxicology 46(1):159-166.			
Duration:	Overall Duration: > 21 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Gammarus pulex</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	732821			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Medium	The chemical was only identified as DBP, a phthalate ester.	
	Metric 2: Test Substance Source	Low	The source was not reported.	
	Metric 3: Test Substance Purity	Low	Purity and/or grade of the test substance were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control.	
	Metric 5: Negative Control Response	Medium	Authors only reported that mortality was the same for control and treated organisms.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations. There is some concern over using plexiglass tanks with phthalates.	
	Metric 8: Consistency of Exposure Administration	Medium	Reporting omissions regarding quality checks are likely to have an impact on results.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Low	Only two exposure levels were used. This is not sufficient to obtain a dose response relationship.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Medium	There are minor reservations about the source of test organisms.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.	
	Metric 15: Number of Organisms and Replicates per Group	Low	Replicates were not used or reported.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Organism environmental conditions were conducive to maintenance of health and biomass loading was appropriate.	
	Metric 17: Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported.	

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Study Citation:	Thurén, A., Woin, P. (1991). Effects of phthalate esters on the locomotor activity of the freshwater amphipod <i>Gammarus pulex</i> . Bulletin of Environmental Contamination and Toxicology 46(1):159-166.			
Duration:	Overall Duration: > 21 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Gammarus pulex</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	732821			
Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Uninformative	Statistical analysis was not conducted.
	Metric 22:	Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments: None				

Overall Quality Determination**Uninformative**

Study Citation:	Thurén, A., Woin, P. (1991). Effects of phthalate esters on the locomotor activity of the freshwater amphipod <i>Gammarus pulex</i> . Bulletin of Environmental Contamination and Toxicology 46(1):159-166.			
Duration:	Overall Duration: > 21 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Gammarus pulex</i> ; Adult			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	732821			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Medium	Chemical only identified as DBP, a phthalate ester	
	Metric 2: Test Substance Source	Low	The source was not reported	
	Metric 3: Test Substance Purity	Low	Purity or grade of test substance were not reported	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control	
	Metric 5: Negative Control Response	Low	The biological response of the negative control groups was not reported	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations, some concern over using plexiglass tanks with phthalates	
	Metric 8: Consistency of Exposure Administration	Medium	Reporting omissions regarding quality checks are likely to have an impact on results	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured	
	Metric 10: Exposure Duration and Frequency	Low	The duration of exposure was reported but steady state or time dependent results were not reported	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Low	Only two exposure levels were used. This is not sufficient to obtain a dose response relationship.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Medium	There are minor reservations about the source of test organisms	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms	
	Metric 15: Number of Organisms and Replicates per Group	Low	Replicates were not used or reported	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Organism environmental conditions were conducive to maintenance of health and biomass loading was appropriate	
	Metric 17: Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported and water concentrations were not used to assess accumulation	
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Study Citation:	Thurén, A., Woin, P. (1991). Effects of phthalate esters on the locomotor activity of the freshwater amphipod <i>Gammarus pulex</i> . Bulletin of Environmental Contamination and Toxicology 46(1):159-166.
Duration:	Overall Duration: > 21 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Gammarus pulex</i> ; Adult
Health Outcome:	ADME (biotransformation)
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	732821

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	Medium	Details regarding the execution of the study protocol for outcome assessment were not reported
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental condition
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Statistical analysis was not conducted, typical for BCF assessments
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes
Additional Comments: None			

Overall Quality Determination**Medium**

Study Citation:	Thurén, A., Woin, P. (1991). Effects of phthalate esters on the locomotor activity of the freshwater amphipod <i>Gammarus pulex</i> . Bulletin of Environmental Contamination and Toxicology 46(1):159-166.			
Duration:	Overall Duration: > 21 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Gammarus pulex</i> ; Adult			
Health Outcome:	Behavioral			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	732821			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Medium	Chemical only identified as DBP, a phthalate ester
	Metric 2:	Test Substance Source	Low	The source was not reported
	Metric 3:	Test Substance Purity	Low	Purity or grade of test substance were not reported
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control
	Metric 5:	Negative Control Response	High	The biological response of the negative control groups was reported
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations, some concern over using plexiglass tanks with phthalates
	Metric 8:	Consistency of Exposure Administration	Medium	
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	Low	The duration of exposure was reported but steady state or time dependent results were not reported
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	Only two exposure levels were used. This is not sufficient to obtain a dose response relationship.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	There are minor reservations about the source of test organisms
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Low	Replicates were not used or reported
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Organism environmental conditions were conducive to maintenance of health and biomass loading was appropriate
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported
	Metric 18:	Consistency of Outcome Assessment	Medium	Some details regarding the execution of the study protocol for outcome assessment were not reported

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Study Citation:	Thurén, A., Woin, P. (1991). Effects of phthalate esters on the locomotor activity of the freshwater amphipod <i>Gammarus pulex</i> . Bulletin of Environmental Contamination and Toxicology 46(1):159-166.
Duration:	Overall Duration: > 21 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Gammarus pulex</i> ; Adult
Health Outcome:	Behavioral
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	732821

Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental condition
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Statistical analysis was performed but not described adequately
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23: Explanation of Unexpected Outcomes	Medium	Highly variable results with no clear trends

Additional Comments: None

Overall Quality Determination**Medium**

Study Citation:	Jr, Mayer, F., Sanders, H. O., Walsh, D. F. (1973). Toxicity, residue dynamics, and reproductive effects of phthalate esters in aquatic invertebrates. Environmental Research 6(1):84-90.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Hexagenia bilineata</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1334646			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The chemical was identified by name.
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of the test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Uninformative	A concurrent negative control group was not included or reported.
	Metric 5:	Negative Control Response	Low	The biological response of the negative control groups was not reported.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.
	Metric 8:	Consistency of Exposure Administration	Low	Few details of exposure administration were reported, but exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	Only one treatment was reported.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test animals was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of test replicates was not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.
	Metric 17:	Outcome Assessment Methodology	Medium	The outcome assessment methodology was not clearly reported.

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Study Citation:	Jr, Mayer, F., Sanders, H. O., Walsh, D. F. (1973). Toxicity, residue dynamics, and reproductive effects of phthalate esters in aquatic invertebrates. Environmental Research 6(1):84-90.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Hexagenia bilineata</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1334646			
Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	Medium	Few details regarding the execution of the study protocol for the outcome assessment were provided.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was not performed.
	Metric 22:	Reporting of Data	Medium	Data for exposure-related findings were presented for the sampling period.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments: None				

Overall Quality Determination**Uninformative**

Study Citation:	Call, D. J., Cox, D. A., Geiger, D. L., Genisot, K. I., Markee, T. P., Brooke, L. T., Polkinghorne, C. N., Vandeventer, F. A., Gorsuch, J. W., Robillard, K. A., Parkerton, T. F., Reiley, M. C., Ankley, G. T., Mount, D. R. (2001). An assessment of the toxicity of phthalate esters to freshwater benthos. 2. Sediment exposures. Environmental Toxicology and Chemistry 20(8):1805-1815.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Sediment; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Hyaella azteca</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	679311			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
Metric 1:	Test Substance Identity	High	Chemical was identified as a "single isomer" and the identify, including CASRN referenced in an outside paper, Call et al 2001.	
Metric 2:	Test Substance Source	High	Source of chemical was defined in Call et al 2001 as AldrichChemical (Milwaukee, WI, USA).	
Metric 3:	Test Substance Purity	High	Purity was identified as >99% in Call et al 2001.	
Domain 2: Test Design				
Metric 4:	Negative Controls	High	Negative controls (sediment and silica sand) were used in this experiment.	
Metric 5:	Negative Control Response	High	Biological response of control groups was appropriate as shown in Table 4.	
Metric 6:	Randomized Allocation	Low	Random allocation not reported.	
Domain 3: Exposure Characterization				
Metric 7:	Experimental System/Test Media Preparation	High	Methods of sediment collection and preparation (including chemical addition) and addition of sediment to test beakers were described in detail.	
Metric 8:	Consistency of Exposure Administration	High	Exposure consistency reported and consistent among different DBP treatments and controls.	
Metric 9:	Measurement of Test Substance Concentration	High	Concentrations measured using HPLC as described in methods and cited reference (Call et al 2001).	
Metric 10:	Exposure Duration and Frequency	High	Duration (10 day exposure) was appropriate for experimental design.	
Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Exposure groups were acceptable and spanned 5 concentrations per test species in addition to control; nominal doses unclear however measured doses reported in sediment and pore water.	
Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure via sediment.	
Domain 4: Test Organism				
Metric 13:	Test Organism Characteristics	Low	Source of test organism not reported.	
Metric 14:	Acclimatization and Pretreatment Conditions	Low	Acclimation of test organisms prior to exposure not reported.	
Metric 15:	Number of Organisms and Replicates per Group	Medium	Tests with DBP utilized 3 replicates of five different concentrations with 10 organisms per beaker and three sediment control replicates with 10 test organisms per beaker and three silica sand control replicates with 10 test organisms per beaker.	

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Study Citation:	Call, D. J., Cox, D. A., Geiger, D. L., Genisot, K. I., Markee, T. P., Brooke, L. T., Polkinghorne, C. N., Vandeventer, F. A., Gorsuch, J. W., Robillard, K. A., Parkerton, T. F., Reiley, M. C., Ankley, G. T., Mount, D. R. (2001). An assessment of the toxicity of phthalate esters to freshwater benthos. 2. Sediment exposures. Environmental Toxicology and Chemistry 20(8):1805-1815.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Sediment; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Hyalella azteca</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	679311

Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions including feeding, intermittent water renewal system, lighting, and temperature were reported. Authors recorded temperature, dissolved oxygen, and pH on days 0 and 10 and conductivity, hardness, alkalinity, and ammonia on days 1 and 9. Sediment TOC conditions described in Table 2.
	Metric 17: Outcome Assessment Methodology	Medium	Survivor count determined after the 10 day exposure but not reported as percent mortality.
	Metric 18: Consistency of Outcome Assessment	High	Outcome assessment conducted at conclusion of 10 day exposure.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	Metric 20: Outcomes Unrelated to Exposure	High	Details regarding test organism attrition and outcomes unrelated to exposure were reported for each study group and there were no differences among groups that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Survival data from toxicity tests were summarized using the trimmed Spearman-Kärber method. Dry weight data were analyzed by one-way analysis of variance and Dunnett's procedure using a SigmaStat Program.
	Metric 22: Reporting of Data	High	Survival data reported in Table 4 and LC50 values shown in Figure 1 and Table 6.
	Metric 23: Explanation of Unexpected Outcomes	Medium	Variability not reported but results suggest no excessive variability within replicates.
Additional Comments: None			

Overall Quality Determination**High**

Study Citation:	Call, D. J., Cox, D. A., Geiger, D. L., Genisot, K. I., Markee, T. P., Brooke, L. T., Polkinghorne, C. N., Vandeventer, F. A., Gorsuch, J. W., Robillard, K. A., Parkerton, T. F., Reiley, M. C., Ankley, G. T., Mount, D. R. (2001). An assessment of the toxicity of phthalate esters to freshwater benthos. 2. Sediment exposures. Environmental Toxicology and Chemistry 20(8):1805-1815.		
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Sediment; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Hyaella azteca</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Development/Growth		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	679311		
Domain	Metric	Rating	Comments
Domain 1: Test Substance			
	Metric 1: Test Substance Identity	High	The chemical was identified as a "single isomer" and the identify, including CASRN was referenced in an outside paper, Call et al 2001.
	Metric 2: Test Substance Source	High	Source of the chemical was defined in Call et al 2001 as AldrichChemical (Milwaukee, WI, USA).
	Metric 3: Test Substance Purity	High	Purity was identified as >99% in Call et al 2001.
Domain 2: Test Design			
	Metric 4: Negative Controls	High	Negative controls (sediment and silica sand) were used in this experiment.
	Metric 5: Negative Control Response	High	Biological response of control groups was appropriate as shown in Table 4.
	Metric 6: Randomized Allocation	Low	Random allocation was not reported.
Domain 3: Exposure Characterization			
	Metric 7: Experimental System/Test Media Preparation	High	Methods of sediment collection and preparation (including chemical addition) and addition of sediment to test beakers were described in detail.
	Metric 8: Consistency of Exposure Administration	High	Exposure consistency was reported and consistent among different DBP treatments and controls.
	Metric 9: Measurement of Test Substance Concentration	High	Concentrations were measured using HPLC as described in methods and the cited reference (Call et al 2001).
	Metric 10: Exposure Duration and Frequency	High	The 10-day exposure duration was appropriate for the experimental design.
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Medium	Exposure groups were acceptable and spanned five concentrations per test species in addition to the control. Nominal doses were unclear, however measured doses were reported in sediment and pore water.
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via sediment.
Domain 4: Test Organism			
	Metric 13: Test Organism Characteristics	Low	Source of test organisms was not reported.
	Metric 14: Acclimatization and Pretreatment Conditions	Low	Acclimation of test organisms prior to the exposure was not reported.
	Metric 15: Number of Organisms and Replicates per Group	Medium	Tests with DBP utilized three replicates of five different concentrations with 10 organisms per beaker, three sediment control replicates with 10 test organisms per beaker, and three silica sand control replicates with 10 test organisms per beaker.
Domain 5: Outcome Assessment			
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Study Citation:	Call, D. J., Cox, D. A., Geiger, D. L., Genisot, K. I., Markee, T. P., Brooke, L. T., Polkinghorne, C. N., Vandeventer, F. A., Gorsuch, J. W., Robillard, K. A., Parkerton, T. F., Reiley, M. C., Ankley, G. T., Mount, D. R. (2001). An assessment of the toxicity of phthalate esters to freshwater benthos. 2. Sediment exposures. Environmental Toxicology and Chemistry 20(8):1805-1815.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Sediment; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Hyalella azteca</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	679311			
Domain	Metric	Rating	Comments	
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions including feeding, intermittent water renewal system, lighting, and temperature were reported. Authors recorded temperature, dissolved oxygen, and pH on days 0 and 10 and conductivity, hardness, alkalinity, and ammonia on days 1 and 9. Sediment TOC conditions were described in Table 2.	
	Metric 17: Outcome Assessment Methodology	High	Sediment was sieved, survivors were collected, dried, and weighed.	
	Metric 18: Consistency of Outcome Assessment	High	Outcome assessment was conducted at conclusion of the 10-day exposure.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.	
	Metric 20: Outcomes Unrelated to Exposure	High	Details regarding test organism attrition and outcomes unrelated to exposure were reported for each study group and there were no differences among groups that could influence the outcome assessment.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Survival data from toxicity tests were summarized using the trimmed Spearman–Kärber method. Dry weight data were analyzed by one-way analysis of variance and Dunnett’s procedure using a SigmaStat Program.	
	Metric 22: Reporting of Data	High	Treatment and control data were reported in Table 4. Results were represented as average dry weight per individual.	
	Metric 23: Explanation of Unexpected Outcomes	Medium	Variability was not reported, but results suggest no excessive variability within replicates.	
Additional Comments: None				
Overall Quality Determination		High		

Study Citation:	Call, D. J., Markee, T. P., Geiger, D. L., Brooke, L. T., Vandeventer, F. A., Cox, D. A., Genisot, K. I., Robillard, K. A., Gorsuch, J. W., Parkerton, T. F., Reiley, M. C., Ankley, G. T., Mount, D. R. (2001). An assessment of the toxicity of phthalate esters to freshwater benthos. 1. Aqueous exposures. Environmental Toxicology and Chemistry 20(8):1798-1804.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Hyaella azteca</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	679312			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The chemical was identified by name, but no structure or CASRN were given.	
	Metric 2: Test Substance Source	High	The source of the phthalate was Aldrich Chemical. The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity was reported as >98%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was adequate.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and exposure frequency were reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were adequate for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.	
Domain 5: Outcome Assessment				
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Study Citation:	Call, D. J., Markee, T. P., Geiger, D. L., Brooke, L. T., Vandeventer, F. A., Cox, D. A., Genisot, K. I., Robillard, K. A., Gorsuch, J. W., Parkerton, T. F., Reiley, M. C., Ankley, G. T., Mount, D. R. (2001). An assessment of the toxicity of phthalate esters to freshwater benthos. 1. Aqueous exposures. Environmental Toxicology and Chemistry 20(8):1798-1804.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Hyalella azteca</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	679312			
Domain	Metric	Rating	Comments	
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.	
	Metric 20: Outcomes Unrelated to Exposure	High	There were no differences among groups that could influence the outcome assessment.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.	
	Metric 22: Reporting of Data	Medium	Mean data for exposure-related findings were shown for each treatment and control group.	
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.	
Additional Comments:	The LC50 for <i>Hyalella</i> was reported for DBP as 0.63 mg/L.			

Overall Quality Determination**High**

Study Citation:	Lake Superior Research Institute, (1997). Sediment toxicity testing program for phthalate esters.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Sediment; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Hyalella azteca</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	7325945			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	Chemical was identified by name, lot # and CAS#	
	Metric 2: Test Substance Source	Low	The test substance identity was NOT analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity reported as >99%	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable	
	Metric 6: Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail	
	Metric 8: Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups	
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source	
	Metric 14: Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms	
	Metric 15: Number of Organisms and Replicates per Group	Medium	10 per replicate with 2 replicates used	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest	
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Study Citation:	Lake Superior Research Institute, (1997). Sediment toxicity testing program for phthalate esters.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Sediment; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Hyalella azteca</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	7325945			
Domain	Metric	Rating	Comments	
	Metric 18: Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions	
	Metric 20: Outcomes Unrelated to Exposure	High	there were no differences among groups	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical methods were adequately described	
	Metric 22: Reporting of Data	High	Data for exposure-related findings were shown for each treatment and control group	
	Metric 23: Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained.	
Additional Comments:	None			

Overall Quality Determination**High**

Study Citation:	Lake Superior Research Institute, (1997). Sediment toxicity testing program for phthalate esters.		
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Sediment; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Hyaella azteca</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Development/Growth		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	7325945		
Domain	Metric	Rating	Comments
Domain 1: Test Substance			
	Metric 1: Test Substance Identity	High	Chemical was identified by name, lot # and CAS#
	Metric 2: Test Substance Source	Low	The test substance identity was NOT analytically verified by the performing laboratory.
	Metric 3: Test Substance Purity	High	Chemical purity reported as >99%
Domain 2: Test Design			
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable
	Metric 6: Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization			
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8: Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods.
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit
Domain 4: Test Organism			
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14: Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15: Number of Organisms and Replicates per Group	Medium	10 per replicate with 2 replicates used
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18: Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
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Study Citation:	Lake Superior Research Institute, (1997). Sediment toxicity testing program for phthalate esters.		
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Sediment; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Hyaella azteca</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Development/Growth		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	7325945		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	High	Statistical methods were adequately described
Metric 22:	Reporting of Data	High	Data for exposure-related findings were shown for each treatment and control group
Metric 23:	Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained.
Additional Comments:	None		

Overall Quality Determination**High**

Study Citation:	Lake Superior Research Institute, (1997). Sediment toxicity testing program for phthalate esters.		
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Sediment; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Hyaella azteca</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Mortality		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	7325945		
Domain	Metric	Rating	Comments
Domain 1: Test Substance			
	Metric 1: Test Substance Identity	High	The chemical was identified by name, lot # and CAS#.
	Metric 2: Test Substance Source	Low	The test substance identity was NOT analytically verified by the performing laboratory.
	Metric 3: Test Substance Purity	High	Chemical purity was reported as >99%.
Domain 2: Test Design			
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable.
	Metric 6: Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups.
Domain 3: Exposure Characterization			
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8: Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods.
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit.
Domain 4: Test Organism			
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 10 organisms per replicate with two replicates used.
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
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Study Citation:	Lake Superior Research Institute, (1997). Sediment toxicity testing program for phthalate esters.		
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Sediment; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Hyaella azteca</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Mortality		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	7325945		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
Metric 20:	Outcomes Unrelated to Exposure	High	There were no differences among groups.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	High	Statistical methods were adequately described.
Metric 22:	Reporting of Data	High	Data for exposure-related findings were shown for each treatment and control group.
Metric 23:	Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.
Additional Comments:	None		

Overall Quality Determination**High**

Study Citation:	Lake Superior Research Institute, (1997). Sediment toxicity testing program for phthalate esters.		
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Sediment; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Hyalella azteca</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Mortality		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	7325945		
Domain	Metric	Rating	Comments
Domain 1: Test Substance			
	Metric 1: Test Substance Identity	High	The chemical was identified by name, lot # and CAS#.
	Metric 2: Test Substance Source	Low	The test substance identity was NOT analytically verified by the performing laboratory.
	Metric 3: Test Substance Purity	High	Chemical purity was reported as >99%.
Domain 2: Test Design			
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable.
	Metric 6: Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups.
Domain 3: Exposure Characterization			
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8: Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods.
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit.
Domain 4: Test Organism			
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 10 organisms per replicate with two replicates used.
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
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Study Citation:	Lake Superior Research Institute, (1997). Sediment toxicity testing program for phthalate esters.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Sediment; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Hyaella azteca</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	7325945

Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	High	There were no differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were shown for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.

Additional Comments: None

Overall Quality Determination**High**

Study Citation:	Lake Superior Research Institute, (1997). Sediment toxicity testing program for phthalate esters.		
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Sediment; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Hyaella azteca</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Development/Growth		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	7325945		
Domain	Metric	Rating	Comments
Domain 1: Test Substance			
	Metric 1: Test Substance Identity	High	Chemical was identified by name, lot # and CAS#
	Metric 2: Test Substance Source	Low	The test substance identity was NOT analytically verified by the performing laboratory.
	Metric 3: Test Substance Purity	High	Chemical purity reported as >99%
Domain 2: Test Design			
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable
	Metric 6: Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization			
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8: Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods.
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit
Domain 4: Test Organism			
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14: Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15: Number of Organisms and Replicates per Group	Medium	10 per replicate with 2 replicates used
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18: Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
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Study Citation:	Lake Superior Research Institute, (1997). Sediment toxicity testing program for phthalate esters.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Sediment; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Hyalella azteca</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	7325945

Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
	Metric 20: Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described
	Metric 22: Reporting of Data	High	Data for exposure-related findings were shown for each treatment and control group
	Metric 23: Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained.

Additional Comments: None

Overall Quality Determination**High**

Study Citation:	Lake Superior Research Institute, (1997). Sediment toxicity testing program for phthalate esters.		
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Hyalella azteca</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Mortality		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	7325945		
Domain	Metric	Rating	Comments
Domain 1: Test Substance			
	Metric 1: Test Substance Identity	High	The chemical was identified by name, lot # and CAS#.
	Metric 2: Test Substance Source	Low	The test substance identity was NOT analytically verified by the performing laboratory.
	Metric 3: Test Substance Purity	High	Chemical purity was reported as >99%.
Domain 2: Test Design			
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable.
	Metric 6: Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups.
Domain 3: Exposure Characterization			
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8: Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods.
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit.
Domain 4: Test Organism			
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 10 organisms per replicate with two replicates used.
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
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Study Citation:	Lake Superior Research Institute, (1997). Sediment toxicity testing program for phthalate esters.		
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Hyaella azteca</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Mortality		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	7325945		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
Metric 20:	Outcomes Unrelated to Exposure	High	There were no differences among groups.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	High	Statistical methods were adequately described.
Metric 22:	Reporting of Data	Medium	Data for exposure-related findings were not shown for each treatment and control group, but results were described in the tables.
Metric 23:	Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.
Additional Comments: None			
Overall Quality Determination		High	

Study Citation:	Lake Superior Research Institute, (1997). Sediment toxicity testing program for phthalate esters.		
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Sediment; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Hyalella azteca</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Mortality		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	7325945		
Domain	Metric	Rating	Comments
Domain 1: Test Substance			
	Metric 1: Test Substance Identity	High	The chemical was identified by name, lot # and CAS#.
	Metric 2: Test Substance Source	Low	The test substance identity was NOT analytically verified by the performing laboratory.
	Metric 3: Test Substance Purity	High	Chemical purity was reported as >99%.
Domain 2: Test Design			
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable.
	Metric 6: Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups.
Domain 3: Exposure Characterization			
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8: Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods.
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit.
Domain 4: Test Organism			
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 10 organisms per replicate with two replicates used.
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
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Study Citation:	Lake Superior Research Institute, (1997). Sediment toxicity testing program for phthalate esters.		
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Sediment; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Hyaella azteca</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Mortality		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	7325945		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
Metric 20:	Outcomes Unrelated to Exposure	High	There were no differences among groups.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	High	Statistical methods were adequately described.
Metric 22:	Reporting of Data	High	Data for exposure-related findings were shown for each treatment and control group.
Metric 23:	Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.
Additional Comments:	None		

Overall Quality Determination**High**

Study Citation:	Jr, Mayer, F., Sanders, H. O., Walsh, D. F. (1973). Toxicity, residue dynamics, and reproductive effects of phthalate esters in aquatic invertebrates. Environmental Research 6(1):84-90.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Ischnura verticalis</i> ; Juvenile			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1334646			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The chemical was identified by name.
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Uninformative	A concurrent negative control group was not included or reported.
	Metric 5:	Negative Control Response	Low	The biological response of the negative control group was not reported.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.
	Metric 8:	Consistency of Exposure Administration	Low	Few details of exposure administration were reported and exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	Only one treatment was reported.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test animals was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of test replicates was not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.
	Metric 17:	Outcome Assessment Methodology	Medium	The outcome assessment methodology was not clearly reported.

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Study Citation:	Jr, Mayer, F., Sanders, H. O., Walsh, D. F. (1973). Toxicity, residue dynamics, and reproductive effects of phthalate esters in aquatic invertebrates. Environmental Research 6(1):84-90.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Ischnura verticalis</i> ; Juvenile			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1334646			
Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	Medium	Few details regarding the execution of the study protocol for outcome assessment were provided.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was not performed.
	Metric 22:	Reporting of Data	Medium	Data for exposure-related findings were presented for the sampling period.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments: None				

Overall Quality Determination**Uninformative**

Study Citation:	Call, D. J., Markee, T. P., Geiger, D. L., Brooke, L. T., Vandeventer, F. A., Cox, D. A., Genisot, K. I., Robillard, K. A., Gorsuch, J. W., Parkerton, T. F., Reiley, M. C., Ankley, G. T., Mount, D. R. (2001). An assessment of the toxicity of phthalate esters to freshwater benthos. 1. Aqueous exposures. Environmental Toxicology and Chemistry 20(8):1798-1804.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); <i>Lumbriculus variegatus</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	679312			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The chemical was identified by name, but no CASRN or structure were provided.	
	Metric 2: Test Substance Source	High	The source of the phthalate was Aldrich Chemical. The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity was reported as >98%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was adequate.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and exposure frequency were reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were adequate for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.	
Domain 5: Outcome Assessment				
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Study Citation:	Call, D. J., Markee, T. P., Geiger, D. L., Brooke, L. T., Vandeventer, F. A., Cox, D. A., Genisot, K. I., Robillard, K. A., Gorsuch, J. W., Parkerton, T. F., Reiley, M. C., Ankley, G. T., Mount, D. R. (2001). An assessment of the toxicity of phthalate esters to freshwater benthos. 1. Aqueous exposures. Environmental Toxicology and Chemistry 20(8):1798-1804.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); <i>Lumbriculus variegatus</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	679312			
Domain	Metric	Rating	Comments	
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.	
	Metric 20: Outcomes Unrelated to Exposure	High	There were no differences among groups that could influence the outcome assessment.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.	
	Metric 22: Reporting of Data	Medium	Mean data for exposure-related findings were shown for each treatment and control group.	
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.	
Additional Comments:	The LC50 for DBP to Lumbriculus was reported as 2.48 mg/L.			
Overall Quality Determination		High		

Study Citation:	Lake Superior Research Institute, (1997). Sediment toxicity testing program for phthalate esters.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); <i>Lumbriculus variegatus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	7325945			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The chemical was identified by name, lot # and CAS#.	
	Metric 2: Test Substance Source	Low	The test substance identity was NOT analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity was reported as >99%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable.	
	Metric 6: Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.	
	Metric 8: Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 10 organisms per replicate with two replicates used.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	
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Study Citation:	Lake Superior Research Institute, (1997). Sediment toxicity testing program for phthalate esters.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); <i>Lumbriculus variegatus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	7325945			
Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	High	There were no differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described.
	Metric 22:	Reporting of Data	Medium	Data for exposure-related findings were not shown for each treatment and control group, but results were described in the tables.
	Metric 23:	Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.
Additional Comments: None				

Overall Quality Determination**High**

Study Citation:	Sung, H. H., Kao, W. Y., Su, Y. J. (2003). Effects and toxicity of phthalate esters to hemocytes of giant freshwater prawn, <i>Macrobrachium rosenbergii</i> . Aquatic Toxicology 64(1):25-37.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Macrobrachium rosenbergii</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Cell signaling/function			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	789598			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Medium	Chemical was identified by correct nomenclature and chemical structure. CASRN was not reported.
	Metric 2:	Test Substance Source	High	Source of BBP was Aldrich Chem. Co.
	Metric 3:	Test Substance Purity	Low	Purity was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Negative controls used.
	Metric 5:	Negative Control Response	High	Biological responses of controls were appropriate.
	Metric 6:	Randomized Allocation	Low	Organisms were purchased from local prawn farms on separate days. The hemocytes isolated from 5 or 10 prawns were used for the assays, but the authors did not specifically mention random allocation of the hemocytes from different prawns for the various assays.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The authors reported that the phthalate stocks were separately dissolved in acetone and diluted with M-199 (hemocyte-culture medium) to a concentration of 1000 mg/ml. They also reported the final concentration once the phthalates were added to the hemocyte suspension. However, the authors did not report the acetone concentration employed.
	Metric 8:	Consistency of Exposure Administration	High	Exposure administration was consistent across groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not reported/measured.
	Metric 10:	Exposure Duration and Frequency	High	Exposure duration was appropriate to assess cell death (necrosis and apoptosis) and cell morphology impacts in the hemocytes.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	In Table 1, the authors reported treatment with different concentration (25, 50 and 100 mg/ml) of PAEs. However, there is limited information in the text and it is unclear how each concentration was achieved and if all three concentrations were included in all assays or only some of the assays.
	Metric 12:	Testing at or Below Solubility Limit	Medium	Below solubility limit as reported. But the authors did not report the concentration of acetone used to dissolve the phthalate and did not measure the phthalate concentration.
Domain 4: Test Organism				
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Study Citation:	Sung, H. H., Kao, W. Y., Su, Y. J. (2003). Effects and toxicity of phthalate esters to hemocytes of giant freshwater prawn, <i>Macrobrachium rosenbergii</i> . Aquatic Toxicology 64(1):25-37.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Macrobrachium rosenbergii</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Cell signaling/function			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	789598			
Domain	Metric	Rating	Comments	
	Metric 13: Test Organism Characteristics	Medium	Test organisms (freshwater prawn) were obtained from a market and hemolymph drawn from these test organisms was then used to isolate the hemocytes employed in the in vitro exposure and assays. Information on the size/age of prawn was not provided.	
	Metric 14: Acclimatization and Pretreatment Conditions	Medium	Test organisms (freshwater prawn) were acclimated in fresh pond water in 120 L plastic containers at 30 8C for 3 days prior to experiments, and stocking densities were maintained at 20 prawns per container. However, there was no indication of the health or stress status of test organism which can affect the immune system. The exposure to phthalates was in vitro using hemocytes (immune cells) isolated from prawns.	
	Metric 15: Number of Organisms and Replicates per Group	Low	This was an in vitro study to determine impacts on hemocytes (immune cells) measured by cell death (necrosis and apoptosis) and cell morphology. The number of organisms used (5-10) to collect a diverse pool of hemocytes and cell suspensions was reported. However, authors offered insufficient details on, for example, the number of replicates examined by gel electrophoresis or the number of sections and replicate fields for the electron microscopy assessment.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	This was an in vitro exposure study to determine impacts on hemocytes measured by cell death (necrosis and apoptosis) and cell morphology. In vitro exposure conditions were adequate.	
	Metric 17: Outcome Assessment Methodology	High	Outcome assessment methodologies using hemocytes [the annexin assay, gel electrophoresis, transmission electron microscopy] were reported and appropriate for the outcomes of interest in hemocytes [cell death by necrosis and apoptosis and cell morphology changes].	
	Metric 18: Consistency of Outcome Assessment	High	Details of the in vitro assays to examine hemocyte toxicity/viability were reported and assessed consistently across groups.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	The study was an in vitro exposure. As described, hemocyte suspensions were prepared in adequate medium conditions and treated consistently.	
	Metric 20: Outcomes Unrelated to Exposure	High	Outcomes unrelated to exposure were not reported.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	Low	Statistical analysis was reported but not explained in detail.	
	Metric 22: Reporting of Data	High	Data were reported for each assay and for all treatment groups per assay. The summary of the effects in Table 1 reported the inhibition or enhancement concentration for each assay.	
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Study Citation:	Sung, H. H., Kao, W. Y., Su, Y. J. (2003). Effects and toxicity of phthalate esters to hemocytes of giant freshwater prawn, <i>Macrobrachium rosenbergii</i> . Aquatic Toxicology 64(1):25-37.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Macrobrachium rosenbergii</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Cell signaling/function
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	789598

Domain	Metric	Rating	Comments
	Metric 23: Explanation of Unexpected Outcomes	Low	Authors did not report SE, SD, CI or such variability details for the cell death (necrosis and apoptosis) and cell morphology data.

Additional Comments: This study was an in vitro experiment where hemocytes (immune cells) isolated from the hemolymph of 5 to 10 giant freshwater prawn (*Macrobrachium rosenbergii*) were exposed to BBP, DBP, DEHP, or DCHP. Endpoints encompassed nonspecific cell-mediated immune function assays as well as hemocyte viability assays: (1) Nonspecific cell-mediated immune defense response assays included determination of hemocytic adhesion and pseudopodia formation (a measure of the initial procedures of either phagocytosis or encapsulation), phenoloxidase activity assay (a measure of pathogen recognition and defense functions), and nitroblue tetrazolium solution (NBT) assay to determine superoxide production (a measure of highly microbicidal activity); (2) Hemocyte toxicity/viability assays included detection of cell death via necrosis, detection of cell death via apoptosis, and impacts on cellular morphology assessed by microscopy. This form was used to evaluate hemocyte toxicity/viability due to DBP.

Overall Quality Determination

Medium

Study Citation:	Sung, H. H., Kao, W. Y., Su, Y. J. (2003). Effects and toxicity of phthalate esters to hemocytes of giant freshwater prawn, <i>Macrobrachium rosenbergii</i> . Aquatic Toxicology 64(1):25-37.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Macrobrachium rosenbergii</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Cell signaling/function			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	789598			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Medium	Chemical was identified by correct nomenclature and chemical structure. CASRN was not reported.
	Metric 2:	Test Substance Source	High	Source of DBP was Riedelde Haen Co.
	Metric 3:	Test Substance Purity	Low	Purity was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Negative controls were used.
	Metric 5:	Negative Control Response	High	Biological responses of controls were appropriate.
	Metric 6:	Randomized Allocation	Low	Organisms were purchased from local prawn farms on separate days. The hemocytes isolated from 5 or 10 prawns were used for the assays, but the authors did not specifically mention random allocation of the hemocytes from different prawns for the various assays.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The authors reported that the phthalate stocks were separately dissolved in acetone and diluted with M-199 (hemocyte-culture medium) to a concentration of 1000 mg/ml. They also reported the final concentration once the phthalates were added to the hemocyte suspension. However, the authors did not report the acetone concentration employed.
	Metric 8:	Consistency of Exposure Administration	High	The exposure was consistent across groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not reported/measured.
	Metric 10:	Exposure Duration and Frequency	High	An appropriate exposure time was used.
	Metric 11:	Number of Exposure Groups/Spacing of Exposure Levels	Medium	In Table 1, the authors reported treatment with different concentration (25, 50 and 100 mg/ml) of PAEs. However, there is limited information in the text and it is unclear how each concentration was achieved and if all three concentrations were included in all assays or only some of the assays.
	Metric 12:	Testing at or Below Solubility Limit	Medium	Testing was below the solubility limit as reported. But the authors did not report the concentration of acetone used to dissolve the phthalate and did not measure the phthalate concentration.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	Test organisms (freshwater prawn) were obtained from a market and hemolymph drawn from these test organisms was then used to isolate the hemocytes employed in the in vitro exposure and assays. Information on the size/age of prawn was not provided.
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Study Citation:	Sung, H. H., Kao, W. Y., Su, Y. J. (2003). Effects and toxicity of phthalate esters to hemocytes of giant freshwater prawn, <i>Macrobrachium rosenbergii</i> . Aquatic Toxicology 64(1):25-37.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Macrobrachium rosenbergii</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Cell signaling/function			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	789598			
Domain	Metric	Rating	Comments	
	Metric 14:	Acclimatization and Pretreatment Conditions	Medium	Test organisms (freshwater prawn) were acclimated in fresh pond water in 120 L plastic containers at 30 C for 3 days prior to experiments, and stocking densities were maintained at 20 prawns per container. However, there was no indication of the health or stress status of test organism which can affect immune function (i.e., stress can modulate immune responses (cell-mediated and humoral). The exposure to phthalates was in vitro using hemocytes (immune cells) isolated from prawns.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	This was an in vitro study to determine impacts on nonspecific cell-mediated immune defense responses. The number of organisms used (5-10) to collect a diverse pool of hemocytes, number of cells and cell suspensions, number of well-plates, and number of replicate fields for microscopic counts were sufficient.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	This was an in vitro exposure study to determine impacts on nonspecific cell-mediated immune defense responses. In vitro exposure conditions were adequate.
	Metric 17:	Outcome Assessment Methodology	High	Outcome assessment methodologies [determination of hemocytic adhesion and pseudopodia formation assay, the phenoloxidase activity assay, and the nitroblue tetrazolium assay] were reported and appropriate for the outcomes of interest [phagocytosis and encapsulation activity, pathogen recognition, and superoxide activity as a measure of microbicidal activity].
	Metric 18:	Consistency of Outcome Assessment	High	Details of the immune function in vitro assays were reported and assessed consistently across groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	The study was an in vitro exposure. As described, hemocyte suspensions were prepared in adequate medium conditions and treated consistently across treatment groups.
	Metric 20:	Outcomes Unrelated to Exposure	High	Outcomes unrelated to exposure were not reported.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was reported but not explained in detail.
	Metric 22:	Reporting of Data	High	Data were reported for each assay and for all treatment groups per assay. The summary of the effects in Table 1 reported the inhibition or enhancement concentration for each assay.
	Metric 23:	Explanation of Unexpected Outcomes	High	Authors reported the standard error of the mean for the nonspecific immune response data: hemocyte adhesion and pseudopodia formation, phenoloxidase activity, and superoxide production (Figures 2 and 3).
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Study Citation:	Sung, H. H., Kao, W. Y., Su, Y. J. (2003). Effects and toxicity of phthalate esters to hemocytes of giant freshwater prawn, <i>Macrobrachium rosenbergii</i> . Aquatic Toxicology 64(1):25-37.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Macrobrachium rosenbergii</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Cell signaling/function
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	789598

Domain	Metric	Rating	Comments
Additional Comments:	This study was an in vitro experiment where hemocytes (immune cells) isolated from the hemolymph of 5 to 10 giant freshwater prawn (<i>Macrobrachium rosenbergii</i>) were exposed to BBP, DBP, DEHP, or DCHP. Endpoints encompassed nonspecific cell-mediated immune function assays as well as hemocyte viability assays: (1) Nonspecific cell-mediated immune defense response assays included determination of hemocytic adhesion and pseudopodia formation (a measure of the initial procedures of either phagocytosis or encapsulation), phenoloxidase activity assay (a measure of pathogen recognition and defense functions), and nitroblue tetrazolium solution (NBT) assay to determine superoxide production (a measure of highly microbicidal activity); (2) Hemocyte toxicity/viability assays included detection of cell death via necrosis, detection of cell death via apoptosis, and impacts on cellular morphology assessed by microscopy. This form was used to evaluate impacts to the nonspecific cell-mediated immune defense responses due to DBP.		

Overall Quality Determination**Medium**

Study Citation:	Jr, Mayer, F., Sanders, H. O., Walsh, D. F. (1973). Toxicity, residue dynamics, and reproductive effects of phthalate esters in aquatic invertebrates. Environmental Research 6(1):84-90.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Orconectus nais</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1334646			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The chemical was identified by name.
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the amount of details provided in the study being reviewed.
	Metric 5:	Negative Control Response	Low	The biological response of the negative control group was not reported.
	Metric 6:	Randomized Allocation	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the amount of details provided in the study being reviewed.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	Concentrations of the test substance were not measured during the study.
	Metric 8:	Consistency of Exposure Administration	Medium	Some details of exposure administration were reported and exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Uninformative	No information is provided on the number of exposure groups and spacing of exposure levels.
	Metric 12:	Testing at or Below Solubility Limit	Low	Reporting omissions prevented determination of whether exposure concentrations exceeded the water solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test animals was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of test replicates was not reported.
Domain 5: Outcome Assessment				
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Study Citation:	Jr, Mayer, F., Sanders, H. O., Walsh, D. F. (1973). Toxicity, residue dynamics, and reproductive effects of phthalate esters in aquatic invertebrates. Environmental Research 6(1):84-90.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Orconectus nais</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1334646			
Domain	Metric	Rating	Comments	
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported.
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for the outcome assessment were not reported.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22:	Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group.
	Metric 23:	Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability.
Additional Comments:	None			

Overall Quality Determination**Uninformative**

Study Citation:	Jr, Mayer, F., Sanders, H. O., Walsh, D. F. (1973). Toxicity, residue dynamics, and reproductive effects of phthalate esters in aquatic invertebrates. Environmental Research 6(1):84-90.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Paleomonetes kadiakensis</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1334646			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The chemical was identified by name.
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Uninformative	A concurrent negative control group was not included or reported.
	Metric 5:	Negative Control Response	Low	The biological response of the negative control group was not reported.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.
	Metric 8:	Consistency of Exposure Administration	Low	Few details of exposure administration were reported and exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	Only one treatment was reported.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test animals was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of test replicates was not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.
	Metric 17:	Outcome Assessment Methodology	Medium	The outcome assessment methodology was not clearly reported.

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Study Citation:	Jr, Mayer, F., Sanders, H. O., Walsh, D. F. (1973). Toxicity, residue dynamics, and reproductive effects of phthalate esters in aquatic invertebrates. Environmental Research 6(1):84-90.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Paleomonetes kadiakensis</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1334646			
Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	Medium	Few details regarding the execution of the study protocol for outcome assessment were provided.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was not performed.
	Metric 22:	Reporting of Data	Medium	Data for exposure-related findings were presented for the sampling period.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments: None				

Overall Quality Determination**Uninformative**

Study Citation:	Adams, W. J., Biddinger, G. R., Robillard, K. A., Gorsuch, J. W. (1995). A summary of the acute toxicity of 14 phthalate esters to representative aquatic organisms. Environmental Toxicology and Chemistry 14(9):1569-1574.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Paratanytarsus parthenogenetica</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1321996; Linked HERO ID(s): 1321996, 1316224			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	Test substance nomenclature was reported without CASRN.	
	Metric 2: Test Substance Source	Low	The source was reported as provided by manufacturer from commercially available batches. Manufacture name and batch number were not provided. No analytical data was reported.	
	Metric 3: Test Substance Purity	High	The chemical was at least 95% pure.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	A negative control was reported.	
	Metric 5: Negative Control Response	High	The control response was acceptable.	
	Metric 6: Randomized Allocation	Low	An allocation method was not reported.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The experimental system was well described. However, headspace or measures to prevent volatilization were not reported.	
	Metric 8: Consistency of Exposure Administration	High	Exposure administration was consistent across groups.	
	Metric 9: Measurement of Test Substance Concentration	Medium	Sample extracts were analyzed by gas chromatography at the start and end of the test. In static studies, final test concentrations frequently were 50% of the initial concentrations. Loss of the phthalate esters was thought to be principally due to adsorption to the test vessels.	
	Metric 10: Exposure Duration and Frequency	High	Duration and frequency of exposure were appropriate for this test.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	Exposure levels were appropriate. A range finding test was performed.	
	Metric 12: Testing at or Below Solubility Limit	High	The test was performed at or below water solubility.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	A source was not reported.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	An appropriate acclimation for the test was reported.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	Each test was performed using duplicate test concentrations with 10 organisms per test vessel.	
Domain 5: Outcome Assessment				
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Study Citation:	Adams, W. J., Biddinger, G. R., Robillard, K. A., Gorsuch, J. W. (1995). A summary of the acute toxicity of 14 phthalate esters to representative aquatic organisms. Environmental Toxicology and Chemistry 14(9):1569-1574.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Paratanytarsus parthenogenetica</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1321996; Linked HERO ID(s): 1321996, 1316224			
Domain	Metric	Rating	Comments	
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions were appropriate for the test.	
	Metric 17: Outcome Assessment Methodology	High	The intended outcomes were reported.	
	Metric 18: Consistency of Outcome Assessment	High	Outcome assessment was consistent across groups.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	Environmental conditions were consistent across groups.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There were no reported differences between groups.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical methods were performed and described.	
	Metric 22: Reporting of Data	Medium	Only treatment endpoints were reported.	
	Metric 23: Explanation of Unexpected Outcomes	High	No unexpected outcomes were reported.	
Additional Comments:	None			
Overall Quality Determination		High		

Study Citation:	Bionomics,, EG&G (1984). Acute toxicity of twelve phthalate esters to <i>Paratanytarsus parthenogenica</i> (final report) report no BW-83-6-1424.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Paratanytarsus parthenogenica</i> ; Larvae			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1316219			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	Test substance was identified by name and CASRN.	
	Metric 2: Test Substance Source	High	The phthalate ester was received from General Electric Company.	
	Metric 3: Test Substance Purity	High	Purity was reported as 100% active ingredient.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Negative controls were used.	
	Metric 5: Negative Control Response	High	Percent mortality for controls was shown in Table 3.	
	Metric 6: Randomized Allocation	Medium	Organisms were impartially distributed into the test vessels.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	Static test conditions were described in detail.	
	Metric 8: Consistency of Exposure Administration	Medium	Details of the exposure administration were reported but limited (volumes of chemicals used to make stock solutions were not reported).	
	Metric 9: Measurement of Test Substance Concentration	High	Test concentrations were verified analytically as shown in Table 1 and described in Appendix I.	
	Metric 10: Exposure Duration and Frequency	High	The exposure duration was reported and adequate (mortality reported at 48hr and 24hr).	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Five concentrations of DBP were tested spanning approximately one order of magnitude between the highest and lowest concentration.	
	Metric 12: Testing at or Below Solubility Limit	High	Concentrations were approximately at or below the solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	Paratanytarsus parthenogenica were obtained from cultured stocks (EG&G Bionomics); age was reported as second or third instars.	
	Metric 14: Acclimatization and Pretreatment Conditions	Medium	It was not specifically stated if organisms were acclimatized.	
	Metric 15: Number of Organisms and Replicates per Group	Low	Three replicate beakers per concentration with five midge larvae per beaker.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Test conditions were well described, reported and followed cited protocols.	
	Metric 17: Outcome Assessment Methodology	Medium	There were limited details of how authors determined mortality of the test organisms.	
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Study Citation:	Bionomics,, EG&G (1984). Acute toxicity of twelve phthalate esters to <i>Paratanytarsus parthenogenica</i> (final report) report no BW-83-6-1424.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Paratanytarsus parthenogenica</i> ; Larvae			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1316219			
Domain	Metric	Rating	Comments	
	Metric 18: Consistency of Outcome Assessment	High	Mortality assessment was conducted at 24 and 48hr and appeared to be consistent among study groups.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	No differences were reported.	
	Metric 20: Outcomes Unrelated to Exposure	High	Details regarding test organism attrition and outcomes unrelated to exposure (e.g., infection) were reported for each study group and there were no differences among groups that could influence the outcome assessment.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	The LC50 at 48 hr exposure was determined via moving angle analysis as described in the footnote of Table 4.	
	Metric 22: Reporting of Data	High	Outcomes were described in Table 3 and LC50 values were shown in Table 4.	
	Metric 23: Explanation of Unexpected Outcomes	Medium	Outcomes were explained in the study.	
Additional Comments:	Authors conducted dose response of DBP and limit tests for DEHP, DIDP, and DINP. Authors report percent morality in replicate groups at 24 and 48 hr exposure. LC50 values for DBP were reported. LC50 values for DEHP, DIDP, and DINP also reported but unclear how authors obtained (or estimated) LC50 values based on a limit test.			

Overall Quality Determination**High**

Study Citation:	Kuang, Q. J., Zhao, W. Y., Cheng, S. P. (2003). Toxicity of dibutyl phthalate to algae. Bulletin of Environmental Contamination and Toxicology 71(3):602-608.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Algae</i> ; natural algae; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332820			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The test substance was identified by name only.	
	Metric 2: Test Substance Source	Low	It was reported the DBP was manufactured in Shanghai, China but this is not enough information on the source. It was not reported if the DBP was analytically verified.	
	Metric 3: Test Substance Purity	High	The purity was reported to be 99%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	Low	Study authors reported the use of a concurrent negative control. It was reported that a vehicle solvent was used, but it was unclear if the control was a solvent control.	
	Metric 5: Negative Control Response	High	The biological response of the negative control was adequate and reported in Table 2.	
	Metric 6: Randomized Allocation	Low	It was not reported how the algae was allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	Limited details were reported on the preparation of the test media and concentrations of test substance were not measured during the study. The solvent concentration was not reported.	
	Metric 8: Consistency of Exposure Administration	Medium	Details of the exposure administration were reported. The test chambers were 50L buckets open to conditions. The temperatures were reported to be between 16-24C, which could lead to variation.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if the test substance was measured at any point in the study.	
	Metric 10: Exposure Duration and Frequency	Medium	The exposure duration was reported to be seven days, which is longer than is typical for acute toxicity studies with algae.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	There were four exposure groups plus a control, and the spacing of the exposure levels was adequate for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	Low	It was reported that a vehicle solvent was used, but it was unclear if the control was a solvent control. The solvent concentration was not reported.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	It was reported that the algae was obtained from East Lake in Wuhan, China. This leaves doubt about the health of the organisms.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the algae was acclimated to the test conditions prior to the start of the test.	
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Study Citation:	Kuang, Q. J., Zhao, W. Y., Cheng, S. P. (2003). Toxicity of dibutyl phthalate to algae. Bulletin of Environmental Contamination and Toxicology 71(3):602-608.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Algae</i> ; natural algae; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332820			
Domain	Metric	Rating	Comments	
	Metric 15: Number of Organisms and Replicates per Group	Low	Initial cell counts for each exposure and control were reported in Table 2. Study authors did not report the number of replicates.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Medium	The test was conducted at 16-24C in 50L buckets with a natural photoperiod length that was not elaborated on. It is unclear if any sort of algal media was used. It was reported that lake water was used in the study.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest–algal density-development/growth.	
	Metric 18: Consistency of Outcome Assessment	Medium	Details of the outcome assessment protocol were not clearly reported but outcomes were assessed consistently across study groups. Cell counts were performed with a 0.1mm^3 plankton counting chamber under a microscope every two days.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions. It was not reported if the algae was acclimated.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	Low	Study authors did not report conducting statistical analysis, but an independent statistical analysis could be conducted from the data provided in Table 2.	
	Metric 22: Reporting of Data	High	Data for the exposure response and the control response is reported in Table 2. Exposure related findings are also presented in Figure 3.	
	Metric 23: Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability.	
Additional Comments:	This portion of the evaluation was on the acute toxicity of DBP on natural algae obtained from East Lake in Wuhan, China. The study measured cell concentrations, so development and growth was selected as the outcome of interest. Algae was selected as the taxa, as the study was on mixed algae species found in the lake. The paper listed several genera that were found, but the results did not elaborate on any genera.			
Overall Quality Determination		Low		

Study Citation:	Melin, C., Egneus, H. (1983). Effects of di-n-butyl phthalate on growth and photosynthesis in algae and on isolated organelles from higher plants. Physiologia Plantarum 59(3):461-466.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Chlorella emersonii</i> ; CCAP strain 211/8h; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Photosynthesis			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1333016			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The DBP was identified by name only.	
	Metric 2: Test Substance Source	Low	The source of the DBP was not reported.	
	Metric 3: Test Substance Purity	Low	The purity/grade of the DBP was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of a negative control, which was a solvent control containing ethanol.	
	Metric 5: Negative Control Response	High	The negative control response was reported in Figure 2.	
	Metric 6: Randomized Allocation	Low	It was not reported how the algae was allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The authors provided limited details on the preparation of the DBP test solution.	
	Metric 8: Consistency of Exposure Administration	Low	Details regarding the exposure administration were limited. Study authors reported the temperature and test volumes though.	
	Metric 9: Measurement of Test Substance Concentration	Low	Study authors did not report if the test concentrations were measured at any point in the study.	
	Metric 10: Exposure Duration and Frequency	Low	It was reported that the algae were exposed for 40 minutes.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Uninformative	The exposure levels for the test on C. emersonii were not reported.	
	Metric 12: Testing at or Below Solubility Limit	High	Study authors reported the use of a vehicle solvent.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the algae was not reported.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if any acclimation occurred.	
	Metric 15: Number of Organisms and Replicates per Group	Low	The algae concentration and the number of replicates was not reported for this part of the study.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Egneus and Blanck 1977 were cited for the culturing techniques, but details were limited in the paper.	

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Study Citation:	Melin, C., Egneus, H. (1983). Effects of di-n-butyl phthalate on growth and photosynthesis in algae and on isolated organelles from higher plants. <i>Physiologia Plantarum</i> 59(3):461-466.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Chlorella emersonii</i> ; CCAP strain 211/8h; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Photosynthesis			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1333016			
Domain	Metric	Rating	Comments	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—CO2 -dependent oxygen evolution as a measurement of photosynthesis.	
	Metric 18: Consistency of Outcome Assessment	Low	Limited details were provided on the protocol for outcome assessment.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. It was not reported if the algae was acclimated.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to the exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Growth curve determination and IC50 value determination were described in the text.	
	Metric 22: Reporting of Data	High	Exposure data and control response were reported in Figure 2.	
	Metric 23: Explanation of Unexpected Outcomes	High	Variability was reported in Figure 2.	
Additional Comments:	This portion of the evaluation is on the effect of DBP on CO2-dependent oxygen evolution as a measure of photosynthesis. The mechanistic photosynthesis outcome was chosen. This portion received an unacceptable rating due to the lack of information on exposure concentrations.			

Overall Quality Determination**Uninformative**

Study Citation:	Melin, C., Egneus, H. (1983). Effects of di-n-butyl phthalate on growth and photosynthesis in algae and on isolated organelles from higher plants. <i>Physiologia Plantarum</i> 59(3):461-466.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Chlorella emersonii</i> ; CCAP strain 211/8h; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1333016			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The DBP was identified by name only.	
	Metric 2: Test Substance Source	Low	The source of the DBP was not reported.	
	Metric 3: Test Substance Purity	Low	The purity/grade of the DBP was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of a negative control, which was a solvent control containing ethanol.	
	Metric 5: Negative Control Response	Low	The negative control response was not reported for <i>C. emersonii</i> .	
	Metric 6: Randomized Allocation	Low	It was not reported how the algae was allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The authors provided limited details on the preparation of the DBP test solution.	
	Metric 8: Consistency of Exposure Administration	Low	Details regarding the exposure administration were limited. Study authors reported the temperature and test volumes though.	
	Metric 9: Measurement of Test Substance Concentration	Low	Study authors did not report if the test concentrations were measured at any point in the study.	
	Metric 10: Exposure Duration and Frequency	Low	Study authors did not explicitly state the duration of the study, but Figure 1 indicates it was seven days.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Uninformative	The exposure levels for the growth test on <i>C. emersonii</i> were not reported. It was reported that the results were similar to the <i>S. capricornutum</i> , but it was unclear if the same exposure levels were used.	
	Metric 12: Testing at or Below Solubility Limit	High	Study authors reported the use of a vehicle solvent.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the algae was not reported.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if any acclimation occurred.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	It was reported that the initial algal density was 0.1ug chlorophyll/mL. There were three culture flasks per concentration.	
Domain 5: Outcome Assessment				
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Study Citation:	Melin, C., Egneus, H. (1983). Effects of di-n-butyl phthalate on growth and photosynthesis in algae and on isolated organelles from higher plants. Physiologia Plantarum 59(3):461-466.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Chlorella emersonii</i> ; CCAP strain 211/8h; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1333016			
Domain	Metric	Rating	Comments	
	Metric 16: Adequacy of Test Conditions	Low	Egneus and Blanck 1977 were cited for the culturing techniques, but details were limited in the paper.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest–algal growth.	
	Metric 18: Consistency of Outcome Assessment	Low	It was reported that algae growth was determined by measuring changes in transmittance at 729nm, but methods and instruments used were not reported.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. It was not reported if the algae was acclimated.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to the exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Growth curve determination and IC50 value determination were described in the text.	
	Metric 22: Reporting of Data	Low	Data was reported in the text, but continuous data was not presented.	
	Metric 23: Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability.	
Additional Comments:	This portion of the evaluation is on the effect of DBP on the growth of algae <i>C. emersonii</i> . Development/growth was selected as the outcome of interest. This portion of the study was rated unacceptable due to the lack of information regarding exposure groups and spacing; test concentrations were not reported.			

Overall Quality Determination**Uninformative**

Study Citation:	Gu, S., Zheng, H., Xu, Q., Sun, C., Shi, M., Wang, Z., Li, F. (2017). Comparative toxicity of the plasticizer dibutyl phthalate to two freshwater algae. <i>Aquatic Toxicology</i> 191:122-130.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Chlorella pyrenoidosa</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5433509			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Medium	The test substance was identified by name in the paper. The supplementary material had information regarding the structure and other physical characteristics.
	Metric 2:	Test Substance Source	Low	The source of the DBP was reported to be Sino-pharm Chemical Reagent Co. Ltd. in Nanjing, China, but it was not reported if it was analytically verified.
	Metric 3:	Test Substance Purity	High	The purity was reported to be 99% pure.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of a concurrent negative control.
	Metric 5:	Negative Control Response	High	The negative control response was reported in Figures 1 and 3 and was adequate for the outcome of interest.
	Metric 6:	Randomized Allocation	Low	It was not reported how the algae was allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The study occurred in flasks. The test solution was prepared by creating a stock solution in which DBP was dissolved into acetone for a concentration of 100g/L. This was then diluted to the proper test concentrations using BG-11 algae growth medium.
	Metric 8:	Consistency of Exposure Administration	High	All exposures occurred in 500mL flasks for 96h using BG-11 growth media. All tests were started with the same initial algal cell count.
	Metric 9:	Measurement of Test Substance Concentration	Low	It was not reported if the test concentrations were measured.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration was reported to be 96h, which is typical of an algae growth study.
	Metric 11:	Number of Exposure Groups/Spacing of Exposure Levels	High	There were five exposure levels, and the spacing was adequate to observe a dose response.
	Metric 12:	Testing at or Below Solubility Limit	Medium	Acetone was reported to be the vehicle solvent, but it was unclear if the negative control was a solvent control.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Both algal species were from the Freshwater Algae Culture Collection at the Chinese Academy of Sciences.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the organisms were acclimated at any point.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	<i>S. obliquus</i> initial concentrations were reported to be 2.02 x 10 ⁶ cells/mL, and each concentration was performed in triplicate.

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Study Citation:	Gu, S., Zheng, H., Xu, Q., Sun, C., Shi, M., Wang, Z., Li, F. (2017). Comparative toxicity of the plasticizer dibutyl phthalate to two freshwater algae. <i>Aquatic Toxicology</i> 191:122-130.		
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)		
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Chlorella pyrenoidosa</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Development/Growth		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	5433509		
Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	The algae was cultured in BG-11 algal medium with a 12L:12D photoperiod at 24C and 8000lux.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest–cell density.
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. Samples were taken at 0, 24, 48, 72, and 96h and counted with a hemocytometer.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were described in the "statistical analysis" section of the paper.
	Metric 22: Reporting of Data	High	Control and exposure responses were reported in Figures 1 and 3 and were adequate for the outcome of interest.
	Metric 23: Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in Figure 1.
Additional Comments:	This portion of the evaluation was on the effect of DBP in the cell density and morphology of the algae <i>C. pyrenoidosa</i> . Development and growth was selected as the outcome of interest. Supplemental material was not included in the Distiller download, but was found via the link in the article.		

Overall Quality Determination**High**

Study Citation:	Gu, S., Zheng, H., Xu, Q., Sun, C., Shi, M., Wang, Z., Li, F. (2017). Comparative toxicity of the plasticizer dibutyl phthalate to two freshwater algae. <i>Aquatic Toxicology</i> 191:122-130.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Chlorella pyrenoidosa</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)-Photosynthesis			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5433509			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Medium	The test substance was identified by name in the paper. The supplementary material had information regarding the structure and other physical characteristics.
	Metric 2:	Test Substance Source	Low	The source of the DBP was reported to be Sino-pharm Chemical Reagent Co. Ltd. in Nanjing, China, but it was not reported if it was analytically verified.
	Metric 3:	Test Substance Purity	High	The purity was reported to be 99% pure.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of a concurrent negative control.
	Metric 5:	Negative Control Response	High	The negative control response was reported in Figures 5 and 6 as well as in the supplemental material.
	Metric 6:	Randomized Allocation	Low	It was not reported how the algae was allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The study occurred in flasks. The test solution was prepared by creating a stock solution in which DBP was dissolved into acetone for a concentration of 100g/L. This was then diluted to the proper test concentrations using BG-11 algae growth medium.
	Metric 8:	Consistency of Exposure Administration	High	All exposures occurred in 500mL flasks for 96h using BG-11 growth media. All tests were started with the same initial algal cell count.
	Metric 9:	Measurement of Test Substance Concentration	Low	It was not reported if the test concentrations were measured.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration was reported to be 96h, which is typical of an algae growth study.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	There were 5 exposure levels, and the spacing was adequate to observe a dose response.
	Metric 12:	Testing at or Below Solubility Limit	Medium	Acetone was reported to be the vehicle solvent, but it was unclear if the negative control was a solvent control.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Both algal species were from the Freshwater Algae Culture Collection at the Chinese Academy of Sciences.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the organisms were acclimated at any point.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	<i>S. obliquus</i> initial concentrations were reported to be 2.02 x 10^6 cells/mL, and each concentration was performed in triplicate.
Domain 5: Outcome Assessment				
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Study Citation:	Gu, S., Zheng, H., Xu, Q., Sun, C., Shi, M., Wang, Z., Li, F. (2017). Comparative toxicity of the plasticizer dibutyl phthalate to two freshwater algae. Aquatic Toxicology 191:122-130.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Chlorella pyrenoidosa</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)-Photosynthesis			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5433509			
Domain		Metric	Rating	Comments
	Metric 16:	Adequacy of Test Conditions	High	The algae was cultured in BG-11 algal medium with a 12L:12D photoperiod at 24C and 8000lux.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—photosynthesis and oxidative stress.
	Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were described in the "statistical analysis" section of the paper.
	Metric 22:	Reporting of Data	High	Control and exposure responses were reported in Figures 5 and 6 as well as in the supplemental material. Results were adequate for the outcomes of interest.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in the figures.
Additional Comments:	This portion of the evaluation was on the effect of DBP on photosynthetic pigments and oxidative stress. Mechanistic outcomes were selected. Data for the photosynthesis outcomes in the paper were not provided because the Figure 4 in the paper is the incorrect figure. There are some data in the supplemental material regarding the photosynthesis outcomes. Supplemental material was not included in the Distiller download, but was found via the link in the article.			
Overall Quality Determination		High		

Study Citation:	Li, Z., Yi, X., Zhou, H., Chi, T., Li, W., Yang, K. (2020). Combined effect of polystyrene microplastics and dibutyl phthalate on the microalgae <i>Chlorella pyrenoidosa</i> . <i>Environmental Pollution</i> 257:113604.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Chlorella pyrenoidosa</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	6966450			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The DBP was identified by name only.	
	Metric 2: Test Substance Source	Low	DBP was purchased from Aladdin Bio-Chem Technology Co in Shanghai, but it was not reported to be analytically verified.	
	Metric 3: Test Substance Purity	High	Purity was reported at 99%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Negative controls and solvent controls are mentioned.	
	Metric 5: Negative Control Response	Low	The biological response of controls was not reported.	
	Metric 6: Randomized Allocation	Low	It was not reported if any methods were put into place to ensure random allocation of algal cells (ie mixing solutions etc.).	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	Exposures were carried out in 10 mL test vials in an incubator with stated light cycles and temperature control. DBP stock solution was prepared in methanol and stored out of direct sunlight.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	Low	Concentrations were only analyzed for the adsorption study; quantified analysis was not mentioned for the acute growth inhibition tests. Because exposure renewal was not carried out, concentrations may have varied throughout the test period.	
	Metric 10: Exposure Duration and Frequency	High	A 96-hour exposure was conducted to analyze acute toxicity.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	A wide range of exposure concentrations from 0.25 to 16 mg/L was included.	
	Metric 12: Testing at or Below Solubility Limit	High	Only the highest concentration studied (16 mg/L) was above the limit of solubility selected for DBP (11.3 mg/L).	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Medium	Organisms were cultured by the researchers, but few details were provided.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	Treatment conditions are stated to be the same as culture conditions.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	5 mL of culture solution containing algal cells at a density of 9.44E6 cells per mL was used. Tests were run in triplicate.	
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Study Citation:	Li, Z., Yi, X., Zhou, H., Chi, T., Li, W., Yang, K. (2020). Combined effect of polystyrene microplastics and dibutyl phthalate on the microalgae <i>Chlorella pyrenoidosa</i> . <i>Environmental Pollution</i> 257:113604.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Chlorella pyrenoidosa</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	6966450

Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Exposures were conducted in an incubator at standard conditions, the same as those used for culture, which appeared suitable for the organisms.
	Metric 17: Outcome Assessment Methodology	High	Cell density was measured using a SpectraMax microplate reader. Growth inhibition was the reported outcome of interest.
	Metric 18: Consistency of Outcome Assessment	High	The outcome was assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences in conditions across study groups.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure (e.g., infection) that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Dose-response analysis was conducted using GraphPad Prism software.
	Metric 22: Reporting of Data	Low	A summary of IC10 and IC50 values was reported with a 95% CI. However, data was not reported for each test concentration, and control results were not reported.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes, or unexpected outcomes were satisfactorily explained.

Additional Comments: This evaluation is for the growth inhibition test conducted on algae, *C. pyrenoidosa*, due to exposure to DBP.

Overall Quality Determination

High

Study Citation:	Chi, J., Li, B., Wang, Q. Y., Liu, H. (2007). Influence of nutrient level on biodegradation and bioconcentration of phthalate acid esters in <i>Chlorella vulgaris</i> . Journal of Environmental Science and Health, Part A: Toxic/Hazardous Substances & Environmental Engineering 42(2):179-183.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Chlorella vulgaris</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	679344			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The chemical was identified by name as dibutyl phthalate (DBP).
	Metric 2:	Test Substance Source	High	The test substance was obtained from Sigma.
	Metric 3:	Test Substance Purity	High	The test substance purity was reported as 99%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Uninformative	Study authors reported using an analytical control (no algae) and what appears to be a baseline control at time 0 hour. They did not use a negative control without test substance.
	Metric 5:	Negative Control Response	Medium	Baseline control responses were noted on graphs and in equations.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were consistently administered to test organisms.
	Metric 9:	Measurement of Test Substance Concentration	Medium	Concentrations were measured but only reported as log values on a graph.
	Metric 10:	Exposure Duration and Frequency	Low	The duration of exposure and/or exposure frequency differed significantly from typical study designs, and concentrations never reached steady state.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	Only one concentration was used.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of test replicates was not reported.
Domain 5: Outcome Assessment				
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Study Citation:	Chi, J., Li, B., Wang, Q. Y., Liu, H. (2007). Influence of nutrient level on biodegradation and bioconcentration of phthalate acid esters in <i>Chlorella vulgaris</i> . Journal of Environmental Science and Health, Part A: Toxic/Hazardous Substances & Environmental Engineering 42(2):179-183.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Chlorella vulgaris</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	679344			
Domain	Metric	Rating	Comments	
	Metric 16:	Adequacy of Test Conditions	Medium	Minor uncertainties or limitations were identified regarding organism environmental conditions as nutrient ratios were altered for each test.
	Metric 17:	Outcome Assessment Methodology	Medium	The outcome assessment methodology partially addressed the intended outcomes but the difference between biodegradation and bioconcentration wasn't clear.
	Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	Metric 20:	Outcomes Unrelated to Exposure	High	There were no differences among groups that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	N/A	Statistical analysis was not possible as only one test concentration was used.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment over time.
	Metric 23:	Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.
Additional Comments:	This was a bioconcentration/bioaccumulation study, with variable nitrogen and phosphorus ratios.			
Overall Quality Determination		Uninformative		

Study Citation:	Jonsson, S., Baun, A. (2003). Toxicity of mono- and diesters of o-phthalic esters to a crustacean, a green alga, and a bacterium. Environmental Toxicology and Chemistry 22(12):3037-3043.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Pseudokirchneriella subcapitata</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	789536			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
Metric 1:	Test Substance Identity	High	The test substance was identified definitively - nomenclature, CASRN, structure were reported.	
Metric 2:	Test Substance Source	High	DBP was purchased from Merck Eurolab (Stockholm, Sweden) but the test substance identity was NOT analytically verified by the performing laboratory.	
Metric 3:	Test Substance Purity	High	Percent purity was reported as >99%.	
Domain 2: Test Design				
Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
Metric 5:	Negative Control Response	High	The biological response of the control group was reported. The control group had a growth rate 1.6-1.8/ d during the 72 hours of incubation.	
Metric 6:	Randomized Allocation	Low	There were minor limitations in the allocation method that are unlikely to have a substantial impact on results. This was an algal study and reporting of random allocations are limited.	
Domain 3: Exposure Characterization				
Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations and/or minimize loss of test substance before and during the exposure.	
Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.	
Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were measured but not reported.	
Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type. An algal growth inhibition test was conducted for 72 hours.	
Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Uninformative	No information is provided on the exposure concentrations and the spacing of exposure levels.	
Metric 12:	Testing at or Below Solubility Limit	Low	Reporting omissions prevented determination of whether exposure concentrations exceeded the water solubility limit.	
Domain 4: Test Organism				
Metric 13:	Test Organism Characteristics	Low	The source of algae was not reported.	
Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized and/or whether pre-treatment conditions were the same for control and exposed groups.	

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Study Citation:	Jonsson, S., Baun, A. (2003). Toxicity of mono- and diesters of o-phthalic esters to a crustacean, a green alga, and a bacterium. Environmental Toxicology and Chemistry 22(12):3037-3043.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Pseudokirchneriella subcapitata</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	789536			
Domain		Metric	Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Test vessels were inoculated to achieve a cell density of 10^4 cells/ml. Replicates were not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions were conducive to maintenance of health. Typical control growth rates of 1.6-1.8/d were observed in controls.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology was reported. The cited reference (Mayer et al. 1997) has the detailed methodology.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups. Samples were taken from each test flask and the controls every 24hrs to determine growth rates.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study on outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Nonlinear regression analysis was conducted to estimate EC values and confidence intervals.
	Metric 22:	Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group. Only the EC 10 and EC 50 values were provided.
	Metric 23:	Explanation of Unexpected Outcomes	Low	There were no unexpected outcomes.
Additional Comments:	The exposure concentrations, spacing of exposure levels and control response were not reported. Measured concentrations were not reported. Growth rate data were not provided for each of the treatment groups and control. Only EC 10 and EC 50 values were reported.			

Overall Quality Determination**Uninformative**

Study Citation:	Gu, S., Zheng, H., Xu, Q., Sun, C., Shi, M., Wang, Z., Li, F. (2017). Comparative toxicity of the plasticizer dibutyl phthalate to two freshwater algae. <i>Aquatic Toxicology</i> 191:122-130.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Scenedesmus obliquus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)-Photosynthesis			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5433509			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
Metric 1:	Test Substance Identity	Medium	The test substance was identified by name in the paper. The supplementary material had information regarding the structure and other physical characteristics.	
Metric 2:	Test Substance Source	Low	The source of the DBP was reported to be Sino-pharm Chemical Reagent Co. Ltd. in Nanjing, China, but it was not reported if it was analytically verified.	
Metric 3:	Test Substance Purity	High	The purity was reported to be 99% pure.	
Domain 2: Test Design				
Metric 4:	Negative Controls	High	Study authors reported the use of a concurrent negative control.	
Metric 5:	Negative Control Response	High	The negative control response was reported in Figures 5 and 6 as well as in the supplemental material.	
Metric 6:	Randomized Allocation	Low	It was not reported how the algae was allocated into study groups.	
Domain 3: Exposure Characterization				
Metric 7:	Experimental System/Test Media Preparation	High	The study occurred in flasks. The test solution was prepared by creating a stock solution in which DBP was dissolved into acetone for a concentration of 100g/L. This was then diluted to the proper test concentrations using BG-11 algae growth medium.	
Metric 8:	Consistency of Exposure Administration	High	All exposures occurred in 500mL flasks for 96h using BG-11 growth media. All tests were started with the same initial algal cell count.	
Metric 9:	Measurement of Test Substance Concentration	Low	It was not reported if the test concentrations were measured.	
Metric 10:	Exposure Duration and Frequency	High	The exposure duration was reported to be 96h, which is typical of an algae growth study.	
Metric 11:	Number of Exposure Groups/Spacing of Exposure Levels	High	There were 5 exposure levels, and the spacing was adequate to observe a dose response.	
Metric 12:	Testing at or Below Solubility Limit	Medium	Acetone was reported to be the vehicle solvent, but it was unclear if the negative control was a solvent control.	
Domain 4: Test Organism				
Metric 13:	Test Organism Characteristics	High	Both algal species were from the Freshwater Algae Culture Collection at the Chinese Academy of Sciences.	
Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the organisms were acclimated at any point.	
Metric 15:	Number of Organisms and Replicates per Group	Medium	S. obliquus initial concentrations were reported to be 2.02 x 10^6 cells/mL, and each concentration was performed in triplicate.	

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Study Citation:	Gu, S., Zheng, H., Xu, Q., Sun, C., Shi, M., Wang, Z., Li, F. (2017). Comparative toxicity of the plasticizer dibutyl phthalate to two freshwater algae. <i>Aquatic Toxicology</i> 191:122-130.		
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)		
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Scenedesmus obliquus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)-Photosynthesis		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	5433509		
Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
Metric 16:	Adequacy of Test Conditions	High	The algae was cultured in BG-11 algal medium with a 12L:12D photoperiod at 24C and 8000lux.
Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—photosynthesis and oxidative stress.
Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	High	Statistical methods were described in the "statistical analysis" section of the paper.
Metric 22:	Reporting of Data	High	Control and exposure responses were reported in Figures 5 and 6 as well as in the supplemental material. Results were adequate for the outcomes of interest.
Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in the figures.
Additional Comments:	This portion of the evaluation was on the effect of DBP on photosynthetic pigments and oxidative stress. Mechanistic outcomes were selected. Data for the photosynthesis outcomes in the paper were not provided because the Figure 4 in the paper is the incorrect figure. There are some data in the supplemental material regarding the photosynthesis outcomes. Supplemental material was not included in the Distiller download, but was found via the link in the article.		

Overall Quality Determination**High**

Study Citation:	Gu, S., Zheng, H., Xu, Q., Sun, C., Shi, M., Wang, Z., Li, F. (2017). Comparative toxicity of the plasticizer dibutyl phthalate to two freshwater algae. <i>Aquatic Toxicology</i> 191:122-130.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Scenedesmus obliquus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5433509			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Medium	The test substance was identified by name in the paper. The supplementary material had information regarding the structure and other physical characteristics.
	Metric 2:	Test Substance Source	Low	The source of the DBP was reported to be Sino-pharm Chemical Reagent Co. Ltd. in Nanjing, China, but it was not reported if it was analytically verified.
	Metric 3:	Test Substance Purity	High	The purity was reported to be 99% pure.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of a concurrent negative control.
	Metric 5:	Negative Control Response	High	The negative control response was reported in Figures 1 and 2 and was adequate for the outcome of interest.
	Metric 6:	Randomized Allocation	Low	It was not reported how the algae was allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The study occurred in flasks. The test solution was prepared by creating a stock solution in which DBP was dissolved into acetone for a concentration of 100g/L. This was then diluted to the proper test concentrations using BG-11 algae growth medium.
	Metric 8:	Consistency of Exposure Administration	High	All exposures occurred in 500mL flasks for 96h using BG-11 growth media. All tests were started with the same initial algal cell count.
	Metric 9:	Measurement of Test Substance Concentration	Low	It was not reported if the test concentrations were measured.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration was reported to be 96h, which is typical of an algae growth study.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	There were five exposure levels, and the spacing was adequate to observe a dose response.
	Metric 12:	Testing at or Below Solubility Limit	Medium	Acetone was reported to be the vehicle solvent, but it was unclear if the negative control was a solvent control.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Both algal species were from the Freshwater Algae Culture Collection at the Chinese Academy of Sciences.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the organisms were acclimated at any point.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	<i>S. obliquus</i> initial concentrations were reported to be 2.02 x 10 ⁶ cells/mL, and each concentration was performed in triplicate.
Domain 5: Outcome Assessment				
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Study Citation:	Gu, S., Zheng, H., Xu, Q., Sun, C., Shi, M., Wang, Z., Li, F. (2017). Comparative toxicity of the plasticizer dibutyl phthalate to two freshwater algae. Aquatic Toxicology 191:122-130.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Scenedesmus obliquus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5433509			
Domain	Metric	Rating	Comments	
	Metric 16:	Adequacy of Test Conditions	High	The algae was cultured in BG-11 algal medium with a 12L:12D photoperiod at 24C and 8000lux.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest–cell density.
	Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. Samples were taken at 0, 24, 48, 72, and 96h and counted with a hemocytometer.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were described in the "statistical analysis" section of the paper.
	Metric 22:	Reporting of Data	High	Control and exposure responses were reported in Figures 1 and 2 and were adequate for the outcome of interest.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in Figure 1.
Additional Comments:	This portion of the evaluation was on the effect of DBP in the cell density and morphology of the algae S. obliquus. Development and growth was selected as the outcome of interest.Supplemental material was not included in the Distiller download, but was found via the link in the article.			
Overall Quality Determination		High		

Study Citation:	Huang, G. L., Sun, H. W., Song, Z. H. (1999). Interactions between dibutyl phthalate and aquatic organisms. Bulletin of Environmental Contamination and Toxicology 63(6):759-765.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Scenedesmus obliquus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5551982			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The source was not reported.	
	Metric 3: Test Substance Purity	Low	The purity and grade of the test substance were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	A concurrent negative control group was reported.	
	Metric 5: Negative Control Response	Low	A concurrent negative control group response was not reported.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.	
	Metric 8: Consistency of Exposure Administration	Low	Only general methods of exposure administration were reported, so it was difficult to determine if the assessment was carried out consistently.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	A standard test duration was used in the study.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Low	No information is provided on the spacing of exposure levels.	
	Metric 12: Testing at or Below Solubility Limit	Low	Reporting omissions prevented determination of whether exposure concentrations exceeded the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups.	
	Metric 15: Number of Organisms and Replicates per Group	Low	There were two replicates, and the initial number of organisms was adequate.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if they were adequate.	
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Study Citation:	Huang, G. L., Sun, H. W., Song, Z. H. (1999). Interactions between dibutyl phthalate and aquatic organisms. Bulletin of Environmental Contamination and Toxicology 63(6):759-765.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Scenedesmus obliquus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5551982			
Domain	Metric		Rating	Comments
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology was reported and adequate.
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22:	Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group.
	Metric 23:	Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability.
Additional Comments: None				

Overall Quality Determination**Low**

Study Citation:	Kuang, Q. J., Zhao, W. Y., Cheng, S. P. (2003). Toxicity of dibutyl phthalate to algae. Bulletin of Environmental Contamination and Toxicology 71(3):602-608.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Scenedesmus obliquus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332820			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The test substance was identified by name only.	
	Metric 2: Test Substance Source	Low	It was reported the DBP was manufactured in Shanghai, China but this is not enough information on the source. It was not reported if the DBP was analytically verified.	
	Metric 3: Test Substance Purity	High	The purity was reported to be 99%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	Low	Study authors reported the use of a concurrent negative control but it was unclear if the control was a solvent control.	
	Metric 5: Negative Control Response	High	The biological response of the negative control was adequate and reported in Table 1.	
	Metric 6: Randomized Allocation	Low	It was not reported how the algae was allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	Limited details were reported on the preparation of the test media and concentrations of test substance were not measured during the study. The concentration of the solvent (acetone) was not provided.	
	Metric 8: Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups. Exposures were conducted in 150mL glass flasks with HB-4 media. Temperatures and photoperiod remained consistent throughout the study, and the same algae concentration was used for all test concentrations.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if the test substance was measured at any point in the study.	
	Metric 10: Exposure Duration and Frequency	High	The exposure duration was reported to be 96h, which is typical of an acute toxicity test with algae.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	There were five exposure groups plus a control, and the spacing of the exposure levels was adequate for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	Low	It was reported that a vehicle solvent was used, but it was unclear if the control was a solvent control. The solvent concentration was not reported.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The source of the <i>S. obliquus</i> was reported to be the Department of Pathology, the Institute of Hydrobiology, Academia Sinica China.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the algae was acclimated to the test conditions prior to the start of the test.	

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Study Citation:	Kuang, Q. J., Zhao, W. Y., Cheng, S. P. (2003). Toxicity of dibutyl phthalate to algae. Bulletin of Environmental Contamination and Toxicology 71(3):602-608.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Scenedesmus obliquus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332820			
Domain	Metric	Rating	Comments	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The study authors reported that a concentration of 5-7 x 10^5cells/mL of algae was used in each test chamber. The test was conducted in triplicate.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	The test was conducted at 24C with a 14L:10D photo period. The algae was cultured in 150mL flasks with HB-4 algal media.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest–algal density-development/growth.	
	Metric 18: Consistency of Outcome Assessment	Medium	Details of the outcome assessment protocol were not clearly reported but outcomes were assessed consistently across study groups. Chlorophyll a was assessed using spectrophotometry at 650nm, and cell counts were done using a hemocytometer every two days, but methodology details were missing.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions. It was not reported if the algae was acclimated.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	Low	SGMI 1979 was used to determine EC50 values. However, it did not appear that statistics were performed on the other data, but independent statistical analysis may be conducted.	
	Metric 22: Reporting of Data	High	Data for the exposure response and the control response is reported in Table 1. Exposure related findings are also presented in Figures 1 and 2.	
	Metric 23: Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability.	
Additional Comments:	This portion of the evaluation was on the acute toxicity of DBP on the single celled algae S. obliquus. Algae concentrations were determined over the 96h test period. Development/growth was selected as the outcome of interest. Study authors also reported a chronic toxicity test was performed, but details regarding the study were not reported nor was any data reported, so an evaluation was not done.			

Overall Quality Determination**Medium**

Study Citation:	Huang, G. L., Sun, H. W., Song, Z. H. (1999). Interactions between dibutyl phthalate and aquatic organisms. Bulletin of Environmental Contamination and Toxicology 63(6):759-765.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Scenedesmus obliquus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5551982			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The source was not reported.	
	Metric 3: Test Substance Purity	Low	The purity and grade of the test substance were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	Low	A concurrent negative control group was reported as containing 5% FA.	
	Metric 5: Negative Control Response	Low	A concurrent negative control group response was not reported.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.	
	Metric 8: Consistency of Exposure Administration	Low	Only general methods of exposure administration were reported, so it was difficult to determine if the assessment was carried out consistently.	
	Metric 9: Measurement of Test Substance Concentration	Medium	Exposure concentrations were measured.	
	Metric 10: Exposure Duration and Frequency	Low	It was unclear if steady state had been reached during the study.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	N/A	Only one concentration was tested.	
	Metric 12: Testing at or Below Solubility Limit	High	The exposure concentration was below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups.	
	Metric 15: Number of Organisms and Replicates per Group	Low	The initial number of organisms was not reported, and the number replicates was not reported.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if they were adequate.	
	Metric 17: Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported.	
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Study Citation:	Huang, G. L., Sun, H. W., Song, Z. H. (1999). Interactions between dibutyl phthalate and aquatic organisms. Bulletin of Environmental Contamination and Toxicology 63(6):759-765.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Scenedesmus obliquus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5551982			
Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	N/A	Statistical analysis is typically not performed for these types of studies.
	Metric 22:	Reporting of Data	Low	Data for exposure-related findings were not shown for the control group.
	Metric 23:	Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability.
Additional Comments: None				

Overall Quality Determination**Low**

Study Citation:	Kuang, Q. J., Zhao, W. Y., Cheng, S. P. (2003). Toxicity of dibutyl phthalate to algae. Bulletin of Environmental Contamination and Toxicology 71(3):602-608.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Scenedesmus obliquus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Cell signaling/function			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332820			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The test substance was identified by name only.	
	Metric 2: Test Substance Source	Low	It was reported the DBP was manufactured in Shanghai, China but this is not enough information on the source. It was not reported if the DBP was analytically verified.	
	Metric 3: Test Substance Purity	High	The purity was reported to be 99%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	Low	Study authors reported the use of a concurrent negative control but it was unclear if the control was a solvent control.	
	Metric 5: Negative Control Response	Low	The biological response of the negative control was mentioned in the text in comparison to treatments.	
	Metric 6: Randomized Allocation	Low	It was not reported how the algae was allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	Limited details were reported on the preparation of the test media and concentrations of test substance were not measured during the study. The concentration of the solvent (acetone) was not provided.	
	Metric 8: Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups. Exposures were conducted in 150mL glass flasks with HB-4 media. Temperatures and photoperiod remained consistent throughout the study, and the same algae concentration was used for all test concentrations.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if the test substance was measured at any point in the study.	
	Metric 10: Exposure Duration and Frequency	High	The exposure duration was reported to be 4 weeks for the chronic study.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	There were 5 exposure groups plus a control, and the spacing of the exposure levels was adequate for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	Low	It was reported that a vehicle solvent was used, but it was unclear if the control was a solvent control. The solvent concentration was not reported.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The source of the <i>S. obliquus</i> was reported to be the Department of Pathology, the Institute of Hydrobiology, Academia Sinica China.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the algae was acclimated to the test conditions prior to the start of the test.	

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Study Citation:	Kuang, Q. J., Zhao, W. Y., Cheng, S. P. (2003). Toxicity of dibutyl phthalate to algae. Bulletin of Environmental Contamination and Toxicology 71(3):602-608.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Scenedesmus obliquus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Cell signaling/function			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332820			
Domain	Metric	Rating	Comments	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The study authors reported that a concentration of 5-7 x 10^5cells/mL of algae was used in each test chamber. The test was conducted in triplicate.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	The test was conducted at 24C with a 14L:10D photo period. The algae was cultured in 150mL flasks with HB-4 algal media.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest–algal density-development/growth.	
	Metric 18: Consistency of Outcome Assessment	Medium	Details of the outcome assessment protocol were not clearly reported but outcomes were assessed consistently across study groups. Chlorophyll a was assessed using spectrophotometry at 650nm, and cell counts were done using a hemocytometer every 2 days but methodology details were missing.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions. It was not reported if the algae was acclimated.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	Low	SGMI 1979 was used to determine EC50 values. However, it did not appear that statistics were performed on the other data, but independent statistical analysis may be conducted.	
	Metric 22: Reporting of Data	Low	Results for the chronic exposure were reported in the text only.	
	Metric 23: Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability.	
Additional Comments:	This portion of the evaluation is for the cellular results reported in the text for the chronic exposure. Cell wall thickening was reported along with cell inclusions. Little details were provided on the chronic portion of the study. This evaluation was for the 4 week chronic exposure.			

Overall Quality Determination**Medium**

Study Citation:	Kuang, Q. J., Zhao, W. Y., Cheng, S. P. (2003). Toxicity of dibutyl phthalate to algae. Bulletin of Environmental Contamination and Toxicology 71(3):602-608.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Scenedesmus obliquus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332820			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The test substance was identified by name only.	
	Metric 2: Test Substance Source	Low	It was reported the DBP was manufactured in Shanghai, China but this is not enough information on the source. It was not reported if the DBP was analytically verified.	
	Metric 3: Test Substance Purity	High	The purity was reported to be 99%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	Low	Study authors reported the use of a concurrent negative control but it was unclear if the control was a solvent control.	
	Metric 5: Negative Control Response	Low	The biological response of the negative control was mentioned in the text in comparison to treatments.	
	Metric 6: Randomized Allocation	Low	It was not reported how the algae was allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	Limited details were reported on the preparation of the test media and concentrations of test substance were not measured during the study. The concentration of the solvent (acetone) was not provided.	
	Metric 8: Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups. Exposures were conducted in 150mL glass flasks with HB-4 media. Temperatures and photoperiod remained consistent throughout the study, and the same algae concentration was used for all test concentrations.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if the test substance was measured at any point in the study.	
	Metric 10: Exposure Duration and Frequency	High	The exposure duration was reported to be four weeks for the chronic study.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	There were five exposure groups plus a control, and the spacing of the exposure levels was adequate for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	Low	It was reported that a vehicle solvent was used, but it was unclear if the control was a solvent control. The solvent concentration was not reported.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The source of the <i>S. obliquus</i> was reported to be the Department of Pathology, the Institute of Hydrobiology, Academia Sinica China.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the algae was acclimated to the test conditions prior to the start of the test.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The study authors reported that a concentration of 5-7 x 10^5cells/mL of algae was used in each test chamber. The test was conducted in triplicate.	
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Study Citation:	Kuang, Q. J., Zhao, W. Y., Cheng, S. P. (2003). Toxicity of dibutyl phthalate to algae. Bulletin of Environmental Contamination and Toxicology 71(3):602-608.		
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days		
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Scenedesmus obliquus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Development/Growth		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	1332820		
Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
Metric 16:	Adequacy of Test Conditions	High	The test was conducted at 24C with a 14L:10D photo period. The algae was cultured in 150mL flasks with HB-4 algal media.
Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest–algal density-development/growth.
Metric 18:	Consistency of Outcome Assessment	Medium	Details of the outcome assessment protocol were not clearly reported but outcomes were assessed consistently across study groups. Chlorophyll a was assessed using spectrophotometry at 650nm, and cell counts were done using a hemocytometer every two days, but methodology details were missing.
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions. It was not reported if the algae was acclimated.
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	Low	SGMI 1979 was used to determine EC50 values. However, it did not appear that statistics were performed on the other data, but independent statistical analysis may be conducted.
Metric 22:	Reporting of Data	Low	Results for the chronic exposure were reported in the text only.
Metric 23:	Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability.
Additional Comments:	This portion of the evaluation was on the acute toxicity of DBP on the single celled algae S. obliquus. Algae concentrations were determined over the 96h test period. Development/growth was selected as the outcome of interest. Study authors also reported a chronic toxicity test was performed, but details regarding the study were not reported nor was any data reported, so an evaluation was not done. This evaluation was for the four week chronic exposure.		

Overall Quality Determination**Medium**

Study Citation:	Cunha, C., Paulo, J., Faria, M., Kaufmann, M., Cordeiro, N. (2019). Ecotoxicological and biochemical effects of environmental concentrations of the plastic-bond pollutant dibutyl phthalate on <i>Scenedesmus</i> sp. <i>Aquatic Toxicology</i> 215:105281.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Scenedesmus</i> sp.; BEA0579B; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	6967432			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	DBP was identified by name only.	
	Metric 2: Test Substance Source	High	The source was reported as Riedel-de Haen, and the DBP was analytically verified using UHPLC as noted in Section 2.3 of the paper.	
	Metric 3: Test Substance Purity	High	The purity was reported as 99%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Negative controls and solvent controls were reported.	
	Metric 5: Negative Control Response	High	The response of the controls (negative and vehicle) were adequate.	
	Metric 6: Randomized Allocation	Low	It was not reported if any measures were taken to randomly allocate the algae (ie. mixing etc.).	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	Waris-H medium was described in detail, and cultures and experimental solutions were maintained in glass flasks. Flasks were topped up with algae solution to ensure that cell density was consistent before the exposure commenced. The test solution was prepared by dissolving DBP in methanol with the maximum amount of solvent set to 0.05% for all conditions and replicates.	
	Metric 8: Consistency of Exposure Administration	High	The exposure was consistent across study groups and was reported adequately.	
	Metric 9: Measurement of Test Substance Concentration	Low	Experimental concentrations were not measured, but a separate DBP stability test was conducted without microalgae. In this test, DBP concentrations were reduced by 32.4% after 48h in solution. However, this correction factor was not applied to the experimental concentrations by the researchers. Therefore, experimental concentrations may be expected to fall considerably from nominal values during the 96-h exposure period.	
	Metric 10: Exposure Duration and Frequency	High	A static 96-hour exposure was conducted, which is appropriate for an acute growth inhibition test with algae.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Four concentrations were used with spacing adequate to characterize a dose-response; however, for both photosynthetic pigment and extracellular carbohydrate production, a non-monotonic dose-response was obtained with lower concentrations, showing a higher effect in some cases than others. More concentrations at a higher dose could have potentially helped to more clearly parameterize the dose-response curve.	
	Metric 12: Testing at or Below Solubility Limit	High	Test concentrations were below the solubility limit of 11.2 mg/L for DBP.	
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Study Citation:	Cunha, C., Paulo, J., Faria, M., Kaufmann, M., Cordeiro, N. (2019). Ecotoxicological and biochemical effects of environmental concentrations of the plastic-bond pollutant dibutyl phthalate on <i>Scenedesmus</i> sp. <i>Aquatic Toxicology</i> 215:105281.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Scenedesmus</i> sp.; BEA0579B; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	6967432

Domain	Metric	Rating	Comments
Domain 4: Test Organism			
Metric 13:	Test Organism Characteristics	High	<i>Scenedesmus</i> sp. (BEA0579B) was obtained from the Spanish Bank of Algae (BEA) of the University of Las Palmas de Gran Canaria (Spain).
Metric 14:	Acclimatization and Pretreatment Conditions	High	Cultures were acclimated to experimental conditions for 96h before inoculation with DBP.
Metric 15:	Number of Organisms and Replicates per Group	Medium	Cell density was 1.6x10E5 cells/mL, and experiments were conducted in triplicate.
Domain 5: Outcome Assessment			
Metric 16:	Adequacy of Test Conditions	High	The culture and experimental groups were maintained at standard conditions (25C, 14/10 photoperiod) in a growth chamber.
Metric 17:	Outcome Assessment Methodology	High	Cell density and growth was observed using a Neubauer Improved counting chamber and an optical microscope.
Metric 18:	Consistency of Outcome Assessment	High	Outcomes were evaluated consistently among study groups.
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	High	Statistical analysis was conducted using SPSS and one-way ANOVA.
Metric 22:	Reporting of Data	Medium	Outcomes were reported for all groups (Fig.2). Although, only a 48-h EC50 was reported, and the reported LOEC for growth inhibition, 1 µg/L, does not match the growth curves shown in Fig.2 in which the 0.02 µg/L group is shown to have a significant effect on growth at 48, 72, and 96 hours.
Metric 23:	Explanation of Unexpected Outcomes	Medium	The paper does not address the non-monotonicity of dose responses at 96-h, in which growth inhibition was not significantly different among exposure groups several orders of magnitude in difference (0.02, 1, and 100 µg/L), and which shows slightly higher mean inhibition among the 0.02 µg/L exposures than the 1 µg/L exposures. Further, this study finds toxicity to <i>Scenedesmus</i> algae at concentrations significantly lower than other studies as noted in the discussion, without adequate explanation.
Additional Comments: This form is for the 96-hour growth inhibition outcome.			

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Study Citation:	Cunha, C., Paulo, J., Faria, M., Kaufmann, M., Cordeiro, N. (2019). Ecotoxicological and biochemical effects of environmental concentrations of the plastic-bond pollutant dibutyl phthalate on <i>Scenedesmus</i> sp. <i>Aquatic Toxicology</i> 215:105281.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Scenedesmus</i> sp.; BEA0579B; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	6967432

Domain	Metric	Rating	Comments
Overall Quality Determination		High	

Study Citation:	Cunha, C., Paulo, J., Faria, M., Kaufmann, M., Cordeiro, N. (2019). Ecotoxicological and biochemical effects of environmental concentrations of the plastic-bond pollutant dibutyl phthalate on <i>Scenedesmus</i> sp. <i>Aquatic Toxicology</i> 215:105281.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Scenedesmus</i> sp.; BEA0579B; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Nutritional & Metabolic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	6967432			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	DBP was identified by name only.	
	Metric 2: Test Substance Source	High	The source was reported as Riedel-de Haen, and the DBP was analytically verified using UHPLC as noted in Section 2.3 of the paper.	
	Metric 3: Test Substance Purity	High	The purity was reported as 99%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Negative controls and solvent controls were reported.	
	Metric 5: Negative Control Response	High	The response of the controls (negative and vehicle) were adequate.	
	Metric 6: Randomized Allocation	Low	It was not reported if any measures were taken to randomly allocate the algae (ie. mixing etc.).	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	Waris-H medium was described in detail, and cultures and experimental solutions were maintained in glass flasks. Flasks were topped up with algae solution to ensure that cell density was consistent before the exposure commenced. The test solution was prepared by dissolving DBP in methanol with the maximum amount of solvent set to 0.05% for all conditions and replicates.	
	Metric 8: Consistency of Exposure Administration	High	The exposure was consistent across study groups and was reported adequately.	
	Metric 9: Measurement of Test Substance Concentration	Low	Experimental concentrations were not measured, but a separate DBP stability test was conducted without microalgae. In this test, DBP concentrations were reduced by 32.4% after 48h in solution. However, this correction factor was not applied to the experimental concentrations by the researchers. Therefore, experimental concentrations may be expected to fall considerably from nominal values during the 96-h exposure period.	
	Metric 10: Exposure Duration and Frequency	High	A static 96-hour exposure was conducted, which is appropriate for an acute growth inhibition test with algae.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Four concentrations were used with spacing adequate to characterize a dose-response; however, for both photosynthetic pigment and extracellular carbohydrate production, a non-monotonic dose-response was obtained with lower concentrations, showing a higher effect in some cases than others. More concentrations at a higher dose could have potentially helped to more clearly parameterize the dose-response curve.	
	Metric 12: Testing at or Below Solubility Limit	High	Test concentrations were below the solubility limit of 11.2 mg/L for DBP.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	Scenedesmus sp. (BEA0579B) was obtained from the Spanish Bank of Algae (BEA) of the University of Las Palmas de Gran Canaria (Spain).	
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Study Citation:	Cunha, C., Paulo, J., Faria, M., Kaufmann, M., Cordeiro, N. (2019). Ecotoxicological and biochemical effects of environmental concentrations of the plastic-bond pollutant dibutyl phthalate on <i>Scenedesmus</i> sp. <i>Aquatic Toxicology</i> 215:105281.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Scenedesmus</i> sp.; BEA0579B; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Nutritional & Metabolic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	6967432			
Domain	Metric	Rating	Comments	
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Cultures were acclimated to experimental conditions for 96h before inoculation with DBP.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Cell density was 1.6x10E5 cells/mL, and experiments were conducted in triplicate.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	The culture and experimental groups were maintained at standard conditions (25C, 14/10 photoperiod) in a growth chamber.
	Metric 17:	Outcome Assessment Methodology	High	Photosynthetic pigments were measured based on the Lichtenthaler (1987) method, cited and described in the test as using a spectrophotometric method. Extracellular carbohydrates were measured using the phenol-sulfuric acid method, also using a spectrophotometer. Extracellular protein was also assessed by measuring absorbance at 750nm.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were evaluated consistently among study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical analysis was conducted using SPSS and one-way ANOVA.
	Metric 22:	Reporting of Data	High	Outcomes were reported for all groups (Fig.4-6).
	Metric 23:	Explanation of Unexpected Outcomes	Medium	The paper does not address the non-monotonicity of dose responses at 96-h, in which extracellular carbohydrate inhibition was not significantly different among exposure groups several orders of magnitude in difference (0.02, 1, and 100 µg/L), and which shows slightly higher mean inhibition among the 0.02 µg/L exposures than the 1 µg/L exposures. Non-monotonicity was also observed in the photosynthetic pigments test. Further, this study finds toxicity to <i>Scenedesmus</i> algae at concentrations significantly lower than other studies, as noted in the discussion, without adequate explanation.
Additional Comments:	This form is for the 96-hour cellular metabolic outcomes including extracellular carbohydrates and extracellular proteins.			

Overall Quality Determination**High**

Study Citation:	Cunha, C., Paulo, J., Faria, M., Kaufmann, M., Cordeiro, N. (2019). Ecotoxicological and biochemical effects of environmental concentrations of the plastic-bond pollutant dibutyl phthalate on <i>Scenedesmus</i> sp. <i>Aquatic Toxicology</i> 215:105281.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Scenedesmus</i> sp.; BEA0579B; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Photosynthesis
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	6967432

Domain	Metric	Rating	Comments
Domain 1: Test Substance			
	Metric 1: Test Substance Identity	Low	DBP was identified by name only.
	Metric 2: Test Substance Source	High	The source was reported as Riedel-de Haen, and the DBP was analytically verified using UHPLC as noted in Section 2.3 of the paper.
	Metric 3: Test Substance Purity	High	The purity was reported as 99%.
Domain 2: Test Design			
	Metric 4: Negative Controls	High	Negative controls and solvent controls were reported.
	Metric 5: Negative Control Response	High	The response of the controls (negative and vehicle) were adequate.
	Metric 6: Randomized Allocation	Low	It was not reported if any measures were taken to randomly allocate the algae (ie. mixing etc.).
Domain 3: Exposure Characterization			
	Metric 7: Experimental System/Test Media Preparation	High	Waris-H medium was described in detail, and cultures and experimental solutions were maintained in glass flasks. Flasks were topped up with algae solution to ensure that cell density was consistent before the exposure commenced. The test solution was prepared by dissolving DBP in methanol with the maximum amount of solvent set to 0.05% for all conditions and replicates.
	Metric 8: Consistency of Exposure Administration	High	The exposure was consistent across study groups and was reported adequately.
	Metric 9: Measurement of Test Substance Concentration	Low	Experimental concentrations were not measured, but a separate DBP stability test was conducted without microalgae. In this test, DBP concentrations were reduced by 32.4% after 48h in solution. However, this correction factor was not applied to the experimental concentrations by the researchers. Therefore, experimental concentrations may be expected to fall considerably from nominal values during the 96-h exposure period.
	Metric 10: Exposure Duration and Frequency	High	A static 96-hour exposure was conducted, which is appropriate for an acute growth inhibition test with algae.
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Medium	Four concentrations were used with spacing adequate to characterize dose-response, however; for both photosynthetic pigment and extracellular carbohydrate production a non-monotonic dose-response was obtained with lower concentrations showing a higher effect in some cases than higher. More concentrations at a higher dose could have potentially helped to more clearly parameterize the dose-response curve.
	Metric 12: Testing at or Below Solubility Limit	High	Test concentrations were below the solubility limit of 11.2 mg/L for DBP.
Domain 4: Test Organism			
	Metric 13: Test Organism Characteristics	High	<i>Scenedesmus</i> sp. (BEA0579B) was obtained from the Spanish Bank of Algae (BEA) of the University of Las Palmas de Gran Canaria (Spain).

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Study Citation:	Cunha, C., Paulo, J., Faria, M., Kaufmann, M., Cordeiro, N. (2019). Ecotoxicological and biochemical effects of environmental concentrations of the plastic-bond pollutant dibutyl phthalate on <i>Scenedesmus</i> sp. <i>Aquatic Toxicology</i> 215:105281.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Scenedesmus</i> sp.; BEA0579B; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Photosynthesis			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	6967432			
Domain	Metric	Rating	Comments	
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Cultures were acclimated to experimental conditions for 96h before inoculation with DBP.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Cell density was 1.6x10E5 cells/mL, and experiments were conducted in triplicate.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	The culture and experimental groups were maintained at standard conditions (25C, 14/10 photoperiod) in a growth chamber.
	Metric 17:	Outcome Assessment Methodology	High	Photosynthetic pigments were measured based on the Lichtenthaler (1987) method, cited and described in the test as using a spectrophotometric method. Extracellular carbohydrates were measured using the phenol-sulfuric acid method, also using a spectrophotometer. Extracellular protein was also assessed by measuring absorbance at 750nm.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were evaluated consistently among study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical analysis was conducted using SPSS and one-way ANOVA.
	Metric 22:	Reporting of Data	High	Outcomes were reported for all groups in Figure 4.
	Metric 23:	Explanation of Unexpected Outcomes	Medium	The paper does not address the non-monotonicity of dose responses at 96-h, in which extracellular carbohydrate inhibition was not significantly different among exposure groups several orders of magnitude in difference (0.02, 1, and 100 µg/L), and which shows slightly higher mean inhibition among the 0.02 µg/L exposures than the 1 µg/L exposures. Non-monotonicity was also observed in the photosynthetic pigments test. Further, this study finds toxicity to <i>Scenedesmus</i> algae at concentrations significantly lower than other studies, as noted in the discussion, without adequate explanation.
Additional Comments:	This form is for the 96-hour photosynthetic outcomes including measurements of chlorophyll a, chlorophyll b, and carotenoids.			

Overall Quality Determination**High**

Study Citation:	Adachi, A., Asa, K., Okano, T. (2006). Efficiency of rice bran for removal of di-n-butyl phthalate and its effect on the growth inhibition of <i>Selenastrum capricornutum</i> by di-n-butyl phthalate. Bulletin of Environmental Contamination and Toxicology 76(5):877-882.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Selenastrum capricornutum</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1323217			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The chemical was identified by name only.
	Metric 2:	Test Substance Source	High	The source was reported as Wake Pure Chemical Industries (Amagasaki, Japan), and verified by gas chromatography.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of the test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using a concurrent negative control group.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.
	Metric 8:	Consistency of Exposure Administration	Medium	A one-time dose was used. Exposures were administered consistently across study groups, but few details were provided.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured by gas chromatography, and given in Figure 1.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Medium	No differences were reported in handling the algae.
	Metric 15:	Number of Organisms and Replicates per Group	Low	An adequate number of algae were used, but replicates weren't reported.
Domain 5: Outcome Assessment				
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Study Citation:	Adachi, A., Asa, K., Okano, T. (2006). Efficiency of rice bran for removal of di-n-butyl phthalate and its effect on the growth inhibition of <i>Selenastrum capricornutum</i> by di-n-butyl phthalate. Bulletin of Environmental Contamination and Toxicology 76(5):877-882.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Selenastrum capricornutum</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	1323217

Domain	Metric	Rating	Comments
	Metric 16: Adequacy of Test Conditions	Medium	Environmental conditions of the test system were most likely conducive to maintenance of organism health but actual measured condition values for control and exposed vessels was not reported.
	Metric 17: Outcome Assessment Methodology	Medium	The outcome assessment methodology reported the intended outcome of interest, but few details were reported.
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups, and rice bran was in all treatments.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This evaluation is for the results with rice bran.

Overall Quality Determination

Medium

Study Citation:	Adachi, A., Asa, K., Okano, T. (2006). Efficiency of rice bran for removal of di-n-butyl phthalate and its effect on the growth inhibition of <i>Selenastrum capricornutum</i> by di-n-butyl phthalate. Bulletin of Environmental Contamination and Toxicology 76(5):877-882.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Selenastrum capricornutum</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1323217			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	High	The source was reported as Wake Pure Chemical Industries (Amagasaki, Japan), and verified by gas chromatography.	
	Metric 3: Test Substance Purity	Low	Purity and/or grade of the test substance were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.	
	Metric 8: Consistency of Exposure Administration	Medium	A one-time dose was used. Exposures were administered consistently across study groups, but few details were provided.	
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured by gas chromatography, and given in Figure 1.	
	Metric 10: Exposure Duration and Frequency	High	The exposure was conducted for 96 hours.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	Medium	No differences were reported in handling the algae.	
	Metric 15: Number of Organisms and Replicates per Group	Low	An adequate number of algae were used, but replicates weren't reported.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Medium	Environmental conditions of the test system were most likely conducive to maintenance of organism health but actual measured condition values for control and exposed vessels was not reported.	
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Study Citation:	Adachi, A., Asa, K., Okano, T. (2006). Efficiency of rice bran for removal of di-n-butyl phthalate and its effect on the growth inhibition of <i>Selenastrum capricornutum</i> by di-n-butyl phthalate. Bulletin of Environmental Contamination and Toxicology 76(5):877-882.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Selenastrum capricornutum</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	1323217

Domain	Metric	Rating	Comments
	Metric 17: Outcome Assessment Methodology	Medium	The outcome assessment methodology reported the intended outcome of interest, but few details were reported.
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: None

Overall Quality Determination**Medium**

Study Citation:	Adams, W. J., Biddinger, G. R., Robillard, K. A., Gorsuch, J. W. (1995). A summary of the acute toxicity of 14 phthalate esters to representative aquatic organisms. Environmental Toxicology and Chemistry 14(9):1569-1574.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Selenastrum capricornutum</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1321996; Linked HERO ID(s): 1321996, 1316224			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The test substance nomenclature was reported without a CASRN.
	Metric 2:	Test Substance Source	Low	The source was reported as provided by a manufacturer from commercially available batches. Manufacture name and batch number were not provided. No analytical data was reported.
	Metric 3:	Test Substance Purity	High	The test substance was at least 95% pure.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	A negative control was reported.
	Metric 5:	Negative Control Response	High	The control response was acceptable.
	Metric 6:	Randomized Allocation	Low	An allocation method was not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The experimental system was well described. However, headspace or measures to prevent volatilization were not reported.
	Metric 8:	Consistency of Exposure Administration	High	Exposure administration was consistent across groups.
	Metric 9:	Measurement of Test Substance Concentration	Medium	Sample extracts were analyzed by gas chromatography at the start and end of the test. In static studies, final test concentrations frequently were 50% of the initial concentrations. Loss of the phthalate esters was thought to be principally due to adsorption to the test vessels.
	Metric 10:	Exposure Duration and Frequency	High	Duration and frequency of the exposure were appropriate for the test.
	Metric 11:	Number of Exposure Groups/Spacing of Exposure Levels	High	Exposure levels were appropriate. A range finding test was performed.
	Metric 12:	Testing at or Below Solubility Limit	High	The test was performed at or below water solubility.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	A source was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Appropriate acclimation for the test was reported.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Each test was performed using duplicate test concentrations with 10 organisms per test vessel.
Domain 5: Outcome Assessment				
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Study Citation:	Adams, W. J., Biddinger, G. R., Robillard, K. A., Gorsuch, J. W. (1995). A summary of the acute toxicity of 14 phthalate esters to representative aquatic organisms. Environmental Toxicology and Chemistry 14(9):1569-1574.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Selenastrum capricornutum</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	1321996; Linked HERO ID(s): 1321996, 1316224

Domain	Metric	Rating	Comments
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions were appropriate for the test.
	Metric 17: Outcome Assessment Methodology	High	Intended outcomes were reported.
	Metric 18: Consistency of Outcome Assessment	High	Outcome assessment was consistent across groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	Environmental conditions were consistent across groups.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There were no reported differences between groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were performed and described.
	Metric 22: Reporting of Data	Medium	Only treatment endpoints were reported.
	Metric 23: Explanation of Unexpected Outcomes	High	No unexpected outcomes were reported.
Additional Comments: None			

Overall Quality Determination**High**

Study Citation:	Bionomics,, Springborn (1984). FYI Submission: Toxicity of fourteen phthalate esters to the freshwater green alga <i>Selenastrum capricornutum</i> .			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Selenastrum capricornutum</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1316196			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The chemical was identified by name and CASRN.	
	Metric 2: Test Substance Source	Low	The source of the chemical was not included in the technical report.	
	Metric 3: Test Substance Purity	Low	Purity or grade were not included in the technical report.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Negative controls were included in the study.	
	Metric 5: Negative Control Response	High	There was an adequate response in the negative controls.	
	Metric 6: Randomized Allocation	Medium	Allocation of algae for experiments was not described in the study.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The study followed OECD 201 (1981) guidelines and included any deviations.	
	Metric 8: Consistency of Exposure Administration	High	The exposure administration was reported as consistent.	
	Metric 9: Measurement of Test Substance Concentration	High	Appendix A includes the analytical measurement of the chemical, gas-liquid chromatography with electron capture detection. Concentrations measured throughout the 10 days, as well as initial and final concentrations can be found in the text.	
	Metric 10: Exposure Duration and Frequency	High	The 10-day exposure was adequate for the study.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Medium	An appropriate number of exposure groups was used.	
	Metric 12: Testing at or Below Solubility Limit	High	Concentrations with responses were below the solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source and details about the algae were limited.	
	Metric 14: Acclimatization and Pretreatment Conditions	Medium	Acclimatization details were limited.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The study followed OECD 201 (1981) guidelines using adequate numbers of organisms.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Test conditions were reported.	
	Metric 17: Outcome Assessment Methodology	High	The outcome was described in detail in the results.	
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Study Citation:	Bionomics,, Springborn (1984). FYI Submission: Toxicity of fourteen phthalate esters to the freshwater green alga <i>Selenastrum capricornutum</i> .			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Selenastrum capricornutum</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1316196			
Domain	Metric	Rating	Comments	
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were reported consistently.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	No differences were reported among the study groups.	
	Metric 20: Outcomes Unrelated to Exposure	High	Outcomes unrelated to exposure were not reported.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistics used include moving average angle analysis, probit analysis, and binomial probability.	
	Metric 22: Reporting of Data	Medium	Data was reported for growth.	
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.	
Additional Comments:	None			
Overall Quality Determination		High		

Study Citation:	Melin, C., Egneus, H. (1983). Effects of di-n-butyl phthalate on growth and photosynthesis in algae and on isolated organelles from higher plants. <i>Physiologia Plantarum</i> 59(3):461-466.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Selenastrum capricornutum</i> ; CCAP strain 278/4; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1333016			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The DBP was identified by name only.	
	Metric 2: Test Substance Source	Low	The source of the DBP was not reported.	
	Metric 3: Test Substance Purity	Low	The purity/grade of the DBP was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of a negative control, which was a solvent control containing ethanol.	
	Metric 5: Negative Control Response	High	The negative control response was reported in Figure 1.	
	Metric 6: Randomized Allocation	Low	It was not reported how the algae was allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The authors provided limited details on the preparation of the DBP test solution.	
	Metric 8: Consistency of Exposure Administration	Low	Details regarding the exposure administration were limited. Study authors reported the temperature and test volumes though.	
	Metric 9: Measurement of Test Substance Concentration	Low	Study authors did not report if the test concentrations were measured at any point in the study.	
	Metric 10: Exposure Duration and Frequency	Low	Study authors did not explicitly state the duration of the study, but Figure 1 indicates it was seven days.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Medium	The exposure levels were provided in Figure 1. The number of exposure groups was adequate, but different spacing (especially between 10 ⁻⁵ and 10 ⁻⁴ M may have yielded more in-depth results.	
	Metric 12: Testing at or Below Solubility Limit	High	Study authors reported the use of a vehicle solvent.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the algae was not reported.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if any acclimation occurred.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	It was reported that the initial algal density was 0.1ug chlorophyll/mL. There were three culture flasks per concentration.	
Domain 5: Outcome Assessment				
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Study Citation:	Melin, C., Egneus, H. (1983). Effects of di-n-butyl phthalate on growth and photosynthesis in algae and on isolated organelles from higher plants. Physiologia Plantarum 59(3):461-466.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Selenastrum capricornutum</i> ; CCAP strain 278/4; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1333016			
Domain	Metric	Rating	Comments	
	Metric 16:	Adequacy of Test Conditions	Medium	S. capricornutum was grown in 10% (v/v) Z-8 medium according to Kotain 1972. Cultures were grown at 20 C with continuous illumination.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest–algal growth.
	Metric 18:	Consistency of Outcome Assessment	Low	It was reported that algae growth was determined by measuring changes in transmittance at 729nm, but methods and instruments used were not reported.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. It was not reported if the algae was acclimated.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Growth curve determination and IC50 value determination were described in the text.
	Metric 22:	Reporting of Data	High	Data for exposure concentrations and for the control response was reported in Figure 1 and was adequate.
	Metric 23:	Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability.
Additional Comments:	This portion of the evaluation is on the effect of DBP on the growth of algae S. capricornutum. Development/growth was selected as the outcome of interest.			

Overall Quality Determination**Medium**

Study Citation:	Melin, C., Egneus, H. (1983). Effects of di-n-butyl phthalate on growth and photosynthesis in algae and on isolated organelles from higher plants. <i>Physiologia Plantarum</i> 59(3):461-466.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Hordeum vulgare</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Photosynthesis			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1333016			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The DBP was identified by name only.	
	Metric 2: Test Substance Source	Low	The source of the DBP was not reported.	
	Metric 3: Test Substance Purity	Low	The purity/grade of the DBP was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of a negative control, which was a solvent control containing ethanol.	
	Metric 5: Negative Control Response	High	The negative control response was mentioned in text under Figure 3.	
	Metric 6: Randomized Allocation	Low	It was not reported how the protoplasts were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The authors provided limited details on the preparation of the DBP test solution.	
	Metric 8: Consistency of Exposure Administration	Low	Details regarding the exposure administration were limited. Study authors reported the temperature and test volumes though.	
	Metric 9: Measurement of Test Substance Concentration	Low	Study authors did not report if the test concentrations were measured at any point in the study.	
	Metric 10: Exposure Duration and Frequency	High	Exposure duration was described and seemed adequate.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	Exposure groups seemed fine and adequate results were obtained.	
	Metric 12: Testing at or Below Solubility Limit	High	Study authors reported the use of a vehicle solvent.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of barley protoplasts was not reported.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if any acclimation occurred.	
	Metric 15: Number of Organisms and Replicates per Group	Low	The protoplast concentration was given but the number of replicates was not.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Test conditions seemed appropriate and were described on page 462 of the paper.	
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Study Citation:	Melin, C., Egneus, H. (1983). Effects of di-n-butyl phthalate on growth and photosynthesis in algae and on isolated organelles from higher plants. <i>Physiologia Plantarum</i> 59(3):461-466.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Hordeum vulgare</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Photosynthesis
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	1333016

Domain	Metric	Rating	Comments
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—photosynthesis/CO2 fixation in protoplasts from barley.
	Metric 18: Consistency of Outcome Assessment	High	The methodology was consistent across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. It was not reported if acclimation occurred.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	IC50 value determination was described in the text.
	Metric 22: Reporting of Data	High	Exposure data and control response were reported in Figure 3.
	Metric 23: Explanation of Unexpected Outcomes	High	Variability was reported in Figure 3.

Additional Comments: This evaluation is for the assessment of photosynthesis/CO2 fixation by barley protoplasts.

Overall Quality Determination

Medium

Study Citation:	Huang, Q., Wang, Q., Tan, W., Song, G., Lu, G., Li, F. (2006). Biochemical responses of two typical duckweeds exposed to dibutyl phthalate. Journal of Environmental Science and Health, Part A: Toxic/Hazardous Substances & Environmental Engineering 41(8):1615-1626.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Lemna minor</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Oxidative stress (including redox biology)-Photosynthesis			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1323213			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	Low	The source was not reported.
	Metric 3:	Test Substance Purity	High	HPLC grade of test substance reported
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an a negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	Each exposures was conducted within a "glass beaker". The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	One time dose, tests repeated twice
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and suitably conditioned to lab culture
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	20 fronds in triplicate is adequate
Domain 5: Outcome Assessment				
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Study Citation:	Huang, Q., Wang, Q., Tan, W., Song, G., Lu, G., Li, F. (2006). Biochemical responses of two typical duckweeds exposed to dibutyl phthalate. Journal of Environmental Science and Health, Part A: Toxic/Hazardous Substances & Environmental Engineering 41(8):1615-1626.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Lemna minor</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Oxidative stress (including redox biology)-Photosynthesis			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1323213			
Domain	Metric	Rating	Comments	
	Metric 16:	Adequacy of Test Conditions	Medium	Environmental conditions of test system were most likely conducive to maintenance of organism health but actual measured condition values for control and exposed vessels was not reported
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes
Additional Comments:	Subtitles of Figure 1 and 2 were apparently switched.This form represents the mechanistic endpoints from the paper and include: Cell signaling/function-Oxidative stress (including redox biology)-Photosynthesis for Lemna			

Overall Quality Determination**High**

Study Citation:	Huang, Q., Wang, Q., Tan, W., Song, G., Lu, G., Li, F. (2006). Biochemical responses of two typical duckweeds exposed to dibutyl phthalate. Journal of Environmental Science and Health, Part A: Toxic/Hazardous Substances & Environmental Engineering 41(8):1615-1626.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Lemna minor</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1323213			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	Chemical was identified by name only and purchased from Sigma.	
	Metric 2: Test Substance Source	Low	"DBP (HPLC grade) was purchased from Sigma (USA)."	
	Metric 3: Test Substance Purity	High	The test substance was reported to be HPLC grade.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	Each exposure was conducted within a "glass beaker". The experimental system and methods for preparation of test media were described in adequate detail.	
	Metric 8: Consistency of Exposure Administration	High	A one-time dose was used. Tests were repeated twice.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and exposure frequency were reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations (<7.5 mg/L) were below the water solubility limit (11.2 mg/L).	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were collected from Tai Lake in China, were adequately described and suitably conditioned to lab culture.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	Twenty fronds in triplicate is adequate.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Medium	Environmental conditions of the test system were most likely conducive to maintenance of organism health but actual measured condition values for control and exposed vessels was not reported.	
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Study Citation:	Huang, Q., Wang, Q., Tan, W., Song, G., Lu, G., Li, F. (2006). Biochemical responses of two typical duckweeds exposed to dibutyl phthalate. Journal of Environmental Science and Health, Part A: Toxic/Hazardous Substances & Environmental Engineering 41(8):1615-1626.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Lemna minor</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	1323213

Domain	Metric	Rating	Comments
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described. Treatments were compared to the control using Student's t-test.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: The subtitles of Figures 1 and 2 were apparently switched.

Overall Quality Determination

High

Study Citation:	Melin, C., Egneus, H. (1983). Effects of di-n-butyl phthalate on growth and photosynthesis in algae and on isolated organelles from higher plants. <i>Physiologia Plantarum</i> 59(3):461-466.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Spinacea oleraceae</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Photosynthesis			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1333016			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The DBP was identified by name only.	
	Metric 2: Test Substance Source	Low	The source of the DBP was not reported.	
	Metric 3: Test Substance Purity	Low	The purity/grade of the DBP was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of a negative control, which was a solvent control containing ethanol.	
	Metric 5: Negative Control Response	High	The negative control response was mentioned in the text under Figure 4.	
	Metric 6: Randomized Allocation	Low	It was not reported how the thylakoids were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The authors provided limited details on the preparation of the DBP test solution.	
	Metric 8: Consistency of Exposure Administration	Low	Details regarding the exposure administration were limited. Study authors reported the temperature and test volumes though.	
	Metric 9: Measurement of Test Substance Concentration	Low	Study authors did not report if the test concentrations were measured at any point in the study.	
	Metric 10: Exposure Duration and Frequency	High	Exposure duration was described and seemed adequate.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	Exposure groups seemed fine and adequate results were obtained.	
	Metric 12: Testing at or Below Solubility Limit	High	Study authors reported the use of a vehicle solvent.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of spinach thylakoids was not reported.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if any acclimation occurred.	
	Metric 15: Number of Organisms and Replicates per Group	Low	The thylakoid concentration and number of replicates was not reported.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Test conditions seemed appropriate and were described on page 462 of the paper.	
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Study Citation:	Melin, C., Egneus, H. (1983). Effects of di-n-butyl phthalate on growth and photosynthesis in algae and on isolated organelles from higher plants. <i>Physiologia Plantarum</i> 59(3):461-466.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Spinacea oleraceae</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Photosynthesis
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	1333016

Domain	Metric	Rating	Comments
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—electron transport reactions in thylakoids from spinach.
	Metric 18: Consistency of Outcome Assessment	High	The methodology was consistent across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. It was not reported if acclimation occurred.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	IC50 value determination was described in the text.
	Metric 22: Reporting of Data	High	Exposure data and control response were reported in Figure 4.
	Metric 23: Explanation of Unexpected Outcomes	High	Variability was reported in Figure 4.

Additional Comments: This evaluation is for the assessment of photosynthesis/electron transfer reactions by spinach thylakoids.

Overall Quality Determination

Medium

Study Citation:	Huang, Q., Wang, Q., Tan, W., Song, G., Lu, G., Li, F. (2006). Biochemical responses of two typical duckweeds exposed to dibutyl phthalate. Journal of Environmental Science and Health, Part A: Toxic/Hazardous Substances & Environmental Engineering 41(8):1615-1626.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Spirodela polyrhiza</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Oxidative stress (including redox biology)-Photosynthesis			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1323213			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	Low	The source was not reported.
	Metric 3:	Test Substance Purity	High	HPLC grade of test substance reported
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an a negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	Each exposures was conducted within a "glass beaker". The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8:	Consistency of Exposure Administration	High	One time dose, tests repeated twice
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and suitably conditioned to lab culture
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	20 fronds in triplicate is adequate
Domain 5: Outcome Assessment				
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Study Citation:	Huang, Q., Wang, Q., Tan, W., Song, G., Lu, G., Li, F. (2006). Biochemical responses of two typical duckweeds exposed to dibutyl phthalate. Journal of Environmental Science and Health, Part A: Toxic/Hazardous Substances & Environmental Engineering 41(8):1615-1626.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Spirodela polyrhiza</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Oxidative stress (including redox biology)-Photosynthesis			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1323213			
Domain	Metric	Rating	Comments	
	Metric 16:	Adequacy of Test Conditions	Medium	Environmental conditions of test system were most likely conducive to maintenance of organism health but actual measured condition values for control and exposed vessels was not reported
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes
Additional Comments:	subtitles of Figure 1 and 2 were apparently switched.This form represents the mechanistic endpoints from the paper and include: Cell signaling/function-Oxidative stress (including redox biology)-Photosynthesis for Spirodela polyrhiza			

Overall Quality Determination**High**

Study Citation:	Huang, Q., Wang, Q., Tan, W., Song, G., Lu, G., Li, F. (2006). Biochemical responses of two typical duckweeds exposed to dibutyl phthalate. Journal of Environmental Science and Health, Part A: Toxic/Hazardous Substances & Environmental Engineering 41(8):1615-1626.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Spirodela polyrhiza</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1323213			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only and purchased from Sigma.	
	Metric 2: Test Substance Source	Low	"DBP (HPLC grade) was purchased from Sigma (USA)."	
	Metric 3: Test Substance Purity	High	HPLC grade of test substance was reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	Each exposure was conducted within a "glass beaker". The experimental system and methods for preparation of test media were described in adequate detail.	
	Metric 8: Consistency of Exposure Administration	High	A one-time dose was used. Tests were repeated twice.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and exposure frequency were reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations (<7.5 mg/L) were below the water solubility limit (11.2 mg/L).	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were collected from Tai Lake in China, adequately described and suitably conditioned to lab culture.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	Twenty fronds in triplicate is adequate.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Medium	Environmental conditions of the test system were most likely conducive to maintenance of organism health, but actual measured condition values for control and exposed vessels was not reported.	
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Study Citation:	Huang, Q., Wang, Q., Tan, W., Song, G., Lu, G., Li, F. (2006). Biochemical responses of two typical duckweeds exposed to dibutyl phthalate. Journal of Environmental Science and Health, Part A: Toxic/Hazardous Substances & Environmental Engineering 41(8):1615-1626.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Spirodela polyrhiza</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	1323213

Domain	Metric	Rating	Comments
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described. Treatments were compared to the control using Student's t-test.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: The subtitles of Figures 1 and 2 were apparently switched.

Overall Quality Determination

High

Study Citation:	Gao, M., Dong, Y., Zhang, Z., Song, W., Qi, Y. (2017). Growth and antioxidant defense responses of wheat seedlings to di-n-butyl phthalate and di (2-ethylhexyl) phthalate stress. Chemosphere 172(Elsevier):418-428.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Triticum sp.</i> ; Jinnong 7; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3515118			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The chemical was identified by name and CAS.	
	Metric 2: Test Substance Source	Low	The source was reported as J&K Scientific Ltd. (Beijing, China), but the test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	The chemical purity was reported as 96.8%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was suitable.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The experimental system and test media preparation methods were adequately reported. Petri dishes were covered with a lid to prevent evaporation. Water loss was checked every 24 hours, but loss of test substance was not measured.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	Medium	The solvent concentration slightly exceeded an appropriate concentration but the biological response of the solvent control was acceptable and no interactions are expected between the solvent and test substance.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.	
Domain 5: Outcome Assessment				
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Study Citation:	Gao, M., Dong, Y., Zhang, Z., Song, W., Qi, Y. (2017). Growth and antioxidant defense responses of wheat seedlings to di-n-butyl phthalate and di (2-ethylhexyl) phthalate stress. Chemosphere 172(Elsevier):418-428.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Triticum sp.</i> ; Jinnong 7; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Reproductive/Teratogenic
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	3515118

Domain	Metric	Rating	Comments
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health. The germination experiment was conducted in a "growth chamber in total darkness at a temperature of $25 \pm 1^{\circ}\text{C}$ and humidity of 80%".
	Metric 17: Outcome Assessment Methodology	Medium	The outcome assessment methodology (germination rate and root and shoot length) reported the intended outcome of interest but were not described adequately.
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to the exposure.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were described well.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group, for germination rate (Fig 1) and root/shoot elongation (Fig 2). IC10 and IC50 values were provided for root and shoot elongation.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments: This evaluation form is relevant for germination rate and shoot/root growth.			

Overall Quality Determination**High**

Study Citation:	Gao, M., Dong, Y., Zhang, Z., Song, W., Qi, Y. (2017). Growth and antioxidant defense responses of wheat seedlings to di-n-butyl phthalate and di (2-ethylhexyl) phthalate stress. Chemosphere 172(Elsevier):418-428.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Triticum sp.</i> ; Jinnong 7; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3515118			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	Chemical was identified by name and CAS	
	Metric 2: Test Substance Source	Low	Source reported as J&K Scientific Ltd. (Beijing, China) but the test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity reported as 96.8%	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group	
	Metric 5: Negative Control Response	Low	The biological response of the negative control group was not reported.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The experimental system and test media preparation methods were adequately reported. The test solutions were replenished daily.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response	
	Metric 12: Testing at or Below Solubility Limit	Medium	The solvent concentration was reported but the biological response of the control was not.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects	
Domain 5: Outcome Assessment				
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Study Citation:	Gao, M., Dong, Y., Zhang, Z., Song, W., Qi, Y. (2017). Growth and antioxidant defense responses of wheat seedlings to di-n-butyl phthalate and di (2-ethylhexyl) phthalate stress. Chemosphere 172(Elsevier):418-428.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Triticum sp.</i> ; Jinnong 7; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3515118			
Domain	Metric	Rating	Comments	
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health. "The experiments were performed in an artificial climate chamber with alternating temperatures of 25 ± 1oC (12 h light) and 20 ± 1oC (12 h dark), 60% relative humidity, and a light intensity of 40 uM/ms".	
	Metric 17: Outcome Assessment Methodology	Medium	The outcome assessment methodology (for determining DBP and DEHP in shoot and root) reported the intended outcome of interest. Key details (percent recovery, method detection limit, etc.) for HPLC analysis were not provided.	
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure that could influence the outcome assessment.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.	
	Metric 22: Reporting of Data	Medium	Data for exposure-related findings were presented for each treatment group. Control response was not provided.	
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.	
Additional Comments:	Control group was not analyzed for DBP. The exposure concentrations were not verified.			
Overall Quality Determination		High		

Study Citation:	Gao, M., Dong, Y., Zhang, Z., Song, W., Qi, Y. (2017). Growth and antioxidant defense responses of wheat seedlings to di-n-butyl phthalate and di (2-ethylhexyl) phthalate stress. Chemosphere 172(Elsevier):418-428.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Triticum sp.</i> ; Jinnong 7; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Oxidative stress (including redox biology)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3515118			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	Chemical was identified by name and CAS	
	Metric 2: Test Substance Source	Low	Source reported as J&K Scientific Ltd. (Beijing, China)but the test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity reported as 96.8%	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was suitable	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The experimental system and/or test media preparation methods were adequately reported. The test solutions were replenished daily.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response	
	Metric 12: Testing at or Below Solubility Limit	Medium	The solvent concentration slightly exceeded an appropriate concentration but the biological response of the solvent control was acceptable and no interactions are expected between the solvent and test substance.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects	
Domain 5: Outcome Assessment				
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Study Citation:	Gao, M., Dong, Y., Zhang, Z., Song, W., Qi, Y. (2017). Growth and antioxidant defense responses of wheat seedlings to di-n-butyl phthalate and di (2-ethylhexyl) phthalate stress. Chemosphere 172(Elsevier):418-428.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Triticum sp.</i> ; Jinnong 7; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Oxidative stress (including redox biology)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3515118			
Domain	Metric	Rating	Comments	
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health. "The experiments were performed in an artificial climate chamber with alternating temperatures of 25 ± 1 oC (12 h light) and 20 ± 1 oC (12 h dark), 60% relative humidity, and a light intensity of 40 uM/ms".	
	Metric 17: Outcome Assessment Methodology	Low	The outcome assessment methodologies for enzyme activities were not clearly reported.	
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical methods were adequately described	
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group for all mechanistic end points (Figures 4, 5 and 6).	
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.	
Additional Comments:	This evaluation form is relevant to all mechanistic endpoints including antioxidant enzyme activities, lipid peroxidation, O2 accumulation and plasma membrane permeability.			

Overall Quality Determination**High**

Study Citation:	Gao, M., Dong, Y., Zhang, Z., Song, W., Qi, Y. (2017). Growth and antioxidant defense responses of wheat seedlings to di-n-butyl phthalate and di (2-ethylhexyl) phthalate stress. Chemosphere 172(Elsevier):418-428.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Triticum sp.</i> ; Jinnong 7; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3515118			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The chemical was identified by name and CAS.	
	Metric 2: Test Substance Source	Low	The source was reported as J&K Scientific Ltd. (Beijing, China), but the test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity was reported as 96.8%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was suitable.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The experimental system and test media preparation methods were adequately reported. The test solutions were replenished daily.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	Medium	The solvent concentration slightly exceeded an appropriate concentration but the biological response of the solvent control was acceptable and no interactions are expected between the solvent and test substance.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.	
Domain 5: Outcome Assessment				
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Study Citation:	Gao, M., Dong, Y., Zhang, Z., Song, W., Qi, Y. (2017). Growth and antioxidant defense responses of wheat seedlings to di-n-butyl phthalate and di (2-ethylhexyl) phthalate stress. Chemosphere 172(Elsevier):418-428.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Triticum sp.</i> ; Jinnong 7; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3515118			
Domain	Metric	Rating	Comments	
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health. "The experiments were performed in an artificial climate chamber with alternating temperatures of 25 ± 1 oC (12 h light) and 20 ± 1 oC (12 h dark), 60% relative humidity, and a light intensity of 40 uM/ms."	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest. Roots were scanned using an EPSON Expression and measurements (total root length, total root surfacearea, average root diameter, and the number of root tips and hairs) were obtained from digital images.	
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure (e.g., infection) that could influence the outcome assessment.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.	
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group (Table 2 and Fig 3).	
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.	
Additional Comments:	Root morphology measurements included total root length, total root surface area, average root diameter, and the number of root tips and hairs. The exposure concentrations were not verified.			

Overall Quality Determination**High**

Study Citation:	Liang, W., Deng, J. Q., Zhan, F. C., Wu, Z. B. (2009). Effects of constructed wetland system on the removal of dibutyl phthalate (DBP). Microbiological Research 164(2):206-211.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Other; Fungus; <i>fungus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1323196			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The chemical was identified by name only.
	Metric 2:	Test Substance Source	Low	The source was not reported.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of the test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Study authors reported using an appropriate concurrent negative control system.
	Metric 5:	Negative Control Response	Medium	The biological response of the negative control group was reported and reasonable for assessed outcomes. There was no report of initial population numbers.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided no details on the measures taken to appropriately prepare test concentrations.
	Metric 8:	Consistency of Exposure Administration	Low	It is unclear when and how DBP was added to the system.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies.
	Metric 10:	Exposure Duration and Frequency	Medium	The duration of exposure was reported and appropriate for the study type, but it is unclear when dosing occurred.
	Metric 11:	Number of Exposure Groups/Spacing of Exposure Levels	Low	Two dissimilar doses were applied at each phase, but the phase was not defined.
	Metric 12:	Testing at or Below Solubility Limit	Medium	A subset of the exposure concentrations exceeded the water solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Uninformative	Test organisms were not added to the system and the source was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	N/A	Organisms may have been previously exposed to the test substance or other unintended stressors.
	Metric 15:	Number of Organisms and Replicates per Group	Uninformative	The initial number of organisms was not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.

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Study Citation:	Liang, W., Deng, J. Q., Zhan, F. C., Wu, Z. B. (2009). Effects of constructed wetland system on the removal of dibutyl phthalate (DBP). Microbiological Research 164(2):206-211.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Other; Fungus; <i>fungus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1323196			
Domain	Metric	Rating	Comments	
	Metric 17:	Outcome Assessment Methodology	Medium	The outcome assessment methodology partially addressed the intended outcomes of interest.
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	N/A	Statistical analysis was not possible (n=1).
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23:	Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability.
Additional Comments:	Enzymes were also measured but not attributed to any entity.			

Overall Quality Determination**Uninformative**

Study Citation:	Bionomics,, Springborn (1984). Acute toxicity of thirteen phthalate esters to the sheepshead minnow (<i>Cyprinodon variegatus</i>) (final report).			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (marine), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>sheepshead minnow (Cyprinodon variegatus)</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1316224; Linked HERO ID(s): 1321996, 1316224			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The test material was identified and a CASRN was given.	
	Metric 2: Test Substance Source	High	The source was listed as EG&G Bionomics- Aquatic Toxicology Laboratory in Wareham, Massachusetts. No other information about the source was given.	
	Metric 3: Test Substance Purity	Low	Purity and/or grade of the test substance were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	A negative control was used.	
	Metric 5: Negative Control Response	High	No mortality was reported in the controls.	
	Metric 6: Randomized Allocation	Medium	Test organisms were impartially distributed to each chamber (pdf pg 136).	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail and accounted for the properties of the test material. For all low-solubility phthalates, enhanced mixing procedure was used, while for butyl benzyl phthalate exposure microbial degradation was accounted for with a cleaning procedure that was implemented daily. The authors reported significant degradation of the test material throughout the test, but quantified the degradation throughout the test and reported the endpoint in terms of mean-measured concentration so this does not have an impact on the results.	
	Metric 8: Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	High	Concentrations were measured using GS-MS to account for poor water solubility.	
	Metric 10: Exposure Duration and Frequency	High	The 96-hour exposure was appropriate for an acute test.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	N/A	Test concentration spacing was limited, as this was designed as a limit test up to the solubility limit of the chemical.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit, which were reported in Appendix A (pdf pg 164).	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	Specimens were either cultured at the Laboratory or purchased commercially. All fish were tested as juveniles at <10weeks old.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	A 96-hour acclimation period was reported.	

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Study Citation:	Bionomics,, Springborn (1984). Acute toxicity of thirteen phthalate esters to the sheepshead minnow (<i>Cyprinodon variegatus</i>) (final report).			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (marine), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>sheepshead minnow (Cyprinodon variegatus)</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1316224; Linked HERO ID(s): 1321996, 1316224			
Domain	Metric	Rating	Comments	
	Metric 15: Number of Organisms and Replicates per Group	Low	Only two replicates of 10 fish were used in each treatment.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Organism housing, environmental conditions, food, water, and nutrients were conducive to maintenance of health, and biomass loading was appropriate.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology partially addressed or reported the intended outcomes(s) of interest (mortality).	
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Medium	The study reported minor differences among the study groups with respect to environmental conditions or other non-treatment-related factors, but these are unlikely to have a substantial impact on results. Authors reported that dissolved oxygen fell below guideline recommended levels and control mortality was high for one phthalate (not this phthalate). As no mortalities were observed in any test concentrations for this chemical, this was not determined to affect this test.	
	Metric 20: Outcomes Unrelated to Exposure	High	Details regarding test organism attrition and outcomes unrelated to exposure (e.g., infection) were reported for each study group and there were no differences among groups that could influence the outcome assessment.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	N/A	No mortality was reported, so no statistical analysis was needed.	
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group and were adequate to determine values for the endpoint(s) of interest. Negative findings were reported quantitatively.	
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes, or unexpected outcomes were satisfactorily explained. Low DO did not have an affect on the outcome and high mortality in controls was not reported for this chemical.	
Additional Comments:	Study report for the Sheepshead minnow test begins on pg 124 of the PDF. DEHP is referred to as phthalate 1H, Dibutyl Phthalate was referred to as 1C, and Butyl Benzyl Phthalate was referred to as 1D.			

Overall Quality Determination**High**

Study Citation:	Tagatz, M. E., Deans, C. H., Moore, J. C., Plaia, G. R. (1983). Alterations in composition of field-developed and laboratory-developed estuarine benthic communities exposed to di-normal-butyl phthalate. <i>Aquatic Toxicology</i> 3(3):239-248.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Other Invertebrate (e.g., sea urchins, ciliates, rotifers); <i>Animalia</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5495608			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	High	The test substance was identified using electron-capture gas-liquid chromatography.	
	Metric 3: Test Substance Purity	Low	The purity of the test substance was 100%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how initial placement of aquaria was determined.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The experimental system and/or test media preparation methods were adequately reported but there is some concern over the use of plastic trays.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Medium	Naturally colonizing organisms were used, and it was assumed there was equal initial population structure.	
	Metric 14: Acclimatization and Pretreatment Conditions	Medium	All pretreatment conditions were most likely the same for control and exposed organisms.	
	Metric 15: Number of Organisms and Replicates per Group	Low	The initial number of test organisms was not reported.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.	
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Study Citation:	Tagatz, M. E., Deans, C. H., Moore, J. C., Plaia, G. R. (1983). Alterations in composition of field-developed and laboratory-developed estuarine benthic communities exposed to di-normal-butyl phthalate. <i>Aquatic Toxicology</i> 3(3):239-248.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Other Invertebrate (e.g., sea urchins, ciliates, rotifers); <i>Animalia</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	5495608

Domain	Metric	Rating	Comments
	Metric 17: Outcome Assessment Methodology	Low	The outcome assessment methodology (harvesting) was not clearly reported.
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.

Additional Comments: None

Overall Quality Determination**Medium**

Study Citation:	Tagatz, M. E., Deans, C. H., Moore, J. C., Plaia, G. R. (1983). Alterations in composition of field-developed and laboratory-developed estuarine benthic communities exposed to di-normal-butyl phthalate. <i>Aquatic Toxicology</i> 3(3):239-248.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Other Invertebrate (e.g., sea urchins, ciliates, rotifers); <i>Animalia</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5495608			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	High	The test substance was identified using electron-capture gas-liquid chromatography.	
	Metric 3: Test Substance Purity	Low	The purity of the test substance was 100%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how initial placement of aquaria was determined.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The experimental system and/or test media preparation methods were adequately reported but there is some concern over the use of plastic trays.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Medium	Naturally colonizing organisms were used, and it is assumed there was equal initial population structure.	
	Metric 14: Acclimatization and Pretreatment Conditions	Medium	All pretreatment conditions were most likely the same for control and exposed organisms.	
	Metric 15: Number of Organisms and Replicates per Group	Low	The initial number of test organisms was not reported.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.	
	Metric 17: Outcome Assessment Methodology	Low	The outcome assessment methodology (harvesting) was not clearly reported.	
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.	
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Study Citation:	Tagatz, M. E., Deans, C. H., Moore, J. C., Plaia, G. R. (1983). Alterations in composition of field-developed and laboratory-developed estuarine benthic communities exposed to di-normal-butyl phthalate. <i>Aquatic Toxicology</i> 3(3):239-248.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Other Invertebrate (e.g., sea urchins, ciliates, rotifers); <i>Animalia</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	5495608

Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.

Additional Comments: None

Overall Quality Determination**Medium**

Study Citation:	Tagatz, M. E., Deans, C. H., Moore, J. C., Plaia, G. R. (1983). Alterations in composition of field-developed and laboratory-developed estuarine benthic communities exposed to di-normal-butyl phthalate. <i>Aquatic Toxicology</i> 3(3):239-248.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); <i>Annelida</i> ; multiple species; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5495608			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	High	The test substance was identified using electron-capture gas-liquid chromatography.	
	Metric 3: Test Substance Purity	Low	Purity of the test substance was 100%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how initial placement of aquaria was determined.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The experimental system and/or test media preparation methods were adequately reported but there is some concern over the use of plastic trays.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Medium	Naturally colonizing organisms were used, and it is assumed there was equal initial population structure.	
	Metric 14: Acclimatization and Pretreatment Conditions	Medium	All pretreatment conditions were most likely the same for control and exposed organisms.	
	Metric 15: Number of Organisms and Replicates per Group	Low	The initial number of test organisms was not reported.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.	
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Study Citation:	Tagatz, M. E., Deans, C. H., Moore, J. C., Plaia, G. R. (1983). Alterations in composition of field-developed and laboratory-developed estuarine benthic communities exposed to di-normal-butyl phthalate. <i>Aquatic Toxicology</i> 3(3):239-248.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); <i>Annelida</i> ; multiple species; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	5495608

Domain	Metric	Rating	Comments
	Metric 17: Outcome Assessment Methodology	Low	The outcome assessment methodology (harvesting) was not clearly reported.
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.

Additional Comments: None

Overall Quality Determination**Medium**

Study Citation:	Tagatz, M. E., Deans, C. H., Moore, J. C., Plaia, G. R. (1983). Alterations in composition of field-developed and laboratory-developed estuarine benthic communities exposed to di-normal-butyl phthalate. <i>Aquatic Toxicology</i> 3(3):239-248.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); <i>Annelida</i> ; multiple species; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5495608			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	High	The test substance was identified using electron-capture gas-liquid chromatography.	
	Metric 3: Test Substance Purity	Low	The purity of test substance was 100%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how initial placement of aquaria was determined.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The experimental system and/or test media preparation methods were adequately reported but there is some concern over the use of plastic trays.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Medium	Naturally colonizing organisms were used, and it is assumed there was equal initial population structure.	
	Metric 14: Acclimatization and Pretreatment Conditions	Medium	All pretreatment conditions were most likely the same for control and exposed organisms.	
	Metric 15: Number of Organisms and Replicates per Group	Low	The initial number of test organisms was not reported.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.	
	Metric 17: Outcome Assessment Methodology	Low	The outcome assessment methodology (harvesting) was not clearly reported.	
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.	

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Study Citation:	Tagatz, M. E., Deans, C. H., Moore, J. C., Plaia, G. R. (1983). Alterations in composition of field-developed and laboratory-developed estuarine benthic communities exposed to di-normal-butyl phthalate. <i>Aquatic Toxicology</i> 3(3):239-248.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); <i>Annelida</i> ; multiple species; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	5495608

Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.

Additional Comments: None

Overall Quality Determination**Medium**

Study Citation:	Sugawara, N. (1974). Effect of phthalate esters on shrimp. Bulletin of Environmental Contamination and Toxicology 12(4):421-424.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Artemia salina</i> ; Larvae			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5569571			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The DBP was identified by name only.
	Metric 2:	Test Substance Source	Low	The source of DBP was not reported.
	Metric 3:	Test Substance Purity	Low	The purity/grade of the DBP was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	A concurrent negative control was used in the study.
	Metric 5:	Negative Control Response	High	The negative control response was reported in Table 1 and was adequate for the outcome of interest.
	Metric 6:	Randomized Allocation	Low	It was not reported how the brine shrimp larvae were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	Petri dishes were used as the test chambers, but little information was provided on the preparation of the test substance.
	Metric 8:	Consistency of Exposure Administration	Low	Details regarding the exposure administration were limited. All exposures were for 24h in 9 x 2cm petri dishes, but more information was needed.
	Metric 9:	Measurement of Test Substance Concentration	Low	It was not reported if the test substance was measured at any point.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration was 24h. This was adequate to determine the hatch.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	There were only three exposure levels, but the spacing was adequate to see a response.
	Metric 12:	Testing at or Below Solubility Limit	Uninformative	The highest two concentrations exceeded the water solubility limit, and it was not reported if a vehicle solvent was used.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the shrimp was only reported to be a commercial source. More information is necessary.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the organisms were acclimated.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	There were 10 larvae in each petri dish with 3-6 replicates per exposure level.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Tests were carried out at 26C, but little other information on the environmental conditions was reported.

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Study Citation:	Sugawara, N. (1974). Effect of phthalate esters on shrimp. Bulletin of Environmental Contamination and Toxicology 12(4):421-424.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Artemia salina</i> ; Larvae			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5569571			
Domain	Metric	Rating	Comments	
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—larvae survival.
	Metric 18:	Consistency of Outcome Assessment	High	After 24h, the number of larvae mortalities was counted. This was determined by whether the larvae moved or not.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described.
	Metric 22:	Reporting of Data	High	Data for each exposure response and the control response was reported in Table 1 and was adequate for the outcome of interest.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in Table 1.
Additional Comments:	This portion of the evaluation was on the effect of DBP on the survival of A. salina larvae. Mortality was selected as the outcome of interest. The study received an unacceptable ranking due to the two highest test concentrations being above the the water solubility limit and the lack of reporting on a vehicle solvent.			

Overall Quality Determination**Uninformative**

Study Citation:	Sugawara, N. (1974). Effect of phthalate esters on shrimp. Bulletin of Environmental Contamination and Toxicology 12(4):421-424.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Artemia salina</i> ; Embryo			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5569571			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The DBP was identified by name only.
	Metric 2:	Test Substance Source	Low	The source of DBP was not reported.
	Metric 3:	Test Substance Purity	Low	The purity/ grade of the DBP was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	A concurrent negative control was used in the study.
	Metric 5:	Negative Control Response	High	The negative control response was reported in Figure 1 and was adequate for the outcome of interest.
	Metric 6:	Randomized Allocation	Low	It was not reported how the brine shrimp embryos were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	Petri dishes were used as the test chambers, but little information was provided on the preparation of the test substance.
	Metric 8:	Consistency of Exposure Administration	Low	Details regarding the exposure administration were limited. All exposures were for 24h in 9 x 2cm petri dishes, but more information was needed.
	Metric 9:	Measurement of Test Substance Concentration	Low	It was not reported if the test substance was measured at any point.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration was 24h. This was adequate to determine the hatch.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	There were only 3 exposure levels, but the spacing was adequate to see a response.
	Metric 12:	Testing at or Below Solubility Limit	Uninformative	The highest 2 concentrations exceeded the water solubility limit, and it was not reported if a vehicle solvent was used.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the shrimp was only reported to be a commercial source. More information is necessary.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the organisms were acclimated.
	Metric 15:	Number of Organisms and Replicates per Group	Low	There were 1-2g of embryos in each petri dish. The number of replicates was not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Tests were carried out at 26C, but little other information on the environmental conditions was reported.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—the number of embryos that hatched.

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Study Citation:	Sugawara, N. (1974). Effect of phthalate esters on shrimp. Bulletin of Environmental Contamination and Toxicology 12(4):421-424.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Artemia salina</i> ; Embryo			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5569571			
Domain	Metric	Rating	Comments	
	Metric 18: Consistency of Outcome Assessment	Low	After 24h, the number of nauplius was counted. More details are needed on the assessment procedure to determine consistency.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Data was analyzed according to the t-test.	
	Metric 22: Reporting of Data	High	Data for each exposure response and the control response was reported in Figure 1 and was adequate for the outcome of interest.	
	Metric 23: Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in Figure 1.	
Additional Comments:	This portion of the evaluation was on the effect of DBP on the hatch of A. salina embryos. Mortality was selected as the outcome of interest. The study received an unacceptable ranking due to the two highest test concentrations being above the the water solubility limit and the lack of reporting on a vehicle solvent			

Overall Quality Determination**Uninformative**

Study Citation:	Tagatz, M. E., Deans, C. H., Moore, J. C., Plaia, G. R. (1983). Alterations in composition of field-developed and laboratory-developed estuarine benthic communities exposed to di-normal-butyl phthalate. <i>Aquatic Toxicology</i> 3(3):239-248.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Arthropoda</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5495608			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	High	The test substance was identified using electron-capture gas-liquid chromatography.	
	Metric 3: Test Substance Purity	Low	Purity of the test substance was 100%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how initial placement of aquaria was determined.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The experimental system and/or test media preparation methods were adequately reported but there is some concern over the use of plastic trays.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Medium	Naturally colonizing organisms were used, and it is assumed there was equal initial population structure.	
	Metric 14: Acclimatization and Pretreatment Conditions	Medium	All pretreatment conditions were most likely the same for control and exposed organisms.	
	Metric 15: Number of Organisms and Replicates per Group	Low	The initial number of test organisms was not reported.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.	
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Study Citation:	Tagatz, M. E., Deans, C. H., Moore, J. C., Plaia, G. R. (1983). Alterations in composition of field-developed and laboratory-developed estuarine benthic communities exposed to di-normal-butyl phthalate. <i>Aquatic Toxicology</i> 3(3):239-248.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Arthropoda</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	5495608

Domain	Metric	Rating	Comments
	Metric 17: Outcome Assessment Methodology	Low	The outcome assessment methodology (harvesting) was not clearly reported.
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.

Additional Comments: None

Overall Quality Determination**Medium**

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Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5495608			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	High	The test substance was identified using electron-capture gas-liquid chromatography.	
	Metric 3: Test Substance Purity	Low	Purity of the test substance was 100%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how initial placement of aquaria was determined.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The experimental system and/or test media preparation methods were adequately reported but there is some concern over the use of plastic trays.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Medium	Naturally colonizing organisms were used, and it is assumed there was equal initial population structure.	
	Metric 14: Acclimatization and Pretreatment Conditions	Medium	All pretreatment conditions were most likely the same for control and exposed organisms.	
	Metric 15: Number of Organisms and Replicates per Group	Low	The initial number of test organisms was not reported.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.	
	Metric 17: Outcome Assessment Methodology	Low	The outcome assessment methodology (harvesting) was not clearly reported.	
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.	
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	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
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Additional Comments: None

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Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5495608			
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Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using a concurrent negative control group.
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	Metric 7:	Experimental System/Test Media Preparation	Medium	The experimental system and/or test media preparation methods were adequately reported but there is some concern over the use of plastic trays.
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Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.

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Chemical:	Dibutyl phthalate (DBP)
HERO ID:	5495608

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Additional Comments: None

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Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group.	
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	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.	
	Metric 17: Outcome Assessment Methodology	Low	The outcome assessment methodology (harvesting) was not clearly reported.	
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Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5495608			
Domain	Metric	Rating	Comments	
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
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Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23:	Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.
Additional Comments:	None			

Overall Quality Determination**Medium**

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Domain 5: Outcome Assessment				
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	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.

Additional Comments: None

Overall Quality Determination**Medium**

Study Citation:	Tagatz, M. E., Deans, C. H., Moore, J. C., Plaia, G. R. (1983). Alterations in composition of field-developed and laboratory-developed estuarine benthic communities exposed to di-normal-butyl phthalate. <i>Aquatic Toxicology</i> 3(3):239-248.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Other Invertebrate (e.g., sea urchins, ciliates, rotifers); <i>Echinodermata</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5495608			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	High	The test substance was identified using electron-capture gas-liquid chromatography.	
	Metric 3: Test Substance Purity	Low	Purity of the test substance was 100%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how initial placement of aquaria was determined.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The experimental system and/or test media preparation methods were adequately reported but there is some concern over the use of plastic trays.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Medium	Naturally colonizing organisms were used, and it is assumed there was equal initial population structure.	
	Metric 14: Acclimatization and Pretreatment Conditions	Medium	All pretreatment conditions were most likely the same for control and exposed organisms.	
	Metric 15: Number of Organisms and Replicates per Group	Low	The initial number of test organisms was not reported.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.	
	Metric 17: Outcome Assessment Methodology	Low	The outcome assessment methodology (harvesting) was not clearly reported.	
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.	
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Study Citation:	Tagatz, M. E., Deans, C. H., Moore, J. C., Plaia, G. R. (1983). Alterations in composition of field-developed and laboratory-developed estuarine benthic communities exposed to di-normal-butyl phthalate. <i>Aquatic Toxicology</i> 3(3):239-248.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Other Invertebrate (e.g., sea urchins, ciliates, rotifers); <i>Echinodermata</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	5495608

Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.

Additional Comments: None

Overall Quality Determination**Medium**

Study Citation:	Liu, Y., Guan, Y., Yang, Z., Cai, Z., Mizuno, T., Tsuno, H., Zhu, W., Zhang, X. (2009). Toxicity of seven phthalate esters to embryonic development of the abalone <i>Haliotis diversicolor supertexta</i> . <i>Ecotoxicology</i> 18(3):293-303.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Mollusks; <i>Haliotis diversicolor supertexta</i> ; Embryo			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	697762			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The chemical was identified only by name. No other information was provided.
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity was reported as at least 99%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative and solvent control group.
	Metric 5:	Negative Control Response	High	The biological responses (percentage of cleavage, normal blastula, larval settlement and metamorphosis) of the negative control group were adequate.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	Methods for preparation of test media were described in adequate detail, however steps taken to minimize loss of test substance was not reported.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Medium	Exposure concentrations were measured at the end of the experiment, but it is not clear whether new test solutions were used for the incubation from swimming stage to the metamorphosis stage. Measured concentrations were similar to nominal concentrations.
	Metric 10:	Exposure Duration and Frequency	High	The total duration of exposure was 96 hours. The experiment was done in two phases covering developmental stages from fertilization to early veliger stage (12 hours) and from veliger to metamorphosis stage (84 hours).
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	There were minor limitations regarding the number of exposure groups and spacing of exposure levels. 96 hr EC-50 values could not established using the concentrations tested.
	Metric 12:	Testing at or Below Solubility Limit	Medium	The solvent concentration (3.75 %v/v) slightly exceeded an appropriate concentration but the biological response of the solvent control was acceptable.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	There are minor reservations or uncertainties about the choice of test species source. Test organisms were collected from the field and prior exposure to phthalates may have occurred.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.
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Study Citation:	Liu, Y., Guan, Y., Yang, Z., Cai, Z., Mizuno, T., Tsuno, H., Zhu, W., Zhang, X. (2009). Toxicity of seven phthalate esters to embryonic development of the abalone <i>Haliotis diversicolor supertexta</i> . <i>Ecotoxicology</i> 18(3):293-303.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Mollusks; <i>Haliotis diversicolor supertexta</i> ; Embryo			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	697762			
Domain	Metric	Rating	Comments	
	Metric 15:	Number of Organisms and Replicates per Group	Low	The numbers of test organisms and replicates were reported. The fertilized egg density used for each treatment group and controls were not verified after adding 10 ml of fertilization medium to each test vessel.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Environmental conditions of the test system were conducive to maintenance of organism health. DBP was found in the dilution medium (1,775 ± 23 ng/l shown in Table 2).
	Metric 17:	Outcome Assessment Methodology	Medium	The outcome assessment methodology partially addressed the intended outcomes. The percentage of embryos undergoing cleavage and percentage of settled larvae were not sensitive endpoints. However, the percentage of normal blastula was a sensitive end point and 9hr EC50 values were calculated. The 96hr EC50 values, based on percentage of larvae that underwent metamorphosis, could not be calculated. Instead, 96hr NOEC values were reported.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical analysis was performed. ANOVA was used to test for differences among treatments and probit analysis was used to calculate 9hr EC50 values.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23:	Explanation of Unexpected Outcomes	Medium	Minor uncertainties or limitations were identified in how the study characterized unexpected outcomes. There was large within study variability for the settlement end point as both normal and abnormal larvae settled.
Additional Comments:	9hr EC50 values were reported based on the percentage of normal blastula and 96hr NOEC values were reported based on the percentage of larvae that underwent metamorphosis. For DBP, 96hr NOEC values were adjusted for background concentration of DBP in the dilution medium.			

Overall Quality Determination**Medium**

Study Citation:	Yang, Z. H., Zhang, X. J., Cai, Z. H. (2009). Toxic effects of several phthalate esters on the embryos and larvae of abalone <i>Haliotis diversicolor supertexta</i> . Chinese Journal of Oceanology and Limnology 27(2):395-399.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Mollusks; <i>Haliotis diversicolor supertexta</i> ; Larvae			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1322103			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The chemical was identified by name.	
	Metric 2: Test Substance Source	Low	The test substance was obtained from Sigma-Aldrich China.	
	Metric 3: Test Substance Purity	High	The chemical purity was reported as >99%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using appropriate concurrent negative and solvent control groups.	
	Metric 5: Negative Control Response	High	The biological response of the negative control groups was reported for settlement rate.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The experimental system and methods for preparation of test media were described in adequate detail but uncertainty over duration and concern for chemical loss caused downgrading.	
	Metric 8: Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	Low	Exposure duration was to trochophore stage, which was somewhat arbitrary.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were justified for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit, solvent aided.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Medium	There are minor reservations about the source of test organisms.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Organism environmental conditions were conducive to maintenance of health.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed the intended outcome of interest.	
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Study Citation:	Yang, Z. H., Zhang, X. J., Cai, Z. H. (2009). Toxic effects of several phthalate esters on the embryos and larvae of abalone <i>Haliotis diversicolor supertexta</i> . Chinese Journal of Oceanology and Limnology 27(2):395-399.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Mollusks; <i>Haliotis diversicolor supertexta</i> ; Larvae
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	1322103

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	High	The outcome assessment protocol was reported and consistently applied across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups for outcomes unrelated to exposures.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: None

Overall Quality Determination**Medium**

Study Citation:	Zhou, J., Cai, Z. H., Xing, K. Z. (2011). Potential mechanisms of phthalate ester embryotoxicity in the abalone <i>Haliotis diversicolor supertexta</i> . <i>Environmental Pollution</i> 159(5):1114-1122.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Mollusks; <i>Haliotis diversicolor supertexta</i> ; Embryo			
Health Outcome:	Mechanistic-Cell signaling/function			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1249532			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Medium	Chemical identified by name
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory
	Metric 3:	Test Substance Purity	High	Purity reported at >=98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using appropriate concurrent negative control groups
	Metric 5:	Negative Control Response	High	The biological responses of the negative control groups were adequate
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were adequate
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit aided by solvent at an appropriate concentration
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test animals was not reported
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Organism environmental conditions were conducive to maintenance of organism health
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed the intended outcomes of interest

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Study Citation:	Zhou, J., Cai, Z. H., Xing, K. Z. (2011). Potential mechanisms of phthalate ester embryotoxicity in the abalone <i>Haliotis diversicolor supertexta</i> . Environmental Pollution 159(5):1114-1122.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Mollusks; <i>Haliotis diversicolor supertexta</i> ; Embryo			
Health Outcome:	Mechanistic-Cell signaling/function			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1249532			
Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	High	There were no differences among groups that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were clearly described
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes
Additional Comments: Gene expression				

Overall Quality Determination**High**

Study Citation:	Zhou, J., Cai, Z. H., Xing, K. Z. (2011). Potential mechanisms of phthalate ester embryotoxicity in the abalone <i>Haliotis diversicolor supertexta</i> . Environmental Pollution 159(5):1114-1122.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Mollusks; <i>Haliotis diversicolor supertexta</i> ; Embryo			
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1249532			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Medium	Chemical identified by name	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory	
	Metric 3: Test Substance Purity	High	Purity reported at >=98%	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using appropriate concurrent negative control groups	
	Metric 5: Negative Control Response	High	The biological responses of the negative control groups were adequate	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail	
	Metric 8: Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were adequate	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit aided by solvent at an appropriate concentration	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the test animals was not reported	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Organism environmental conditions were conducive to maintenance of organism health	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed the intended outcomes of interest	
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups	

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Study Citation:	Zhou, J., Cai, Z. H., Xing, K. Z. (2011). Potential mechanisms of phthalate ester embryotoxicity in the abalone <i>Haliotis diversicolor supertexta</i> . Environmental Pollution 159(5):1114-1122.		
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days		
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Invertebrate; Mollusks; <i>Haliotis diversicolor supertexta</i> ; Embryo		
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	1249532		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
	Metric 20: Outcomes Unrelated to Exposure	High	There were no differences among groups that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were clearly described
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes
Additional Comments: MDA and POD changes			
Overall Quality Determination		High	

Study Citation:	Zhou, J., Cai, Z. H., Xing, K. Z. (2011). Potential mechanisms of phthalate ester embryotoxicity in the abalone <i>Haliotis diversicolor supertexta</i> . <i>Environmental Pollution</i> 159(5):1114-1122.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Mollusks; <i>Haliotis diversicolor supertexta</i> ; Embryo			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1249532			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Medium	The chemical was identified by name.
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Purity was reported at >=98%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using appropriate concurrent negative control groups.
	Metric 5:	Negative Control Response	High	The biological responses of the negative control groups were adequate.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were adequate.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit aided by solvent at an appropriate concentration.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test animals was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Organism housing and environmental conditions were conducive to maintenance of organism health.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed the intended outcomes of interest.
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Study Citation:	Zhou, J., Cai, Z. H., Xing, K. Z. (2011). Potential mechanisms of phthalate ester embryotoxicity in the abalone <i>Haliotis diversicolor supertexta</i> . Environmental Pollution 159(5):1114-1122.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Mollusks; <i>Haliotis diversicolor supertexta</i> ; Embryo			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1249532			
Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	High	There were no differences among groups that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were clearly described.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments: Metamorphosis rate, hatch rate, abnormality rate, surface structure changes				

Overall Quality Determination**High**

Study Citation:	Yang, Z. H., Zhang, X. J., Cai, Z. H. (2009). Toxic effects of several phthalate esters on the embryos and larvae of abalone <i>Haliotis diversicolor supertexta</i> . Chinese Journal of Oceanology and Limnology 27(2):395-399.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Mollusks; <i>Haliotis diversicolor supertexta</i> ; Embryo			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1322103			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The chemical was identified by name.	
	Metric 2: Test Substance Source	High	The test substance was obtained from Sigma-Aldrich China.	
	Metric 3: Test Substance Purity	High	The chemical purity was reported as >99%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using appropriate concurrent negative and solvent control groups.	
	Metric 5: Negative Control Response	High	The biological response of the negative control groups was reported for abnormal development.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The experimental system and methods for preparation of test media were described in adequate detail but uncertainty over duration and concern for chemical loss caused downgrading.	
	Metric 8: Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	Low	The exposure duration was to trochophore stage, somewhat arbitrary.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were justified for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit with a solvent aid.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Medium	There are minor reservations about the source of test organisms.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Organism environmental conditions were conducive to maintenance of health.	
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Study Citation:	Yang, Z. H., Zhang, X. J., Cai, Z. H. (2009). Toxic effects of several phthalate esters on the embryos and larvae of abalone <i>Haliotis diversicolor supertexta</i> . Chinese Journal of Oceanology and Limnology 27(2):395-399.
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Mollusks; <i>Haliotis diversicolor supertexta</i> ; Embryo
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	1322103

Domain	Metric	Rating	Comments
	Metric 17: Outcome Assessment Methodology	Medium	The outcome assessment methodology partially addressed the intended outcomes of interest, not all organisms were examined.
	Metric 18: Consistency of Outcome Assessment	Low	Somewhat subjective assessments were made, the criteria for abnormal development was not well defined.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups unrelated to test substance exposure.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22: Reporting of Data	Low	Abnormal was not well defined, and it was unclear which abnormalities were assessed.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: None

Overall Quality Determination**Medium**

Study Citation:	Tagatz, M. E., Deans, C. H., Moore, J. C., Plaia, G. R. (1983). Alterations in composition of field-developed and laboratory-developed estuarine benthic communities exposed to di-normal-butyl phthalate. <i>Aquatic Toxicology</i> 3(3):239-248.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Mollusks; <i>Mollusca</i> ; multiple species; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5495608			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	High	The test substance was identified using electron-capture gas-liquid chromatography.	
	Metric 3: Test Substance Purity	Low	Purity of the test substance was 100%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how initial placement of aquaria was determined.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The experimental system and/or test media preparation methods were adequately reported but there is some concern over the use of plastic trays.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Medium	Naturally colonizing organisms were used, and it is assumed there was equal initial population structure.	
	Metric 14: Acclimatization and Pretreatment Conditions	Medium	All pretreatment conditions were most likely the same for control and exposed organisms.	
	Metric 15: Number of Organisms and Replicates per Group	Low	The initial number of test organisms was not reported.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.	
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Study Citation:	Tagatz, M. E., Deans, C. H., Moore, J. C., Plaia, G. R. (1983). Alterations in composition of field-developed and laboratory-developed estuarine benthic communities exposed to di-normal-butyl phthalate. <i>Aquatic Toxicology</i> 3(3):239-248.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Mollusks; <i>Mollusca</i> ; multiple species; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	5495608

Domain	Metric	Rating	Comments
	Metric 17: Outcome Assessment Methodology	Low	The outcome assessment methodology (harvesting) was not clearly reported.
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.

Additional Comments: None

Overall Quality Determination**Medium**

Study Citation:	Tagatz, M. E., Deans, C. H., Moore, J. C., Plaia, G. R. (1983). Alterations in composition of field-developed and laboratory-developed estuarine benthic communities exposed to di-normal-butyl phthalate. <i>Aquatic Toxicology</i> 3(3):239-248.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Mollusks; <i>Mollusca</i> ; multiple species; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5495608			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	High	The test substance was identified using electron-capture gas-liquid chromatography.	
	Metric 3: Test Substance Purity	Low	Purity of the test substance was 100%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how initial placement of aquaria was determined.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The experimental system and/or test media preparation methods were adequately reported but there is some concern over the use of plastic trays.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Medium	Naturally colonizing organisms were used, and it is assumed there was equal initial population structure.	
	Metric 14: Acclimatization and Pretreatment Conditions	Medium	All pretreatment conditions were most likely the same for control and exposed organisms.	
	Metric 15: Number of Organisms and Replicates per Group	Low	The initial number of test organisms was not reported.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.	
	Metric 17: Outcome Assessment Methodology	Low	The outcome assessment methodology (harvesting) was not clearly reported.	
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.	

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Study Citation:	Tagatz, M. E., Deans, C. H., Moore, J. C., Plaia, G. R. (1983). Alterations in composition of field-developed and laboratory-developed estuarine benthic communities exposed to di-normal-butyl phthalate. <i>Aquatic Toxicology</i> 3(3):239-248.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Mollusks; <i>Mollusca</i> ; multiple species; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	5495608

Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.

Additional Comments: None

Overall Quality Determination**Medium**

Study Citation:	Adams, W. J., Biddinger, G. R., Robillard, K. A., Gorsuch, J. W. (1995). A summary of the acute toxicity of 14 phthalate esters to representative aquatic organisms. Environmental Toxicology and Chemistry 14(9):1569-1574.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Mysidopsis bahia</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1321996; Linked HERO ID(s): 1321996, 1316224			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The test substance nomenclature was reported without a CASRN.	
	Metric 2: Test Substance Source	Low	The source was reported as provided by manufacturer from commercially available batches. Manufacture name and batch number were not provided. No analytical data was reported.	
	Metric 3: Test Substance Purity	High	The test substance was at least 95% pure.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	A negative control was reported.	
	Metric 5: Negative Control Response	High	The control response was acceptable.	
	Metric 6: Randomized Allocation	Low	An allocation method was not reported.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The experimental system was well described. However, headspace or measures to prevent volatilization were not reported.	
	Metric 8: Consistency of Exposure Administration	High	Exposure administration was consistent across groups.	
	Metric 9: Measurement of Test Substance Concentration	Medium	Sample extracts were analyzed by gas chromatography at the start and end of the test. In static studies, final test concentrations frequently were 50% of the initial concentrations. Loss of the phthalate esters was thought to be principally due to adsorption to the test vessels.	
	Metric 10: Exposure Duration and Frequency	High	Duration and frequency of exposure were appropriate for the test.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	Exposure levels were appropriate. A range finding test was performed.	
	Metric 12: Testing at or Below Solubility Limit	High	The test was performed at or below water solubility.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	A source was not reported.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	An appropriate acclimation for the test was reported.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	Each test was performed using duplicate test concentrations with 10 organisms per test vessel.	
Domain 5: Outcome Assessment				
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Study Citation:	Adams, W. J., Biddinger, G. R., Robillard, K. A., Gorsuch, J. W. (1995). A summary of the acute toxicity of 14 phthalate esters to representative aquatic organisms. <i>Environmental Toxicology and Chemistry</i> 14(9):1569-1574.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Mysidopsis bahia</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	1321996; Linked HERO ID(s): 1321996, 1316224

Domain	Metric	Rating	Comments
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions were appropriate for the test.
	Metric 17: Outcome Assessment Methodology	High	Intended outcomes were reported.
	Metric 18: Consistency of Outcome Assessment	High	Outcome assessment was consistent across groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	Environmental conditions were consistent across groups.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There were no reported differences between groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were performed and described.
	Metric 22: Reporting of Data	Medium	Only treatment endpoints were reported.
	Metric 23: Explanation of Unexpected Outcomes	High	No unexpected outcomes were reported.
Additional Comments: None			

Overall Quality Determination**High**

Study Citation:	Bionomics,, EG&G (1984). Acute toxicity of twelve phthalate esters to mysid shrimp (Mysidopsis bahia).			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (marine), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Mysidopsis bahia</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1316220			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
Metric 1:	Test Substance Identity	Low	The test substance was identified by chemical name, but no CASRN or structure were reported.	
Metric 2:	Test Substance Source	High	The source of the phthalates was Bionomics Aquatic Toxicology Laboratory (Wareham, MA).	
Metric 3:	Test Substance Purity	Low	Purity and/or substance were not included in the study.	
Domain 2: Test Design				
Metric 4:	Negative Controls	High	Authors reported using negative controls.	
Metric 5:	Negative Control Response	High	The response of the negative controls was adequate.	
Metric 6:	Randomized Allocation	Medium	Mysid shrimp were maintained 1-3 days before they were distributed into test vessels.	
Domain 3: Exposure Characterization				
Metric 7:	Experimental System/Test Media Preparation	High	The experimental design followed protocol guidelines.	
Metric 8:	Consistency of Exposure Administration	High	Authors reported consistent administration.	
Metric 9:	Measurement of Test Substance Concentration	High	Phthalates were analytically verified and measured.	
Metric 10:	Exposure Duration and Frequency	High	The test duration followed protocol.	
Metric 11:	Number of Exposure Groups/Spacing of Exposure Levels	High	The number of replicates used was adequate to the guidelines.	
Metric 12:	Testing at or Below Solubility Limit	High	Concentrations used in the analysis were below the solubility limit.	
Domain 4: Test Organism				
Metric 13:	Test Organism Characteristics	Medium	The source of organisms was reported, but details beyond that were not.	
Metric 14:	Acclimatization and Pretreatment Conditions	High	Organisms were housed for 1-3 days prior to treatment.	
Metric 15:	Number of Organisms and Replicates per Group	Medium	The replicates followed protocol.	
Domain 5: Outcome Assessment				
Metric 16:	Adequacy of Test Conditions	High	Environmental conditions were adequate and described in detail.	
Metric 17:	Outcome Assessment Methodology	High	Outcomes were reported and addressed.	
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Study Citation:	Bionomics,, EG&G (1984). Acute toxicity of twelve phthalate esters to mysid shrimp (<i>Mysidopsis bahia</i>).			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (marine), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Mysidopsis bahia</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1316220			
Domain		Metric	Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed and reported.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	No differences were reported.
	Metric 20:	Outcomes Unrelated to Exposure	High	No outcomes unrelated to exposure were reported.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistics performed included moving average angle analysis, probit analysis, and binomial probability.
	Metric 22:	Reporting of Data	High	Data was reported adequately.
	Metric 23:	Explanation of Unexpected Outcomes	High	No unexpected outcomes were reported.
Additional Comments:	None			

Overall Quality Determination**High**

Study Citation:	RB, Laughlin, J. R., Neff, J. M., Hrung, Y. C., Goodwin, T. C., Giam, C. S. (1978). The effects of three phthalate esters on the larval development of the grass shrimp <i>Palaemonetes pugio</i> (Holthuis). <i>Water, Air, and Soil Pollution</i> 9(3):323-336.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>PALAEMONETES PUGIO</i> ; Larvae			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1333217			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The chemical was identified by accepted name [di-n-butyl phthalate (DBP)].
	Metric 2:	Test Substance Source	Low	The source was not reported.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of the test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent solvent control group.
	Metric 5:	Negative Control Response	High	The biological response of the solvent control group was adequate.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations, and differences from nominal values varied considerably.
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported, and exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were sampled at 0 hours and after 24 hours. They were measured via gas chromatography and were not similar to nominal concentrations.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were adequate for a dose response.
	Metric 12:	Testing at or Below Solubility Limit	Low	Exposure concentrations were at or below the water solubility limit, however droplets of chemical were noticed in test chambers.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	There are minor reservations – test organisms were collected from the wild, with minimal characteristic information given: "Gravid female <i>Palaemonetes pugio</i> were collected from salt marshes at the eastern end of Galveston Island, Texas. Separate collections were made between June and October, 1976, for testing each phthalate ester. Previous observations have established that the overall health and viability of adults do not vary significantly during this time of year (Tatem et al., 1976)."
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.
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Study Citation:	RB, Laughlin, J. R., Neff, J. M., Hrung, Y. C., Goodwin, T. C., Giam, C. S. (1978). The effects of three phthalate esters on the larval development of the grass shrimp <i>Palaemonetes pugio</i> (Holthuis). <i>Water, Air, and Soil Pollution</i> 9(3):323-336.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>PALAEMONETES PUGIO</i> ; Larvae
Health Outcome:	ADME (biotransformation)
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	1333217

Domain	Metric	Rating	Comments
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects (75 larvae per concentration with three replicates).
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	Low	Reporting environmental conditions was not sufficiently reported to evaluate if adequate.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability, some variability was considered to be attributed to incomplete dissolution of test chemical.

Additional Comments: This form is for the ADME outcome with DBP.

Overall Quality Determination

Medium

Study Citation:	RB, Laughlin, J. R., Neff, J. M., Hrung, Y. C., Goodwin, T. C., Giam, C. S. (1978). The effects of three phthalate esters on the larval development of the grass shrimp <i>Palaemonetes pugio</i> (Holthuis). <i>Water, Air, and Soil Pollution</i> 9(3):323-336.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>PALAEMONETES PUGIO</i> ; Larvae			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1333217			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by accepted name [di-n-butyl phthalate (DBP)]
	Metric 2:	Test Substance Source	Low	The source was not reported
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent solvent control group
	Metric 5:	Negative Control Response	High	The biological response of the solvent control group was adequate
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations, differences from nominal values varied considerably
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were sampled at 0hr and after 24hr and measured via gas chromatography but were not similar to nominal concentrations
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were adequate for a dose response
	Metric 12:	Testing at or Below Solubility Limit	Low	Exposure concentrations were at or below the water solubility limit, however droplets of chemical were noticed in test chambers
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	There are minor reservations – test organisms were collected from the wild, with minimal characteristic information given:”Gravid female <i>Palaemonetes pugio</i> were collected from salt marshes at the eastern end of Galveston Island, Texas. Separate collections were made between June and October, 1976, for testing each phthalate ester. Previous observations have established that the overall health and viability of adults do not vary significantly during this time of year (Tatem et al., 1976).”
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects (75 larvae per concentration, three replicates)
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Study Citation:	RB, Laughlin, J. R., Neff, J. M., Hrung, Y. C., Goodwin, T. C., Giam, C. S. (1978). The effects of three phthalate esters on the larval development of the grass shrimp <i>Palaemonetes pugio</i> (Holthuis). Water, Air, and Soil Pollution 9(3):323-336.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>PALAEMONETES PUGIO</i> ; Larvae
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	1333217

Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	Low	Reporting environmental conditions was not sufficiently reported to evaluate if adequate
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Statistical analysis was performed but not described adequately
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23: Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability, some variability was considered to be attributed to incomplete dissolution of test chemical
Additional Comments: None			

Overall Quality Determination**Medium**

Study Citation:	RB, Laughlin, J. R., Neff, J. M., Hrung, Y. C., Goodwin, T. C., Giam, C. S. (1978). The effects of three phthalate esters on the larval development of the grass shrimp <i>Palaemonetes pugio</i> (Holthuis). <i>Water, Air, and Soil Pollution</i> 9(3):323-336.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>PALAEMONETES PUGIO</i> ; Larvae			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1333217			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by accepted name [di-n-butyl phthalate (DBP)]
	Metric 2:	Test Substance Source	Low	The source was not reported
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent solvent control group
	Metric 5:	Negative Control Response	High	The biological response of the solvent control group was adequate
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations, differences from nominal values varied considerably
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were sampled at 0hr and after 24hr and measured via gas chromatography but were not similar to nominal concentrations
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were adequate for a dose response
	Metric 12:	Testing at or Below Solubility Limit	Low	Exposure concentrations were at or below the water solubility limit, however droplets of chemical were noticed in test chambers
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	There are minor reservations – test organisms were collected from the wild, with minimal characteristic information given:”Gravid female <i>Palaemonetes pugio</i> were collected from salt marshes at the eastern end of Galveston Island, Texas. Separate collections were made between June and October, 1976, for testing each phthalate ester. Previous observations have established that the overall health and viability of adults do not vary significantly during this time of year (Tatem et al., 1976).”
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects (75 larvae per concentration, three replicates)
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Study Citation:	RB, Laughlin, J. R., Neff, J. M., Hrung, Y. C., Goodwin, T. C., Giam, C. S. (1978). The effects of three phthalate esters on the larval development of the grass shrimp <i>Palaemonetes pugio</i> (Holthuis). Water, Air, and Soil Pollution 9(3):323-336.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>PALAEMONETES PUGIO</i> ; Larvae
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	1333217

Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	Low	Reporting environmental conditions was not sufficiently reported to evaluate if adequate
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Statistical analysis was performed but not described adequately
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23: Explanation of Unexpected Outcomes	Low	Some variability was considered to be attributed to incomplete dissolution of test chemical
Additional Comments: None			

Overall Quality Determination**Medium**

Study Citation:	Clark, J. R., JR, Patrick, J. M., Jr, More, J. C., Lores, E. M. (1987). Waterborne and sediment-source toxicities of six organic chemicals to grass shrimp (<i>Palaemonetes pugio</i>) and amphioxus (<i>Branchiostoma caribaeum</i>). Archives of Environmental Contamination and Toxicology 16(4):401-408.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Palaemonetes pugio</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5557723			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The chemical was identified by name only.
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Medium	The authors state that "reagent grade" test chemicals were purchased from chemical supply companies.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	Low	The data from the biological response of the negative control groups was not reported.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The experimental system and methods for preparation of test media were described in adequate detail, but there is some concern over solubility. Glass beakers and jars were used in the bioassays.
	Metric 8:	Consistency of Exposure	High	Exposures were administered consistently across study groups.
	Metric 9:	Administration Measurement of Test Substance Concentration	Low	Ranges of measured concentrations across a variety of experiments were reported, but the methods used to make these measurements were not reported.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	The study does not report the number of exposure groups, what the exposure levels were, or the spacing of exposure levels. Only the highest concentration is reported in the results.
	Metric 12:	Testing at or Below Solubility Limit	Low	The solvent concentration and biological response were not reported.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The test organisms were wild caught, with only a few in lab generations cultured.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of test organisms was not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.

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Study Citation:	Clark, J. R., JR, Patrick, J. M., Jr, More, J. C., Lores, E. M. (1987). Waterborne and sediment-source toxicities of six organic chemicals to grass shrimp (<i>Palaemonetes pugio</i>) and amphioxus (<i>Branchiostoma caribaeum</i>). Archives of Environmental Contamination and Toxicology 16(4):401-408.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Palaemonetes pugio</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5557723			
Domain	Metric	Rating	Comments	
	Metric 17:	Outcome Assessment Methodology	Low	The intended outcome was mortality and was straightforwardly stated, but the details of the methodology such as the exposure concentrations, or the spacing of the exposure levels were not reported.
	Metric 18:	Consistency of Outcome Assessment	Low	
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described.
	Metric 22:	Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group.
	Metric 23:	Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability.
Additional Comments:	The study does not report the test’s concentrations of exposure. It only reports that no mortality (the endpoint of interest) occurred.			
Overall Quality Determination		Low		

Study Citation:	Clark, J. R., JR, Patrick, J. M., Jr, More, J. C., Lores, E. M. (1987). Waterborne and sediment-source toxicities of six organic chemicals to grass shrimp (<i>Palaemonetes pugio</i>) and amphioxus (<i>Branchiostoma caribaeum</i>). Archives of Environmental Contamination and Toxicology 16(4):401-408.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Aquatic (marine); Sediment; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Palaemonetes pugio</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5557723			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Medium	The authors state that "reagent grade" test chemicals were purchased from chemical supply companies.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	Low	The biological response of the negative control group was not reported.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The experimental system and methods for preparation of test media were described in adequate detail, but there is some concern over solubility. Glass containers were used.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	Low	Measured exposure concentrations were not reported.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Low	The study does not report the number of exposure groups, what the exposure levels were, or the spacing of exposure levels. Only the highest concentration is reported in the results.	
	Metric 12: Testing at or Below Solubility Limit	Medium	The highest concentration was below solubility.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Medium	The organisms were wild caught, with only a few in lab generations cultured.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.	
	Metric 15: Number of Organisms and Replicates per Group	Low	The number of test organisms was not reported, but duplicates were used.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.	
	Metric 17: Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported.	

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Study Citation:	Clark, J. R., JR, Patrick, J. M., Jr, More, J. C., Lores, E. M. (1987). Waterborne and sediment-source toxicities of six organic chemicals to grass shrimp (<i>Palaemonetes pugio</i>) and amphioxus (<i>Branchiostoma caribaeum</i>). Archives of Environmental Contamination and Toxicology 16(4):401-408.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (marine); Sediment; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Palaemonetes pugio</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	5557723

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability.

Additional Comments: The study did not report the exposure concentrations at all. No mortality was observed at all.

Overall Quality Determination

Low

Study Citation:	Tagatz, M. E., Deans, C. H., Moore, J. C., Plaia, G. R. (1983). Alterations in composition of field-developed and laboratory-developed estuarine benthic communities exposed to di-normal-butyl phthalate. <i>Aquatic Toxicology</i> 3(3):239-248.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Other Invertebrate (e.g., sea urchins, ciliates, rotifers); <i>Rhynchocoela</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5495608			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	High	The test substance was identified using electron-capture gas-liquid chromatography.	
	Metric 3: Test Substance Purity	Low	Purity of the test substance was 100%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how initial placement of aquaria was determined.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The experimental system and/or test media preparation methods were adequately reported but there is some concern over the use of plastic trays.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Medium	Naturally colonizing organisms were used, and it is assumed there was equal initial population structure.	
	Metric 14: Acclimatization and Pretreatment Conditions	Medium	All pretreatment conditions were most likely the same for control and exposed organisms.	
	Metric 15: Number of Organisms and Replicates per Group	Low	The initial number of test organisms was not reported.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.	
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Study Citation:	Tagatz, M. E., Deans, C. H., Moore, J. C., Plaia, G. R. (1983). Alterations in composition of field-developed and laboratory-developed estuarine benthic communities exposed to di-normal-butyl phthalate. <i>Aquatic Toxicology</i> 3(3):239-248.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Other Invertebrate (e.g., sea urchins, ciliates, rotifers); <i>Rhynchocoela</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	5495608

Domain	Metric	Rating	Comments
	Metric 17: Outcome Assessment Methodology	Low	The outcome assessment methodology (harvesting) was not clearly reported.
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.

Additional Comments: None

Overall Quality Determination**Medium**

Study Citation:	Acey, R., Healy, P., Unger, T. F., Ford, C. E., Hudson, R. A. (1987). Growth and aggregation behavior of representative phytoplankton as affected by the environmental contaminant di-n-butyl phthalate. Bulletin of Environmental Contamination and Toxicology 39(1):1-6.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Dunaliella parva</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	790153			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The test substance was identified by name only.	
	Metric 2: Test Substance Source	High	The source of the DBP was reported to be Sigma Aldrich in St. Louis, MO. The DBP was analyzed by HPLC to be sure no monohydrolysis had occurred.	
	Metric 3: Test Substance Purity	High	The DBP was reported to be an analytical reagent grade.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of a concurrent negative control.	
	Metric 5: Negative Control Response	High	The negative control response was reported in Figure 1 and was adequate for the outcome of interest.	
	Metric 6: Randomized Allocation	Low	It was not reported how the algae was allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	Limited details were provided on the preparation of the DBP concentrations and media.	
	Metric 8: Consistency of Exposure Administration	Low	Little information was provided on the exposure, so it is difficult to determine if the exposures were administered consistently. The number of organisms added to each exposure were just reported to be within a range. The exact time duration was not reported for each test. It appeared that organisms counts were not always performed on the same day or at the same time from Figure 1.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if the exposure concentrations were measured throughout the study.	
	Metric 10: Exposure Duration and Frequency	Uninformative	The exposure duration for each test was not reported. Exposure durations can be estimated from Figure 1, but not all counts appeared to be conducted at the same time.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Medium	There were only two exposure concentrations. This is less than is typical, but it was enough for a response.	
	Metric 12: Testing at or Below Solubility Limit	High	Study authors reported that all test concentrations were below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	Test organisms were reported to be either from the Marine Biology Laboratory of California State University Long Beach stock cultures or from Carolina Biological Supply.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the organisms were acclimated prior to the study.	
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Study Citation:	Acey, R., Healy, P., Unger, T. F., Ford, C. E., Hudson, R. A. (1987). Growth and aggregation behavior of representative phytoplankton as affected by the environmental contaminant di-n-butyl phthalate. Bulletin of Environmental Contamination and Toxicology 39(1):1-6.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Dunaliella parva</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	790153			
Domain		Metric	Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Low	Test organism concentrations were reported to be somewhere between 200 and 700 organisms/uL. This was quite a bit of variation. All tests were performed in triplicate.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Test organisms were grown in continuous light at 26C in F2 media modified with artificial seawater. More information on light intensity would be ideal.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—growth of the organisms in terms of aggregation and growth rate.
	Metric 18:	Consistency of Outcome Assessment	Medium	Organisms were counted with a hemocytometer. It appeared from Figure 1 that not all concentrations were counted at the same time.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Uninformative	Study authors did not conduct statistical analysis. Data is in graph form and exact numbers are hard to determine.
	Metric 22:	Reporting of Data	High	Exposure data and control response data are reported in Figure 1 and appear adequate for the outcome of interest.
	Metric 23:	Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability.
Additional Comments:	This portion of the evaluation is on the growth of green algae <i>D. parva</i> during DBP exposure. Development/growth was chosen as the outcome. The study received an unacceptable ranking due to lack of information regarding exposure duration and due to lack of statistical analysis.			

Overall Quality Determination**Uninformative**

Study Citation:	Liu, N., Wen, F., Li, F., Zheng, X., Liang, Z., Zheng, H. (2016). Inhibitory mechanism of phthalate esters on <i>Karenia brevis</i> . Chemosphere 155:498-508.		
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)		
Exposure Route, Media, Path:	Aquatic (marine); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Karenia brevis</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Development/Growth		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	3230225		
Domain	Metric	Rating	Comments
Domain 1: Test Substance			
Metric 1:	Test Substance Identity	Low	Relevant test materials are described as di-iso-butyl ortho-phthalate (DIBP), benzyl-n-butyl ortho-phthalate (BBP), Di-n-butylortho-phthalate (DBP), and bis(2-ethylhexyl)ortho-phthalate(DEHP). No further details are provided.
Metric 2:	Test Substance Source	Low	Chemicals are sourced by the Sigma Company, no additional information provided.
Metric 3:	Test Substance Purity	High	purity is reported as > 99%
Domain 2: Test Design			
Metric 4:	Negative Controls	High	Negative and solvent controls were used. The authors indicated that no significant differences were observed between the solvent controls (acetone concentration (0.5 mL L-1)) and the negative control.
Metric 5:	Negative Control Response	High	No adverse effects reported in the control.
Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization			
Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations and minimize loss of test substance before and during the exposure for these degradable substances. A solvent (acetone) was used to facilitate the preparation of the stock solution.
Metric 8:	Consistency of Exposure Administration	Low	Reporting omissions are likely to have a substantial impact on results. No measurement of test material concentration was conducted at the end of the test, so the actual exposure concentration was uncertain. Similarly, the different biodegradation rates of the chemicals meant that exposure concentrations may have differed because of degradation, but this was not discussed.
Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured or measurements were not reported. As degradation is likely to be observed, the reported nominal concentrations are not likely to be representative of the final concentration and reporting in terms of nominal concentrations may underestimate the effects observed.
Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type and/or outcome(s) of interest (96-hour algae growth inhibition test).
Metric 11:	Number of Exposure Groups/Spacing of Exposure Levels	Medium	Exposure concentrations were reported to be 0, 1, 5, 10, 20, 30, 50, 100, 150, 200 mL/L. These appear to reflect the nominal concentration and no final test quantification was conducted, so these are likely not representative of the actual exposure concentration throughout the test. The number of exposure groups and spacing of exposure levels were adequate to show results relevant to the outcome of interest .
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Study Citation:	Liu, N., Wen, F., Li, F., Zheng, X., Liang, Z., Zheng, H. (2016). Inhibitory mechanism of phthalate esters on <i>Karenia brevis</i> . Chemosphere 155:498-508.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (marine); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Karenia brevis</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3230225			
Domain	Metric	Rating	Comments	
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate (the authors demonstrated that no significant difference (p > 0.05) was observed between the growth in controls and acetone treatment.).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test animals was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized or whether pretreatment conditions were the same for control and exposed groups.
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of test organisms (starting algal density) was not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	The algal cells were cultured in a GXZ-380Z intelligent illumination incubator under the following conditions: 14 h of light at 24C (4000 ± 500 lux) and 10 h of dark at 22C. The medium was shaken once daily to prevent cell adherence growth.
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported; it was unclear whether methods were sensitive for the outcome of interest. The authors reported that significant effects were observed on growth following exposure to some of the test materials, but did not provide the measures of significance for each test concentration, so it was not clear what levels elicited these inhibitions on growth. There appeared to be a dose-response based on the graphs provided. This is likely to have a substantial impact on results.
	Metric 18:	Consistency of Outcome Assessment	High	Outcome assessments appears to be consistent across group. Cell numbers were determined every 24 hours.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure (e.g., infection) that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	One-way ANOVA was adopted to determine the significant differences between experimental and control groups. The calculations and measures of significance were not provided so no conclusions about a dose response could be made.
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Study Citation:	Liu, N., Wen, F., Li, F., Zheng, X., Liang, Z., Zheng, H. (2016). Inhibitory mechanism of phthalate esters on <i>Karenia brevis</i> . Chemosphere 155:498-508.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (marine); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Karenia brevis</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	3230225

Domain	Metric	Rating	Comments
	Metric 22: Reporting of Data	High	Data for exposure-related findings were shown for each treatment and control group (Figure 1). The author's discussion of the growth inhibition data for each phthalate was not clear, and their conclusions were made without incorporating any discussion of statistical significance.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes, or unexpected outcomes were satisfactorily explained.

Additional Comments: The discussion of growth inhibition following exposure to DEHP, DIBP, BBP and DBP was lacking.

Overall Quality Determination

Low

Study Citation:	Liu, N., Wen, F., Li, F., Zheng, X., Liang, Z., Zheng, H. (2016). Inhibitory mechanism of phthalate esters on <i>Karenia brevis</i> . Chemosphere 155:498-508.		
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)		
Exposure Route, Media, Path:	Aquatic (marine); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Karenia brevis</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)-Photosynthesis		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	3230225		
Domain	Metric	Rating	Comments
Domain 1: Test Substance			
	Metric 1: Test Substance Identity	Low	Relevant test materials are described as di-iso-butyl ortho-phthalate (DIBP), benzyl-n-butyl ortho-phthalate (BBP), Di-n-butylortho-phthalate (DBP), and bis(2-ethylhexyl)ortho-phthalate(DEHP). No further details are provided.
	Metric 2: Test Substance Source	Low	Chemicals are sourced by the Sigma Company, and no additional information was provided.
	Metric 3: Test Substance Purity	High	Purity is reported as > 99%.
Domain 2: Test Design			
	Metric 4: Negative Controls	High	Negative and solvent controls were used. The authors indicated that no significant differences were observed between the solvent controls (acetone concentration (0.5 mL L-1)) and the negative control.
	Metric 5: Negative Control Response	High	The biological response of the control group was reported for mechanistic endpoints, and was acceptable.
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization			
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations and minimize loss of test substance before and during the exposure for these degradable substances. A solvent (acetone) was used to facilitate the preparation of the stock solution.
	Metric 8: Consistency of Exposure Administration	Low	Reporting omissions are likely to have a substantial impact on results. No measurement of test material concentration was conducted at the end of the test, so the actual exposure concentration was uncertain. Similarly, the different biodegradation rates of the chemicals meant that exposure concentrations may have differed because of degradation, but this was not discussed.
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured or measurements were not reported. As degradation is likely to be observed, the reported nominal concentrations are not likely to be representative of the final concentration and reporting in terms of nominal concentrations may underestimate the effects observed.
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type and/or outcome(s) of interest (96-hour algae test).
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Medium	Exposure concentrations were reported to be 0,10, and 30 mL/L-1 and three replicates were used. These appear to reflect the nominal concentrations and no final test quantification was conducted, so these are likely not representative of the actual exposure concentrations throughout the test.
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Study Citation:	Liu, N., Wen, F., Li, F., Zheng, X., Liang, Z., Zheng, H. (2016). Inhibitory mechanism of phthalate esters on <i>Karenia brevis</i> . Chemosphere 155:498-508.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (marine); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Karenia brevis</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)-Photosynthesis			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3230225			
Domain	Metric	Rating	Comments	
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate (the authors demonstrated that no significant difference (p > 0.05) was observed between the growth in controls and acetone treatment.).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test organisms was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized or whether pretreatment conditions were the same for control and exposed groups.
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of test organisms (initial algal density) was not reported, and three replicates were used.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	The algal cells were cultured in a GXZ-380Z intelligent illumination incubator under the following conditions: 14 h of light at 24C (4000 ± 500 lux) and 10 h of dark at 22C. The medium was shaken once daily to prevent cell adherence growth.
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology (MDA content, SOD, CAT and free radicals) was not clearly reported. It was unclear whether methods were sensitive for the outcome of interest. The authors reported that significant effects were observed on MDA and antioxidant levels following exposure to some of the test materials, but did not provide the measures of significance for each test concentration, so it was not clear what levels elicited these inhibitions on growth. There appeared to be a dose-response based on the graphs provided, but limited test concentrations limit the utility of these conclusions. This is likely to have a substantial impact on results.
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment (e.g., timing of assessment across groups) were confusing. It was reported that algal cell numbers were counted but results were not provided.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure (e.g., infection) that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	One-way ANOVA was adopted to determine the significant differences between experimental and control groups. The calculations and measures of significance were not provided so no conclusions about a dose response could be made.

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Study Citation:	Liu, N., Wen, F., Li, F., Zheng, X., Liang, Z., Zheng, H. (2016). Inhibitory mechanism of phthalate esters on <i>Karenia brevis</i> . Chemosphere 155:498-508.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (marine); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Karenia brevis</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)-Photosynthesis
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	3230225

Domain	Metric	Rating	Comments
	Metric 22: Reporting of Data	Low	Data for exposure-related findings (MDA content, SOD, CAT, free radical concentration) were shown for each treatment and control group (Figures 3, 4, 5 and 6), but algal density data were not provided.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes, or unexpected outcomes were satisfactorily explained.

Additional Comments: The discussion of growth inhibition following exposure to DBP and BBP was lacking.

Overall Quality Determination

Low

Study Citation:	Medlin, L. K. (1980). Effects of di-n-butyl phthalate and salinity on the growth of the diatom <i>Skeletonema costatum</i> . Bulletin of Environmental Contamination and Toxicology 25(1):75-78.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Skeletonema costatum</i> ; diatoms; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	789981			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The test substance was identified by name only.	
	Metric 2: Test Substance Source	Low	The source of the DBP was not reported.	
	Metric 3: Test Substance Purity	Low	The purity and grade were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of a concurrent negative control.	
	Metric 5: Negative Control Response	High	The negative control response was reported in Table 3.	
	Metric 6: Randomized Allocation	Low	It was not reported how the diatoms were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	A 0.2mL portion of DBP was added to each salinity level of NH-15. Dilutions of this were used to make the DBP concentrations. It is unclear if the 0.2mL portion of the DBP was prepared in lab or if it was manufactured that way. Experiments were conducted in 16 x 125 mm Pyrex brand, disposable test tubes with a polypropylene cap.	
	Metric 8: Consistency of Exposure Administration	Medium	All exposures were conducted in 16 x 125mm test tubes at 24c in a growth chamber with continuous light. The concentration of diatoms was not reported, leaving uncertainties in diatom concentration for each replicate. It was only stated that 5 ml of <i>S. costatum</i> culture in logarithmic growth phase was added to each dilution.	
	Metric 9: Measurement of Test Substance Concentration	Medium	It was reported that test concentrations were frozen and later analyzed to determine DBP concentrations. The methods used were not reported, and the actual concentrations were not reported. Concentrations were only reported as % of saturation.	
	Metric 10: Exposure Duration and Frequency	High	The exposure was reported to be for 4d, which is typical of this type of study.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Low	There were seven exposure levels for each salinity and a control. The spacing was adequate to see a response. Study authors did not report actual exposure concentrations. Concentrations were reported as percentage of saturation. The stock solution was reported to be saturated with DBP, but the exact concentration was not reported.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit. The stock solution was reported to be saturated with DBP, and thus would not have been over the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the diatoms was not reported.	
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Study Citation:	Medlin, L. K. (1980). Effects of di-n-butyl phthalate and salinity on the growth of the diatom <i>Skeletonema costatum</i> . Bulletin of Environmental Contamination and Toxicology 25(1):75-78.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Skeletonema costatum</i> ; diatoms; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	789981			
Domain	Metric	Rating	Comments	
	Metric 14:	Acclimatization and Pretreatment Conditions	High	It was reported that the diatom cultures were "preconditioned" to each salinity level prior to the study. The concentration of diatoms used in each test chamber was not reported. There were three replicates for each treatment level.
	Metric 15:	Number of Organisms and Replicates per Group	Low	
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Cultures were kept at 24C with continuous light at the appropriate salinity for the study being conducted. Artificial seawater (NH-15) was used. The culture medium used for nutrition was not reported.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—development/growth.
	Metric 18:	Consistency of Outcome Assessment	High	Relative chlorophyll concentrations were measured at the start of the test and daily thereafter using a fluorometer. Cultures were fixed and initial and final cell counts were taken as well.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Medium	There could potentially have been differences in diatom concentrations between study groups as this was not reported in the paper.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Data were fitted to a quadratic model. A two-way analysis of variance and the student t-test was used to analyze the data.
	Metric 22:	Reporting of Data	High	Data was presented for all exposure concentrations and salinities as well as for the control in Table 3. Figure 1 presented data for exposure responses only.
	Metric 23:	Explanation of Unexpected Outcomes	Low	The study did not report measures of variability.
Additional Comments:	This study was on the effect of DBP on diatom growth at different salinities. A new evaluation was created for each salinity. This evaluation is for the 14 0/00 salinity.			
Overall Quality Determination		Medium		

Study Citation:	Medlin, L. K. (1980). Effects of di-n-butyl phthalate and salinity on the growth of the diatom <i>Skeletonema costatum</i> . Bulletin of Environmental Contamination and Toxicology 25(1):75-78.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Skeletonema costatum</i> ; diatoms; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	789981			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The test substance was identified by name only.
	Metric 2:	Test Substance Source	Low	The source of the DBP was not reported.
	Metric 3:	Test Substance Purity	Low	The purity and grade were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of a concurrent negative control.
	Metric 5:	Negative Control Response	High	The negative control response was reported in Table 3.
	Metric 6:	Randomized Allocation	Low	It was not reported how the diatoms were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	A 0.2mL portion of DBP was added to each salinity level of NH-15. Dilutions of this were used to make the DBP concentrations. It is unclear if the 0.2mL portion of the DBP was prepared in lab or if it was manufactured that way. Experiments were conducted in 16 x 125 mm Pyrex brand, disposable test tubes with a polypropylene cap.
	Metric 8:	Consistency of Exposure Administration	Medium	All exposures were conducted in 16 x 125mm test tubes at 24c in a growth chamber with continuous light. The concentration of diatoms was not reported, leaving uncertainties in diatom concentration for each replicate.
	Metric 9:	Measurement of Test Substance Concentration	Medium	It was reported that test concentrations were frozen and later analyzed to determine DBP concentrations. The methods used were not reported, and the actual concentrations were not reported. Concentrations were only reported as % of saturation.
	Metric 10:	Exposure Duration and Frequency	High	The exposure was reported to be for 4d, which is typical of this type of study.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	There were seven exposure levels for each salinity and a control. The spacing was adequate to see a response. Study authors did not report actual exposure concentrations. Concentrations were reported as percentage of saturation. The stock solution was reported to be saturated with DBP, but the exact concentration was not reported.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit. The stock solution was reported to be saturated with DBP, and thus would not have been over the water solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the diatoms was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	It was reported that the diatom cultures were "preconditioned" to each salinity level prior to the study.
	Metric 15:	Number of Organisms and Replicates per Group	Low	The concentration of diatoms used in each test chamber was not reported. There were three replicates for each treatment level.

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Study Citation:	Medlin, L. K. (1980). Effects of di-n-butyl phthalate and salinity on the growth of the diatom <i>Skeletonema costatum</i> . Bulletin of Environmental Contamination and Toxicology 25(1):75-78.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Skeletonema costatum</i> ; diatoms; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	789981			
Domain	Metric	Rating	Comments	
Domain 5: Outcome Assessment				
Metric 16:	Adequacy of Test Conditions	Medium	Cultures were kept at 24C with continuous light at the appropriate salinity for the study being conducted. Artificial seawater (NH-15) was used. The culture medium used for nutrition was not reported.	
Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest–development/growth.	
Metric 18:	Consistency of Outcome Assessment	High	Relative chlorophyll concentrations were measured at the start of the test and daily thereafter using a fluorometer. Cultures were fixed and initial and final cell counts were taken as well.	
Domain 6: Confounding / Variable Control				
Metric 19:	Confounding Variables in Test Design and Procedures	Medium	There could potentially have been differences in diatom concentrations between study groups as this was not reported in the paper.	
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
Metric 21:	Statistical Methods	High	Data were fitted to a quadratic model. A two-way analysis of variance and the student t-test was used to analyze the data.	
Metric 22:	Reporting of Data	High	Data was presented for all exposure concentrations and salinities as well as for the control in Table 3. Figure 1 presented data for exposure responses only.	
Metric 23:	Explanation of Unexpected Outcomes	Low	The study did not report measures of variability.	
Additional Comments:	This study was on the effect of DBP on diatom growth at different salinities. A new evaluation was created for each salinity. This evaluation is for the 22 0/00 salinity.			

Overall Quality Determination**Medium**

Study Citation:	Medlin, L. K. (1980). Effects of di-n-butyl phthalate and salinity on the growth of the diatom <i>Skeletonema costatum</i> . Bulletin of Environmental Contamination and Toxicology 25(1):75-78.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Skeletonema costatum</i> ; diatoms; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	789981			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The test substance was identified by name only.	
	Metric 2: Test Substance Source	Low	The source of the DBP was not reported.	
	Metric 3: Test Substance Purity	Low	The purity and grade were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of a concurrent negative control.	
	Metric 5: Negative Control Response	High	The negative control response was reported in Table 3.	
	Metric 6: Randomized Allocation	Low	It was not reported how the diatoms were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	A 0.2mL portion of DBP was added to each salinity level of NH-15. Dilutions of this were used to make the DBP concentrations. It is unclear if the 0.2mL portion of the DBP was prepared in lab or if it was manufactured that way. Experiments were conducted in 16 x 125 mm Pyrex brand, disposable test tubes with a polypropylene cap.	
	Metric 8: Consistency of Exposure Administration	Medium	All exposures were conducted in 16 x 125mm test tubes at 24c in a growth chamber with continuous light. The concentration of diatoms was not reported, leaving uncertainties in diatom concentration for each replicate.	
	Metric 9: Measurement of Test Substance Concentration	Medium	It was reported that test concentrations were frozen and later analyzed to determine DBP concentrations. The methods used were not reported, and the actual concentrations were not reported. Concentrations were only reported as % of saturation.	
	Metric 10: Exposure Duration and Frequency	High	The exposure was reported to be for 4d, which is typical of this type of study.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Low	There were seven exposure levels for each salinity and a control. The spacing was adequate to see a response. Study authors did not report actual exposure concentrations. Concentrations were reported as percentage of saturation. The stock solution was reported to be saturated with DBP, but the exact concentration was not reported.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit. The stock solution was reported to be saturated with DBP, and thus would not have been over the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the diatoms was not reported.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	It was reported that the diatom cultures were "preconditioned" to each salinity level prior to the study.	
	Metric 15: Number of Organisms and Replicates per Group	Low	The concentration of diatoms used in each test chamber was not reported. There were three replicates for each treatment level.	

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Study Citation:	Medlin, L. K. (1980). Effects of di-n-butyl phthalate and salinity on the growth of the diatom <i>Skeletonema costatum</i> . Bulletin of Environmental Contamination and Toxicology 25(1):75-78.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Skeletonema costatum</i> ; diatoms; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	789981			
Domain	Metric	Rating	Comments	
Domain 5: Outcome Assessment				
Metric 16:	Adequacy of Test Conditions	Medium	Cultures were kept at 24C with continuous light at the appropriate salinity for the study being conducted. Artificial seawater (NH-15) was used. The culture medium used for nutrition was not reported.	
Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—development/growth.	
Metric 18:	Consistency of Outcome Assessment	High	Relative chlorophyll concentrations were measured at the start of the test and daily thereafter using a fluorometer. Cultures were fixed and initial and final cell counts were taken as well.	
Domain 6: Confounding / Variable Control				
Metric 19:	Confounding Variables in Test Design and Procedures	Medium	There could potentially have been differences in diatom concentrations between study groups as this was not reported in the paper.	
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
Metric 21:	Statistical Methods	High	Data were fitted to a quadratic model. A two-way analysis of variance and the student t-test was used to analyze the data.	
Metric 22:	Reporting of Data	High	Data for was presented for all exposure concentrations and salinities as well as for the control in Table 3. Figure 1 presented data for exposure responses only.	
Metric 23:	Explanation of Unexpected Outcomes	Low	The study did not report measures of variability.	
Additional Comments:	This study was on the effect of DBP on diatom growth at different salinities. A new evaluation was created for each salinity. This evaluation is for the 36 0/00 salinity.			

Overall Quality Determination**Medium**

Study Citation:	Medlin, L. K. (1980). Effects of di-n-butyl phthalate and salinity on the growth of the diatom <i>Skeletonema costatum</i> . Bulletin of Environmental Contamination and Toxicology 25(1):75-78.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Skeletonema costatum</i> ; diatoms; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	789981			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The test substance was identified by name only.
	Metric 2:	Test Substance Source	Low	The source of the DBP was not reported.
	Metric 3:	Test Substance Purity	Low	The purity and grade were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of a concurrent negative control.
	Metric 5:	Negative Control Response	High	The negative control response was reported in Table 3.
	Metric 6:	Randomized Allocation	Low	It was not reported how the diatoms were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	A 0.2mL portion of DBP was added to each salinity level of NH-15. Dilutions of this were used to make the DBP concentrations. It is unclear if the 0.2mL portion of the DBP was prepared in lab or if it was manufactured that way. Experiments were conducted in 16 x 125 mm Pyrex brand, disposable test tubes with a polypropylene cap.
	Metric 8:	Consistency of Exposure Administration	Medium	All exposures were conducted in 16 x 125mm test tubes at 24c in a growth chamber with continuous light. The concentration of diatoms was not reported, leaving uncertainties in diatom concentration for each replicate.
	Metric 9:	Measurement of Test Substance Concentration	Medium	It was reported that test concentrations were frozen and later analyzed to determine DBP concentrations. The methods used were not reported, and the actual concentrations were not reported. Concentrations were only reported as % of saturation.
	Metric 10:	Exposure Duration and Frequency	High	The exposure was reported to be for 4d, which is typical of this type of study.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	There were seven exposure levels for each salinity and a control. The spacing was adequate to see a response. Study authors did not report actual exposure concentrations. Concentrations were reported as percentage of saturation. The stock solution was reported to be saturated with DBP, but the exact concentration was not reported.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit. The stock solution was reported to be saturated with DBP, and thus would not have been over the water solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the diatoms was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	It was reported that the diatom cultures were "preconditioned" to each salinity level prior to the study.
	Metric 15:	Number of Organisms and Replicates per Group	Low	The concentration of diatoms used in each test chamber was not reported. There were three replicates for each treatment level.

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Study Citation:	Medlin, L. K. (1980). Effects of di-n-butyl phthalate and salinity on the growth of the diatom <i>Skeletonema costatum</i> . Bulletin of Environmental Contamination and Toxicology 25(1):75-78.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Skeletonema costatum</i> ; diatoms; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	789981			
Domain	Metric	Rating	Comments	
Domain 5: Outcome Assessment				
Metric 16:	Adequacy of Test Conditions	Medium	Cultures were kept at 24C with continuous light at the appropriate salinity for the study being conducted. Artificial seawater (NH-15) was used. The culture medium used for nutrition was not reported.	
Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest–development/growth.	
Metric 18:	Consistency of Outcome Assessment	High	Relative chlorophyll concentrations were measured at the start of the test and daily thereafter using a fluorometer. Cultures were fixed and initial and final cell counts were taken as well.	
Domain 6: Confounding / Variable Control				
Metric 19:	Confounding Variables in Test Design and Procedures	Medium	There could potentially have been differences in diatom concentrations between study groups as this was not reported in the paper.	
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
Metric 21:	Statistical Methods	High	Data were fitted to a quadratic model. A two-way analysis of variance and the student t-test was used to analyze the data.	
Metric 22:	Reporting of Data	High	Data for was presented for all exposure concentrations and salinities as well as for the control in Table 3. Figure 1 presented data for exposure responses only.	
Metric 23:	Explanation of Unexpected Outcomes	Low	The study did not report measures of variability.	
Additional Comments:	This study was on the effect of DBP on diatom growth at different salinities. A new evaluation was created for each salinity. This evaluation is for the 27 0/00 salinity.			

Overall Quality Determination**Medium**

Study Citation:	Acey, R., Healy, P., Unger, T. F., Ford, C. E., Hudson, R. A. (1987). Growth and aggregation behavior of representative phytoplankton as affected by the environmental contaminant di-n-butyl phthalate. Bulletin of Environmental Contamination and Toxicology 39(1):1-6.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Synechococcus lividus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	790153			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The test substance was identified by name only.	
	Metric 2: Test Substance Source	High	The source of the DBP was reported to be Sigma Aldrich in St. Louis, MO. The DBP was analyzed by HPLC to be sure no monohydrolysis had occurred.	
	Metric 3: Test Substance Purity	High	The DBP was reported to be an analytical reagent grade.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of a concurrent negative control.	
	Metric 5: Negative Control Response	High	The negative control response was reported in Figure 1 and in Table 1 and was adequate for the outcome of interest.	
	Metric 6: Randomized Allocation	Low	It was not reported how the diatoms were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	Limited details were provided on the preparation of the DBP concentrations and media.	
	Metric 8: Consistency of Exposure Administration	Low	Little information was provided on the exposure, so it is difficult to determine if the exposures were administered consistently. The number of organisms added to each exposure were just reported to be within a range. The exact time duration was not reported for each test. It appeared that organisms counts were not always performed on the same day or at the same time from Figure 1.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if the exposure concentrations were measured throughout the study.	
	Metric 10: Exposure Duration and Frequency	Uninformative	The exposure duration for each test was not reported. Exposure durations can be estimated from Figure 1, but not all counts appeared to be conducted at the same time.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	There were five exposure concentrations. This was adequate to observe a response. Spacing was adequate.	
	Metric 12: Testing at or Below Solubility Limit	High	Study authors reported that all test concentrations were below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	Test organisms were reported to be either from the Marine Biology Laboratory of California State University Long Beach stock cultures or from Carolina Biological Supply.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the organisms were acclimated prior to the study.	
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Study Citation:	Acey, R., Healy, P., Unger, T. F., Ford, C. E., Hudson, R. A. (1987). Growth and aggregation behavior of representative phytoplankton as affected by the environmental contaminant di-n-butyl phthalate. Bulletin of Environmental Contamination and Toxicology 39(1):1-6.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Synechococcus lividus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	790153			
Domain	Metric	Rating	Comments	
	Metric 15: Number of Organisms and Replicates per Group	Low	Test organism concentrations were reported to be somewhere between 200 and 700 organisms/uL. This was quite a bit of variation. All tests were performed in triplicate.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Medium	Test organisms were grown in continuous light at 26C in F2 media modified with artificial seawater. More information on light intensity would be ideal.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—growth of the organisms in terms of aggregation and growth rate.	
	Metric 18: Consistency of Outcome Assessment	Medium	Organisms were counted with a hemocytometer. It appeared from Figure 1 that not all concentrations were counted at the same time.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	Low	Study authors reported not conducting statistical analysis, but independent statistical analysis may be conducted from data in Table 1.	
	Metric 22: Reporting of Data	High	Exposure data and control response data are reported in Figure 1 and appear adequate for the outcome of interest.	
	Metric 23: Explanation of Unexpected Outcomes	Medium	Variability was reported in Table 1, but not in Figure 1.	
Additional Comments:	This portion of the evaluation is on the growth of blue-green algae <i>S. lividus</i> during DBP exposure. Development/growth was chosen as the outcome. The study received an unacceptable ranking due to lack of information regarding exposure duration.			

Overall Quality Determination**Uninformative**

Study Citation:	Acey, R., Healy, P., Unger, T. F., Ford, C. E., Hudson, R. A. (1987). Growth and aggregation behavior of representative phytoplankton as affected by the environmental contaminant di-n-butyl phthalate. Bulletin of Environmental Contamination and Toxicology 39(1):1-6.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Thalassioria pseudomona</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	790153			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The test substance was identified by name only.
	Metric 2:	Test Substance Source	High	The source of the DBP was reported to be Sigma Aldrich in St. Louis, MO. The DBP was analyzed by HPLC to be sure no monohydrolysis had occurred.
	Metric 3:	Test Substance Purity	High	The DBP was reported to be an analytical reagent grade.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of a concurrent negative control.
	Metric 5:	Negative Control Response	High	The negative control response was reported in Figure 1 and was adequate for the outcome of interest.
	Metric 6:	Randomized Allocation	Low	It was not reported how the diatoms were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	Limited details were provided on the preparation of the DBP concentrations and media.
	Metric 8:	Consistency of Exposure Administration	Low	Little information was provided on the exposure, so it is difficult to determine if the exposures were administered consistently. The number of organisms added to each exposure were just reported to be within a range. The exact time duration was not reported for each test. It appeared that organisms counts were not always performed on the same day or at the same time from Figure 1.
	Metric 9:	Measurement of Test Substance Concentration	Low	It was not reported if the exposure concentrations were measured throughout the study.
	Metric 10:	Exposure Duration and Frequency	Uninformative	The exposure duration for each test was not reported. Exposure durations can be estimated from Figure 1, but not all counts appeared to be conducted at the same time.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	There were four exposure concentrations. This was adequate to observe a response.
	Metric 12:	Testing at or Below Solubility Limit	High	Study authors reported that all test concentrations were below the water solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Test organisms were reported to be either from the Marine Biology Laboratory of California State University Long Beach stock cultures or from Carolina Biological Supply.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the organisms were acclimated prior to the study.

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Study Citation:	Acey, R., Healy, P., Unger, T. F., Ford, C. E., Hudson, R. A. (1987). Growth and aggregation behavior of representative phytoplankton as affected by the environmental contaminant di-n-butyl phthalate. Bulletin of Environmental Contamination and Toxicology 39(1):1-6.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Non-vascular Plants; <i>Thalassioria pseudomona</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	790153			
Domain		Metric	Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Low	Test organism concentrations were reported to be somewhere between 200 and 700 organisms/uL. This was quite a bit of variation. All tests were performed in triplicate.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Test organisms were grown in continuous light at 26C in F2 media modified with artificial seawater. More information on light intensity would be ideal.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—growth of the organisms in terms of aggregation and growth rate.
	Metric 18:	Consistency of Outcome Assessment	Medium	Organisms were counted with a hemocytometer. It appeared from Figure 1 that not all concentrations were counted at the same time.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Uninformative	Study authors did not conduct statistical analysis. Data is in graph form and exact numbers are hard to determine.
	Metric 22:	Reporting of Data	High	Exposure data and control response data are reported in Figure 1 and appear adequate for the outcome of interest.
	Metric 23:	Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability.
Additional Comments:	This portion of the evaluation is on the growth of the diatom T. pseudomona during DBP exposure. Development/growth was chosen as the outcome. The study received an unacceptable ranking due to lack of information regarding exposure duration and due to lack of statistical analysis.			

Overall Quality Determination**Uninformative**

Study Citation:	Wofford, H. W., Wilsey, C. D., Neff, G. S., Giam, C. S., Neff, J. M. (1981). Bioaccumulation and metabolism of phthalate esters by oysters, brown shrimp, and sheepshead minnows. <i>Ecotoxicology and Environmental Safety</i> 5(2):202-210.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Cyprinodon variegatus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	789995			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The chemical was identified by name, but no CASRN or structure were given.
	Metric 2:	Test Substance Source	High	The test substance source's include Aldrich Chemical Company for the unlabeled phthalate and California Bionuclear Corporation for the 14C-labeled phthalate.
	Metric 3:	Test Substance Purity	Low	The purity of the chemical was not included in the study.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Uninformative	No control was reported.
	Metric 5:	Negative Control Response	N/A	No control was reported.
	Metric 6:	Randomized Allocation	Low	The allocation method into groups was not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The experimental design for test preparation was described.
	Metric 8:	Consistency of Exposure Administration	High	No variations in exposure administration were reported.
	Metric 9:	Measurement of Test Substance Concentration	High	Concentrations were measured using analytical techniques, gas-liquid chromatography and liquid scintillation.
	Metric 10:	Exposure Duration and Frequency	Medium	A 24-hour exposure period was used.
	Metric 11:	Number of Exposure Groups/Spacing of Exposure Levels	Low	Standard deviations were reported as two.
	Metric 12:	Testing at or Below Solubility Limit	High	Two concentrations were below the solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	Characteristics were not described in the study.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Organisms were acclimatized for four days prior to phthalate exposure.
	Metric 15:	Number of Organisms and Replicates per Group	Low	There was a low number of exposure groups.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Test conditions were adequate.
	Metric 17:	Outcome Assessment Methodology	High	Methodology was addressed.

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Study Citation:	Wofford, H. W., Wilsey, C. D., Neff, G. S., Giam, C. S., Neff, J. M. (1981). Bioaccumulation and metabolism of phthalate esters by oysters, brown shrimp, and sheepshead minnows. <i>Ecotoxicology and Environmental Safety</i> 5(2):202-210.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>Cyprinodon variegatus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	789995			
Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	Medium	There was consistency in outcome assessment.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	No confounding variables were indicated in the assessment.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	Outcomes unrelated to exposure were not reported.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	A three-way analysis of variance (ANOVA) on the data was performed using the General Linear Model procedure of SAS 76.
	Metric 22:	Reporting of Data	High	Data for each outcome was reported.
	Metric 23:	Explanation of Unexpected Outcomes	High	No unexpected outcomes were reported.
Additional Comments: None				

Overall Quality Determination**Uninformative**

Study Citation:	Bionomics,, Springborn (1984). Acute toxicity of thirteen phthalate esters to the sheepshead minnow (<i>Cyprinodon variegatus</i>) (final report).			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (marine), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>sheepshead minnow (Cyprinodon variegatus)</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1316224; Linked HERO ID(s): 1321996, 1316224			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The test material was identified and a CASRN was given.	
	Metric 2: Test Substance Source	High	The source was listed as EG&G Bionomics- Aquatic Toxicology Laboratory in Wareham, Massachusetts. No other information about the source was given.	
	Metric 3: Test Substance Purity	Low	Purity and/or grade of the test substance were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	A negative control was used.	
	Metric 5: Negative Control Response	High	No mortality was reported in the controls.	
	Metric 6: Randomized Allocation	Medium	Test organisms were impartially distributed to each chamber (pdf pg 136).	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail and accounted for the properties of the test material. For all low-solubility phthalates, enhanced mixing procedure was used, while for butyl benzyl phthalate exposure microbial degradation was accounted for with a cleaning procedure that was implemented daily. The authors reported significant degradation of the test material throughout the test, but quantified the degradation throughout the test and reported the endpoint in terms of mean-measured concentration so this does not have an impact on the results.	
	Metric 8: Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	High	Concentrations were measured using GS-MS to account for poor water solubility.	
	Metric 10: Exposure Duration and Frequency	High	The 96-hour exposure was appropriate for an acute test.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	N/A	Test concentration spacing was limited, as this was designed as a limit test up to the solubility limit of the chemical.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit, which were reported in Appendix A (pdf pg 164).	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	Specimens were either cultured at the Laboratory or purchased commercially. All fish were tested as juveniles at <10weeks old.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	A 96-hour acclimation period was reported.	

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Study Citation:	Bionomics,, Springborn (1984). Acute toxicity of thirteen phthalate esters to the sheepshead minnow (<i>Cyprinodon variegatus</i>) (final report).			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (marine), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Fish; <i>sheepshead minnow (Cyprinodon variegatus)</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1316224; Linked HERO ID(s): 1321996, 1316224			
Domain	Metric	Rating	Comments	
	Metric 15: Number of Organisms and Replicates per Group	Low	Only two replicates of 10 fish were used in each treatment.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Organism housing, environmental conditions, food, water, and nutrients were conducive to maintenance of health, and biomass loading was appropriate.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology partially addressed or reported the intended outcomes(s) of interest (mortality).	
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Medium	The study reported minor differences among the study groups with respect to environmental conditions or other non-treatment-related factors, but these are unlikely to have a substantial impact on results. Authors reported that dissolved oxygen fell below guideline recommended levels and control mortality was high for one phthalate (not this phthalate). As no mortalities were observed in any test concentrations for this chemical, this was not determined to affect this test.	
	Metric 20: Outcomes Unrelated to Exposure	High	Details regarding test organism attrition and outcomes unrelated to exposure (e.g., infection) were reported for each study group and there were no differences among groups that could influence the outcome assessment.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	N/A	No mortality was reported, so no statistical analysis was needed.	
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group and were adequate to determine values for the endpoint(s) of interest. Negative findings were reported quantitatively.	
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes, or unexpected outcomes were satisfactorily explained. Low DO did not have an affect on the outcome and high mortality in controls was not reported for this chemical.	
Additional Comments:	Study report for the Sheepshead minnow test begins on pg 124 of the PDF. DEHP is referred to as phthalate 1H, Dibutyl Phthalate was referred to as 1C, and Butyl Benzyl Phthalate was referred to as 1D.			

Overall Quality Determination**High**

Study Citation:	Lee, S. K., Veeramachaneni, R., D.N. (2005). Subchronic exposure to low concentrations of di-n-butyl phthalate disrupts spermatogenesis in <i>Xenopus laevis</i> frogs. <i>Toxicological Sciences</i> 84(2):394-407.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Larvae			
Health Outcome:	Mechanistic-Endocrine toxicity			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	128004			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The DBP was identified by CASRN.	
	Metric 2: Test Substance Source	Low	The source was reported to be Sigma Aldrich, and the lot number was reported, but it was not reported if the DBP was analytically verified.	
	Metric 3: Test Substance Purity	High	The DBP was reported to be 99.8% pure.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a negative control in the FETAX solution and a negative control containing 0.01% DMSO.	
	Metric 5: Negative Control Response	High	The biological response of the negative controls was reported in the text under results.	
	Metric 6: Randomized Allocation	Medium	It was reported that tadpoles were randomly assigned to treatment groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	Stock solutions were prepared in 100mL amber glass bottles weekly by adding DBP directly to DMSO. Glass pipettes were used to add the proper quantity to the test tanks. It was then manually stirred. Renewal were conducted every M-W-F.	
	Metric 8: Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups. The same solvent was used in all test concentrations and in the solvent control. All test concentrations were run until complete metamorphosis of the control organisms.	
	Metric 9: Measurement of Test Substance Concentration	Low	Study authors did not report if the exposure concentrations were measured.	
	Metric 10: Exposure Duration and Frequency	High	The exposure duration was reported to be from stage 52 (3wks) to stage 66 (8wks). This is typical of amphibian studies.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	There were 5 exposure levels as well as a negative control and a solvent control. This was adequate to see a response.	
	Metric 12: Testing at or Below Solubility Limit	High	A solvent was used in this study, and the solvent control response was adequate indicating the concentration was appropriate.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	Breeding adults were reported to be from Nasco in Fort Atkinson, WI. Adults were bred in the laboratory performing the study, and the tadpoles were reared there. The tadpole stages were appropriate for the study.	
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Study Citation:	Lee, S. K., Veeramachaneni, R., D.N. (2005). Subchronic exposure to low concentrations of di-n-butyl phthalate disrupts spermatogenesis in <i>Xenopus laevis</i> frogs. <i>Toxicological Sciences</i> 84(2):394-407.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Larvae
Health Outcome:	Mechanistic-Endocrine toxicity
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	128004

Domain	Metric	Rating	Comments
	Metric 14: Acclimatization and Pretreatment Conditions	High	Pretreatment conditions were the same as treatment conditions. Temperature and photoperiod were the same for each.
	Metric 15: Number of Organisms and Replicates per Group	Low	The number of test organisms for each test concentration were reported, but the number of replicates for each test concentration was not reported.
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Tadpoles were housed at a loading rate of 2.5 tadpoles/L. They were kept at 20-24C with a 12:12 photoperiod. The organisms were fed daily after 96h in age throughout the study. A slurry was used for the tadpoles and pellets were used after metamorphosis.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—plasma testosterone levels.
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	StatView was used for statistical analysis. ANOVA was used as well as Tukey-Kramer post hoc test.
	Metric 22: Reporting of Data	Low	Data for the exposure related findings was reported in the text.
	Metric 23: Explanation of Unexpected Outcomes	Low	Variability was not reported.
Additional Comments:	This portion of the evaluation was on the effect of DBP on testosterone levels in <i>Xenopus laevis</i> . Plasma testosterone levels were measured, so the mechanistic endocrine outcome was chosen.		

Overall Quality Determination

High

Study Citation:	Lee, S. K., Veeramachaneni, R., D.N. (2005). Subchronic exposure to low concentrations of di-n-butyl phthalate disrupts spermatogenesis in <i>Xenopus laevis</i> frogs. <i>Toxicological Sciences</i> 84(2):394-407.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Larvae			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	128004			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The DBP was identified by CASRN.	
	Metric 2: Test Substance Source	Low	The source was reported to be Sigma Aldrich, and the lot number was reported, but it was not reported if the DBP was analytically verified.	
	Metric 3: Test Substance Purity	High	The DBP was reported to be 99.8% pure.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a negative control in the FETAX solution and a negative control containing 0.01% DMSO.	
	Metric 5: Negative Control Response	High	The biological response of the negative control was reported in Table 1.	
	Metric 6: Randomized Allocation	Medium	It was reported that tadpoles were randomly assigned to treatment groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	Stock solutions were prepared in 100mL amber glass bottles weekly by adding DBP directly to DMSO. Glass pipettes were used to add the proper quantity to the test tanks. It was then manually stirred. Renewals were conducted every M-W-F.	
	Metric 8: Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups. The same solvent was used in all test concentrations and in the solvent control. All test concentrations were run until complete metamorphosis of the control organisms.	
	Metric 9: Measurement of Test Substance Concentration	Low	Study authors did not report if the exposure concentrations were measured.	
	Metric 10: Exposure Duration and Frequency	High	The exposure duration was reported to be from stage 52 (3wks) to stage 66 (8wks). This is typical of amphibian studies.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	There were five exposure levels as well as a negative control and a solvent control. This was adequate to see a response.	
	Metric 12: Testing at or Below Solubility Limit	High	A solvent was used in this study, and the solvent control response was adequate, indicating the concentration was appropriate.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	Breeding adults were reported to be from Nasco in Fort Atkinson, WI. Adults were bred in the laboratory performing the study, and the tadpoles were reared there. The tadpole stages were appropriate for the study.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	Pretreatment conditions were the same as treatment conditions. Temperature and photoperiod were the same for each.	
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Study Citation:	Lee, S. K., Veeramachaneni, R., D.N. (2005). Subchronic exposure to low concentrations of di-n-butyl phthalate disrupts spermatogenesis in <i>Xenopus laevis</i> frogs. <i>Toxicological Sciences</i> 84(2):394-407.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Larvae
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	128004

Domain	Metric	Rating	Comments
	Metric 15: Number of Organisms and Replicates per Group	Low	The number of test organisms for each test concentration were reported, but the number of replicates for each test concentration was not reported.
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Tadpoles were housed at a loading rate of 2.5 tadpoles/L. They were kept at 20-24C with a 12:12 photoperiod. The organisms were fed daily after 96h in age throughout the study. A slurry was used for the tadpoles and pellets were used after metamorphosis.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—body weights/development/growth.
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	StatView was used for statistical analysis. ANOVA was used as well as Tukey-Kramer post hoc test.
	Metric 22: Reporting of Data	High	Data for exposure related findings including control results can be found in Table 1.
	Metric 23: Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in Table 1.
Additional Comments:	This portion of the evaluation was on the effect of DBP on body weight in <i>Xenopus laevis</i> . Body weights for each treatment were obtained, so the development/growth outcome was chosen.		

Overall Quality Determination**High**

Study Citation:	Lee, S. K., Veeramachaneni, R., D.N. (2005). Subchronic exposure to low concentrations of di-n-butyl phthalate disrupts spermatogenesis in <i>Xenopus laevis</i> frogs. <i>Toxicological Sciences</i> 84(2):394-407.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Larvae			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	128004			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The DBP was identified by CASRN.	
	Metric 2: Test Substance Source	Low	The source was reported to be Sigma Aldrich, and the lot number was reported, but it was not reported if the DBP was analytically verified.	
	Metric 3: Test Substance Purity	High	The DBP was reported to be 99.8% pure.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a negative control in the FETAX solution and a negative control containing 0.01% DMSO.	
	Metric 5: Negative Control Response	High	The biological response of the negative controls was reported in Figures 2,3, and 6, as well as in Tables 1 and 2.	
	Metric 6: Randomized Allocation	Medium	It was reported that tadpoles were randomly assigned to treatment groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	Stock solutions were prepared in 100mL amber glass bottles weekly by adding DBP directly to DMSO. Glass pipettes were used to add the proper quantity to the test tanks. It was then manually stirred. Renewals were conducted every M-W-F.	
	Metric 8: Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups. The same solvent was used in all test concentrations and in the solvent control. All test concentrations were run until complete metamorphosis of the control organisms.	
	Metric 9: Measurement of Test Substance Concentration	Low	Study authors did not report if the exposure concentrations were measured.	
	Metric 10: Exposure Duration and Frequency	High	The exposure duration was reported to be from stage 52 (3wks) to stage 66 (8wks). This is typical of amphibian studies.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	There were five exposure levels as well as a negative control and a solvent control. This was adequate to see a response.	
	Metric 12: Testing at or Below Solubility Limit	High	A solvent was used in this study, and the solvent control response was adequate indicating the concentration was appropriate.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	Breeding adults were reported to be from Nasco in Fort Atkinson, WI. Adults were bred in the laboratory performing the study, and the tadpoles were reared there. The tadpole stages were appropriate for the study.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	Pretreatment conditions were the same as treatment conditions. Temperature and photoperiod were the same for each.	
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Study Citation:	Lee, S. K., Veeramachaneni, R., D.N. (2005). Subchronic exposure to low concentrations of di-n-butyl phthalate disrupts spermatogenesis in <i>Xenopus laevis</i> frogs. <i>Toxicological Sciences</i> 84(2):394-407.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Larvae
Health Outcome:	Reproductive/Teratogenic
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	128004

Domain	Metric	Rating	Comments
	Metric 15: Number of Organisms and Replicates per Group	Low	The number of test organisms for each test concentration were reported, but the number of replicates for each test concentration was not reported.
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Tadpoles were housed at a loading rate of 2.5 tadpoles/L. They were kept at 20-24C with a 12:12 photoperiod. The organisms were fed daily after 96h in age throughout the study. A slurry was used for the tadpoles and pellets were used after metamorphosis.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—histopathological changes in reproductive organs.
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	StatView was used for statistical analysis. ANOVA was used as well as Tukey-Kramer post hoc test.
	Metric 22: Reporting of Data	High	Data for exposure related findings including control results can be found in Tables 1 and 2 as well as in Figures 2,3 and 6. Other figures provided exposure related results only.
	Metric 23: Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in the tables.
Additional Comments:	This portion of the evaluation was on the effect of DBP on spermatogenesis in <i>Xenopus laevis</i> . Histopathological analysis was performed to observe the effects on the reproductive system in male frogs. Reproduction was therefore selected as the outcome of interest.		

Overall Quality Determination**High**

Study Citation:	Lee, S. K., Veeramachaneni, R., D.N. (2005). Subchronic exposure to low concentrations of di-n-butyl phthalate disrupts spermatogenesis in <i>Xenopus laevis</i> frogs. <i>Toxicological Sciences</i> 84(2):394-407.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Aquatic (freshwater), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Larvae			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	128004			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The DBP was identified by CASRN.	
	Metric 2: Test Substance Source	Low	The source was reported to be Sigma Aldrich, and the lot number was reported, but it was not reported if the DBP was analytically verified.	
	Metric 3: Test Substance Purity	High	The DBP was reported to be 99.8% pure.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a negative control in the FETAX solution and a negative control containing 0.01% DMSO.	
	Metric 5: Negative Control Response	High	Mortality in the control groups were less than 10% throughout the exposure.	
	Metric 6: Randomized Allocation	Medium	It was reported that tadpoles were randomly assigned to treatment groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	Stock solutions were prepared in 100mL amber glass bottles weekly by adding DBP directly to DMSO. Glass pipettes were used to add the proper quantity to the test tanks. It was then manually stirred. Renewal were conducted every M-W-F.	
	Metric 8: Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups. The same solvent was used in all test concentrations and in the solvent control. All test concentrations were run until complete metamorphosis of the control organisms.	
	Metric 9: Measurement of Test Substance Concentration	Low	Study authors did not report if the exposure concentrations were measured.	
	Metric 10: Exposure Duration and Frequency	High	The exposure duration was reported to be from stage 52 (3wks) to stage 66 (8wks). This is typical of amphibian studies.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	There were 5 exposure levels as well as a negative control and a solvent control. This was adequate to see a response.	
	Metric 12: Testing at or Below Solubility Limit	High	A solvent was used in this study, and the solvent control response was adequate indicating the concentration was appropriate.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	Breeding adults were reported to be from Nasco in Fort Atkinson, WI. Adults were bred in the laboratory performing the study, and the tadpoles were reared there. The tadpole stages were appropriate for the study.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	Pretreatment conditions were the same as treatment conditions. Temperature and photoperiod were the same for each.	
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Study Citation:	Lee, S. K., Veeramachaneni, R., D.N. (2005). Subchronic exposure to low concentrations of di-n-butyl phthalate disrupts spermatogenesis in <i>Xenopus laevis</i> frogs. <i>Toxicological Sciences</i> 84(2):394-407.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; <i>Xenopus laevis</i> ; Larvae
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	128004

Domain	Metric	Rating	Comments
	Metric 15: Number of Organisms and Replicates per Group	Low	The number of test organisms for each test concentration were reported, but the number of replicates for each test concentration was not reported.
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Tadpoles were housed at a loading rate of 2.5 tadpoles/L. They were kept at 20-24C with a 12:12 photoperiod. The organisms were fed daily after 96h in age throughout the study. A slurry was used for the tadpoles and pellets were used after metamorphosis.
	Metric 17: Outcome Assessment Methodology	Low	The mortality assessment was not described in the methods section.
	Metric 18: Consistency of Outcome Assessment	Low	How mortality was assessed was not reported.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	StatView was used for statistical analysis. ANOVA was used as well as Tukey-Kramer post hoc test.
	Metric 22: Reporting of Data	Medium	Mortality was just briefly described in text in the results section. Cumulative percent mortalities per treatment group were given there but no data was shown in any table or figure.
	Metric 23: Explanation of Unexpected Outcomes	Medium	Study authors did not report any unexpected outcomes. No variability was shown for mortality data.
Additional Comments: This evaluation is for the mortality assessment during the study.			

Overall Quality Determination**High**

Study Citation:	Sugawara, N. (1974). Toxic effect of a normal series of phthalate esters on the hatching of shrimp eggs. Toxicology and Applied Pharmacology 30(1):87-89.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Artemia salina</i> ; Embryo			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1315792			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The source was not reported.	
	Metric 3: Test Substance Purity	Low	Purity or grade of the test substance were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	Medium	A hatch rate of ~47% in the control seems low.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The study provided only limited details on the measures taken to appropriately prepare test concentrations. Triton X-100 was used as a carrier. The experimental set up and the type of experimental vessel used were not reported.	
	Metric 8: Consistency of Exposure Administration	Medium	A one-time dose was used. The volume of test solution in experimental dishes was not reported.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	Medium	Figure 1 is for 72-hour values. The text states 40 or 72 hours was the exposure duration.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	Three treatment levels were used with adequate spacing between them.	
	Metric 12: Testing at or Below Solubility Limit	Medium	The solvent concentration seemed high. "The concentration of Triton X-100 in the control, 10 ppm, and 20-ppm solutions was adjusted to 10 ppm by adding this reagent."	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the test eggs was not reported.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups.	
	Metric 15: Number of Organisms and Replicates per Group	Low	One or two mg of eggs were placed in test dishes. This seems like a wide range. No replicates were reported.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.	
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Study Citation:	Sugawara, N. (1974). Toxic effect of a normal series of phthalate esters on the hatching of shrimp eggs. Toxicology and Applied Pharmacology 30(1):87-89.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Artemia salina</i> ; Embryo			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1315792			
Domain	Metric	Rating	Comments	
	Metric 17: Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported.	
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical analysis was performed. The Fig. 1 caption states that student's t test was used.	
	Metric 22: Reporting of Data	Low	Data for exposure-related findings were presented for each treatment and control group in Figure 1. The legend for the graph is not included. The labeling of phthalate esters is incorrect. Cannot locate DBP on the graph.	
	Metric 23: Explanation of Unexpected Outcomes	Low	Insufficient information was provided to determine if excessive variability or unexpected outcomes occurred.	
Additional Comments:	Mortality of shrimp eggs in the control was ~47% and DMP data could not be located in Figure 1.			
Overall Quality Determination		Low		

Study Citation:	Wofford, H. W., Wilsey, C. D., Neff, G. S., Giam, C. S., Neff, J. M. (1981). Bioaccumulation and metabolism of phthalate esters by oysters, brown shrimp, and sheepshead minnows. <i>Ecotoxicology and Environmental Safety</i> 5(2):202-210.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Mollusks; <i>Crassostrea virginica</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	789995			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The chemical was identified by name, but no CASRN or structure were given.
	Metric 2:	Test Substance Source	High	The test substance sources include Aldrich Chemical Company for the unlabeled phthalate and California Bionuclear Corporation for the 14C-labeled phthalate.
	Metric 3:	Test Substance Purity	Low	Purity of the chemical was not included in the study.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Uninformative	An experiment with a control to test the absorbance of empty oyster shells was conducted. However, no controls were used for the experiment that measured concentrations in tissue samples.
	Metric 5:	Negative Control Response	N/A	No negative controls were reported.
	Metric 6:	Randomized Allocation	Low	Organisms were collected from Galveston Bay. An allocation method for test groups was not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The experimental design for test preparation was described. Procedures to account for P-chem properties were not reported.
	Metric 8:	Consistency of Exposure Administration	High	No variations in exposure administration were reported.
	Metric 9:	Measurement of Test Substance Concentration	High	Concentrations were measured using analytical techniques, gas-liquid chromatography and liquid scintillation.
	Metric 10:	Exposure Duration and Frequency	Medium	A 24-hour exposure period was used.
	Metric 11:	Number of Exposure Groups/Spacing of Exposure Levels	Low	Only two exposure groups were used.
	Metric 12:	Testing at or Below Solubility Limit	High	Two concentrations were below the solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	Characteristics were not described in the study.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Organisms were acclimatized for four days prior to phthalate exposure.
	Metric 15:	Number of Organisms and Replicates per Group	Low	A low number of exposure groups was used.
Domain 5: Outcome Assessment				
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Study Citation:	Wofford, H. W., Wilsey, C. D., Neff, G. S., Giam, C. S., Neff, J. M. (1981). Bioaccumulation and metabolism of phthalate esters by oysters, brown shrimp, and sheepshead minnows. <i>Ecotoxicology and Environmental Safety</i> 5(2):202-210.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Mollusks; <i>Crassostrea virginica</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	789995			
Domain	Metric	Rating	Comments	
	Metric 16:	Adequacy of Test Conditions	High	Test conditions were adequate.
	Metric 17:	Outcome Assessment Methodology	High	Methodology was addressed.
	Metric 18:	Consistency of Outcome Assessment	Medium	Outcome assessment was consistent for all groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Medium	The concentrations during the 24-hr exposure period remained relatively constant with the exception of the oyster experiments. The concentrations decreased 30-70% during the oyster exposures, probably due to the high adsorptive capacity of the shells. The assertion was supported by an experiment showing absorption of chemical concentration in empty oyster shells.
	Metric 20:	Outcomes Unrelated to Exposure	High	Outcomes unrelated to exposure were not reported.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	A three-way analysis of variance (ANOVA) on the data was performed using the General Linear Model procedure of SAS 76.
	Metric 22:	Reporting of Data	High	Data for each outcome was reported.
	Metric 23:	Explanation of Unexpected Outcomes	Medium	Study size was small but data was reported.
Additional Comments:	None			

Overall Quality Determination**Uninformative**

Study Citation:	Bionomics,, EG&G (1984). Acute toxicity of twelve phthalate esters to mysid shrimp (Mysidopsis bahia).			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (marine), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Mysidopsis bahia</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1316220			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The test substance was identified by chemical name, but no CASRN or structure were reported.	
	Metric 2: Test Substance Source	High	The source of the phthalates was Bionomics Aquatic Toxicology Laboratory (Wareham, MA).	
	Metric 3: Test Substance Purity	Low	Purity and/or substance were not included in the study.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Authors reported using negative controls.	
	Metric 5: Negative Control Response	High	The response of the negative controls was adequate.	
	Metric 6: Randomized Allocation	Medium	Mysid shrimp were maintained 1-3 days before they were distributed into test vessels.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental design followed protocol guidelines.	
	Metric 8: Consistency of Exposure Administration	High	Authors reported consistent administration.	
	Metric 9: Measurement of Test Substance Concentration	High	Phthalates were analytically verified and measured.	
	Metric 10: Exposure Duration and Frequency	High	The test duration followed protocol.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of replicates used was adequate to the guidelines.	
	Metric 12: Testing at or Below Solubility Limit	High	Concentrations used in the analysis were below the solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Medium	The source of organisms was reported, but details beyond that were not.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	Organisms were housed for 1-3 days prior to treatment.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The replicates followed protocol.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions were adequate and described in detail.	
	Metric 17: Outcome Assessment Methodology	High	Outcomes were reported and addressed.	
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Study Citation:	Bionomics,, EG&G (1984). Acute toxicity of twelve phthalate esters to mysid shrimp (<i>Mysidopsis bahia</i>).			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (marine), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Mysidopsis bahia</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1316220			
Domain		Metric	Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed and reported.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	No differences were reported.
	Metric 20:	Outcomes Unrelated to Exposure	High	No outcomes unrelated to exposure were reported.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistics performed included moving average angle analysis, probit analysis, and binomial probability.
	Metric 22:	Reporting of Data	High	Data was reported adequately.
	Metric 23:	Explanation of Unexpected Outcomes	High	No unexpected outcomes were reported.
Additional Comments:	None			

Overall Quality Determination**High**

Study Citation:	Linden, E., Bengtsson, B. E., Svanberg, O., Sundstrom, G. (1979). The acute toxicity of 78 chemicals and pesticide formulations against two brackish water organisms, the bleak (<i>Alburnus alburnus</i>) and the harpacticoid <i>Nitocra spinipes</i> . Chemosphere 8(11-12):843-851.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Nitocra spinipes</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	51937			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The chemical was identified by name only.
	Metric 2:	Test Substance Source	Low	The source was not reported.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of the test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	Low	The biological response of the negative control group was not reported.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations. They stated that "No control analyses for the actual substance(s) were made of the test solutions."
	Metric 8:	Consistency of Exposure Administration	Medium	Only general methods of exposure administration were reported so assessment was difficult to determine.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	Medium	The duration of exposure and exposure frequency were reported and suitable, but slightly longer than typical for the study type (96h).
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	At least six concentrations were tested, but the range of exposure groups was not reported.
	Metric 12:	Testing at or Below Solubility Limit	Low	Reporting omissions prevented determination of whether exposure concentrations exceeded the water solubility limit. However, the reported LC50, 1.7 mg/L, is below the solubility reported in the Final Scope for DBP (11.2 mg/L at 25C).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The source of the test animals was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It is unclear if test organisms were acclimatized to test conditions.
	Metric 15:	Number of Organisms and Replicates per Group	Low	Twenty organisms with no replicates per treatment were used.
Domain 5: Outcome Assessment				
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Study Citation:	Linden, E., Bengtsson, B. E., Svanberg, O., Sundstrom, G. (1979). The acute toxicity of 78 chemicals and pesticide formulations against two brackish water organisms, the bleak (<i>Alburnus alburnus</i>) and the harpacticoid <i>Nitocra spinipes</i> . Chemosphere 8(11-12):843-851.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Nitocra spinipes</i> ; Adult
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	51937

Domain	Metric	Rating	Comments
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported and seemed consistent.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group, but results were described in Table 3.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments: None			

Overall Quality Determination**Medium**

Study Citation:	Wofford, H. W., Wilsey, C. D., Neff, G. S., Giam, C. S., Neff, J. M. (1981). Bioaccumulation and metabolism of phthalate esters by oysters, brown shrimp, and sheepshead minnows. <i>Ecotoxicology and Environmental Safety</i> 5(2):202-210.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Penaecus aztecus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	789995			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The chemical was identified by name, but no CASRN or structure were given.
	Metric 2:	Test Substance Source	High	The test substance sources include Aldrich Chemical Company for the unlabeled phthalate and California Bionuclear Corporation for the 14C-labeled phthalate.
	Metric 3:	Test Substance Purity	Low	Purity of the chemical was not included in the study.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Uninformative	No negative controls were reported.
	Metric 5:	Negative Control Response	N/A	No negative controls were reported.
	Metric 6:	Randomized Allocation	Low	Organisms were collected from Galveston Bay. Allocation method into exposure groups was not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The experimental design for test preparation was described. Measures taken to account for P-chem properties were not reported.
	Metric 8:	Consistency of Exposure Administration	High	No variations in exposure administration were reported.
	Metric 9:	Measurement of Test Substance Concentration	High	Concentrations were measured using analytical techniques, gas-liquid chromatography and liquid scintillation.
	Metric 10:	Exposure Duration and Frequency	Medium	A 24-hour exposure period was used.
	Metric 11:	Number of Exposure Groups/Spacing of Exposure Levels	Low	Only two exposure groups were reported.
	Metric 12:	Testing at or Below Solubility Limit	High	Two concentrations were below the solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	Characteristics were not described in the study.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Organisms were acclimatized for four days prior to phthalate exposure.
	Metric 15:	Number of Organisms and Replicates per Group	Low	A low number of exposure groups was used.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Test conditions were adequate.
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Study Citation:	Wofford, H. W., Wilsey, C. D., Neff, G. S., Giam, C. S., Neff, J. M. (1981). Bioaccumulation and metabolism of phthalate esters by oysters, brown shrimp, and sheepshead minnows. Ecotoxicology and Environmental Safety 5(2):202-210.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Penaecus aztecus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	789995			
Domain	Metric	Rating	Comments	
	Metric 17:	Outcome Assessment Methodology	High	Methodology was addressed.
	Metric 18:	Consistency of Outcome Assessment	High	Outcome assessment was consistent for all groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	No confounding variables were indicated in the assessment reported.
	Metric 20:	Outcomes Unrelated to Exposure	High	Outcomes unrelated to exposure were not reported.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	A three-way analysis of variance (ANOVA) on the data was performed using the General Linear Model procedure of SAS 76.
	Metric 22:	Reporting of Data	High	Data for each outcome was reported.
	Metric 23:	Explanation of Unexpected Outcomes	High	Outcomes unrelated to exposure were not reported.
Additional Comments:	None			

Overall Quality Determination**Uninformative**

Study Citation:	Cornell University, (1931). Report upon the toxicity of plasticizers.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection			
Taxa, Species, Age:	Vertebrate; Mammalian; <i>Capra hircus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332948			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The DBP was identified by name and CASRN.
	Metric 2:	Test Substance Source	Low	The source of the DBP was not reported.
	Metric 3:	Test Substance Purity	Low	The purity/grade of the DBP was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Uninformative	Study authors did not report the use of a negative control. Only one goat was injected with DBP. The other goat was injected with substance 3.
	Metric 5:	Negative Control Response	Uninformative	Study authors did not report the use of a negative control, so a negative control response was not reported.
	Metric 6:	Randomized Allocation	Low	It was not reported how the two goats were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The preparation of the DBP for the injection was not reported. The exposure was via a subcutaneous injection.
	Metric 8:	Consistency of Exposure Administration	Low	Reporting omissions are likely to have a substantial impact on results. Injections appeared to be administered daily for 11 days, but no other information was provided on the administration.
	Metric 9:	Measurement of Test Substance Concentration	Low	Study authors did not report if the test doses were measured.
	Metric 10:	Exposure Duration and Frequency	Medium	The exposure duration was reported to be one injection a day for 11 days at doses ranging from 5cc to 50cc. This appeared to be sufficient time to assess the outcomes of interest.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	There were several different doses administered over an 11 day period, but there was only one goat to which they were administered. It's possible the effect of the doses administered could have been cumulative.
	Metric 12:	Testing at or Below Solubility Limit	Low	Reporting omissions prevented determination of whether exposure concentrations exceeded the water solubility limit. Exposures were reported in ccs. The concentration of the DBP being dispensed in the injection was not reported.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the goats and the age of the goats were not reported. They were simply reported to be 15kg. The breed of the goat was not reported either.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the goats were acclimated in any way to test conditions.

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Study Citation:	Cornell University, (1931). Report upon the toxicity of plasticizers.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection			
Taxa, Species, Age:	Vertebrate; Mammalian; <i>Capra hircus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332948			
Domain	Metric	Rating	Comments	
	Metric 15: Number of Organisms and Replicates per Group	Uninformative	There was only one goat exposed to DBP in this study. There was also no control, so no comparison could be statistically made.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	No information on environmental conditions was provided.	
	Metric 17: Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported. It was reported autopsy was performed, but it is unclear what was looked at for this.	
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited. Little details on the autopsy protocol were reported other than autopsy was performed one day after the final injection.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. Environmental conditions were not reported, nor was it reported if the goat was acclimated.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	Uninformative	There was only one organism that was exposed to DBP and no control, so statistical comparison was not possible.	
	Metric 22: Reporting of Data	Low	Results were described in the text only.	
	Metric 23: Explanation of Unexpected Outcomes	Low	Insufficient information was provided to determine if excessive variability or unexpected outcomes occurred. There was only one organism exposed, so variability or unexpected outcomes were uncertain.	
Additional Comments:	This evaluation was on the effect of DBP administered via subcutaneous injection to a goat. This study received an unacceptable ranking due to the lack of negative control as well as because there was only one organism used in the study. No statistical analysis could be conducted because there was only one organism and no control. Autopsy was performed. It was reported the goat exposed to DBP did not have any visible injury in the autopsy. Little other information was reported. The goat exposed to substance 3 (not PECO relevant) reported effects in the gastrointestinal tract, lymph nodes, heart rate, lungs, liver, and ovaries as well as a change in body temperature. So it is inferred these systems were checked in the goat exposed to DBP.			

Overall Quality Determination**Uninformative**

Study Citation:	Cornell University, (1931). Report upon the toxicity of plasticizers.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection			
Taxa, Species, Age:	Vertebrate; Mammalian; <i>Capra hircus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Respiratory			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332948			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The DBP was identified by name and CASRN.	
	Metric 2: Test Substance Source	Low	The source of the DBP was not reported.	
	Metric 3: Test Substance Purity	Low	The purity/grade of the DBP was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	Uninformative	Study authors did not report the use of a negative control. Only one goat was injected with DBP. The other goat was injected with substance 3.	
	Metric 5: Negative Control Response	Uninformative	Study authors did not report the use of a negative control, so a negative control response was not reported.	
	Metric 6: Randomized Allocation	Low	It was not reported how the 2 goats were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The preparation of the DBP for the injection was not reported. The exposure was via a subcutaneous injection.	
	Metric 8: Consistency of Exposure Administration	Low	Reporting omissions are likely to have a substantial impact on results. Injections appeared to be administered daily for 11 days, but no other information was provided on the administration.	
	Metric 9: Measurement of Test Substance Concentration	Low	Study authors did not report if the test doses were measured.	
	Metric 10: Exposure Duration and Frequency	Medium	The exposure duration was reported to be one injection a day for 11 days at doses ranging from 5cc to 50cc. This appeared to sufficient time to assess the outcomes of interest.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Low	There were several different doses administered over an 11 day period, but there was only one goat to which they were administered. It's possible the effect of the doses administered could have been cumulative.	
	Metric 12: Testing at or Below Solubility Limit	Low	Reporting omissions prevented determination of whether exposure concentrations exceeded the water solubility limit. Exposures were reported in ccs. The concentration of the DBP being dispensed in the injection was not reported.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the goats and the age of the goats were not reported. They were simply reported to be 15kg. The breed of the goat was not reported either.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the goats were acclimated in any way to test conditions.	
	Metric 15: Number of Organisms and Replicates per Group	Uninformative	There was only one goat exposed to DBP in this study. There was also no control, so no comparison could be statistically made.	
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Study Citation:	Cornell University, (1931). Report upon the toxicity of plasticizers.		
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days		
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection		
Taxa, Species, Age:	Vertebrate; Mammalian; <i>Capra hircus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Respiratory		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	1332948		
Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	Low	No information on environmental conditions was provided.
	Metric 17: Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported. It was reported autopsy was performed, but it is unclear what was looked at for this.
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment limited. Little details on the autopsy protocol were reported other than autopsy was performed 1 day after the final injection.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. Environmental conditions were not reported, nor was it reported if the goat was acclimated.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Uninformative	There was only one organism that was exposed to DBP and no control, so statistical comparison was not possible.
	Metric 22: Reporting of Data	Low	Results were described in the text only.
	Metric 23: Explanation of Unexpected Outcomes	Low	Insufficient information was provided to determine if excessive variability or unexpected outcomes occurred. There was only one organism exposed, so variability or unexpected outcomes were uncertain.
Additional Comments:	This evaluation was on the effect of DBP administered via subcutaneous injection to a goat. This study received an unacceptable ranking due to the lack of negative control as well as because there was only one organism used in the study. No statistical analysis could be conducted because there was only one organism and no control. Autopsy was performed. It was reported the goat exposed to DBP did not have any visible injury in the autopsy. Little other information was reported. The goat exposed to substance 3 (not PECO relevant) reported effects in the gastrointestinal tract, lymph nodes, heart rate, lungs, liver, and ovaries as well as a change in body temperature. So it is inferred these systems were checked in the goat exposed to DBP.		

Overall Quality Determination**Uninformative**

Study Citation:	Cornell University, (1931). Report upon the toxicity of plasticizers.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection			
Taxa, Species, Age:	Vertebrate; Mammalian; <i>Capra hircus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Hepatic/Liver			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332948			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The DBP was identified by name and CASRN.
	Metric 2:	Test Substance Source	Low	The source of the DBP was not reported.
	Metric 3:	Test Substance Purity	Low	The purity/grade of the DBP was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Uninformative	Study authors did not report the use of a negative control. Only one goat was injected with DBP. The other goat was injected with substance 3.
	Metric 5:	Negative Control Response	Uninformative	Study authors did not report the use of a negative control, so a negative control response was not reported.
	Metric 6:	Randomized Allocation	Low	It was not reported how the 2 goats were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The preparation of the DBP for the injection was not reported. The exposure was via a subcutaneous injection.
	Metric 8:	Consistency of Exposure Administration	Low	Reporting omissions are likely to have a substantial impact on results. Injections appeared to be administered daily for 11 days, but no other information was provided on the administration.
	Metric 9:	Measurement of Test Substance Concentration	Low	Study authors did not report if the test doses were measured.
	Metric 10:	Exposure Duration and Frequency	Medium	The exposure duration was reported to be one injection a day for 11 days at doses ranging from 5cc to 50cc. This appeared to sufficient time to assess the outcomes of interest.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	There were several different doses administered over an 11 day period, but there was only one goat to which they were administered. It's possible the effect of the doses administered could have been cumulative.
	Metric 12:	Testing at or Below Solubility Limit	Low	Reporting omissions prevented determination of whether exposure concentrations exceeded the water solubility limit. Exposures were reported in ccs. The concentration of the DBP being dispensed in the injection was not reported.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the goats and the age of the goats were not reported. They were simply reported to be 15kg. The breed of the goat was not reported either.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the goats were acclimated in any way to test conditions.
	Metric 15:	Number of Organisms and Replicates per Group	Uninformative	There was only one goat exposed to DBP in this study. There was also no control, so no comparison could be statistically made.
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Study Citation:	Cornell University, (1931). Report upon the toxicity of plasticizers.		
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days		
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection		
Taxa, Species, Age:	Vertebrate; Mammalian; <i>Capra hircus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Hepatic/Liver		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	1332948		
Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	Low	No information on environmental conditions was provided.
	Metric 17: Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported. It was reported autopsy was performed, but it is unclear what was looked at for this.
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment limited. Little details on the autopsy protocol were reported other than autopsy was performed 1 day after the final injection.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. Environmental conditions were not reported, nor was it reported if the goat was acclimated.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Uninformative	There was only one organism that was exposed to DBP and no control, so statistical comparison was not possible.
	Metric 22: Reporting of Data	Low	Results were described in the text only.
	Metric 23: Explanation of Unexpected Outcomes	Low	Insufficient information was provided to determine if excessive variability or unexpected outcomes occurred. There was only one organism exposed, so variability or unexpected outcomes were uncertain.
Additional Comments:	This evaluation was on the effect of DBP administered via subcutaneous injection to a goat. This study received an unacceptable ranking due to the lack of negative control as well as because there was only one organism used in the study. No statistical analysis could be conducted because there was only one organism and no control. Autopsy was performed. It was reported the goat exposed to DBP did not have any visible injury in the autopsy. Little other information was reported. The goat exposed to substance 3 (not PECO relevant) reported effects in the gastrointestinal tract, lymph nodes, heart rate, lungs, liver, and ovaries as well as a change in body temperature. So it is inferred these systems were checked in the goat exposed to DBP.		

Overall Quality Determination**Uninformative**

Study Citation:	Cornell University, (1931). Report upon the toxicity of plasticizers.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection			
Taxa, Species, Age:	Vertebrate; Mammalian; <i>Capra hircus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332948			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The DBP was identified by name and CASRN.	
	Metric 2: Test Substance Source	Low	The source of the DBP was not reported.	
	Metric 3: Test Substance Purity	Low	The purity/grade of the DBP was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	Uninformative	Study authors did not report the use of a negative control. Only one goat was injected with DBP. The other goat was injected with substance 3.	
	Metric 5: Negative Control Response	Uninformative	Study authors did not report the use of a negative control, so a negative control response was not reported.	
	Metric 6: Randomized Allocation	Low	It was not reported how the 2 goats were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The preparation of the DBP for the injection was not reported. The exposure was via a subcutaneous injection.	
	Metric 8: Consistency of Exposure Administration	Low	Reporting omissions are likely to have a substantial impact on results. Injections appeared to be administered daily for 11 days, but no other information was provided on the administration.	
	Metric 9: Measurement of Test Substance Concentration	Low	Study authors did not report if the test doses were measured.	
	Metric 10: Exposure Duration and Frequency	Medium	The exposure duration was reported to be one injection a day for 11 days at doses ranging from 5cc to 50cc. This appeared to sufficient time to assess the outcomes of interest.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Low	There were several different doses administered over an 11 day period, but there was only one goat to which they were administered. It's possible the effect of the doses administered could have been cumulative.	
	Metric 12: Testing at or Below Solubility Limit	Low	Reporting omissions prevented determination of whether exposure concentrations exceeded the water solubility limit. Exposures were reported in ccs. The concentration of the DBP being dispensed in the injection was not reported.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the goats and the age of the goats were not reported. They were simply reported to be 15kg. The breed of the goat was not reported either.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the goats were acclimated in any way to test conditions.	
	Metric 15: Number of Organisms and Replicates per Group	Uninformative	There was only one goat exposed to DBP in this study. There was also no control, so no comparison could be statistically made.	
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Study Citation:	Cornell University, (1931). Report upon the toxicity of plasticizers.		
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days		
Exposure Route,	Terrestrial; N/A (e.g., injection); Injection		
Media, Path:			
Taxa, Species, Age:	Vertebrate; Mammalian; <i>Capra hircus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Mortality		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	1332948		
Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	Low	No information on environmental conditions was provided.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest–mortality. Observations for mortality were made for the duration of the test and then one day after the last exposure.
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. The goat was observed daily for mortality.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. Environmental conditions were not reported, nor was it reported if the goat was acclimated.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Uninformative	There was only one organism that was exposed to DBP and no control, so statistical comparison was not possible.
	Metric 22: Reporting of Data	Low	Results were described in the text only.
	Metric 23: Explanation of Unexpected Outcomes	Low	Insufficient information was provided to determine if excessive variability or unexpected outcomes occurred. There was only one organism exposed, so variability or unexpected outcomes were uncertain.
Additional Comments:	This evaluation was on the effect of DBP administered via subcutaneous injection to a goat. This study received an unacceptable ranking due to the lack of negative control as well as because there was only one organism used in the study. No statistical analysis could be conducted because there was only one organism and no control.		

Overall Quality Determination**Uninformative**

Study Citation:	Cornell University, (1931). Report upon the toxicity of plasticizers.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection			
Taxa, Species, Age:	Vertebrate; Mammalian; <i>Capra hircus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Gastrointestinal			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332948			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The DBP was identified by name and CASRN.	
	Metric 2: Test Substance Source	Low	The source of the DBP was not reported.	
	Metric 3: Test Substance Purity	Low	The purity/grade of the DBP was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	Uninformative	Study authors did not report the use of a negative control. Only one goat was injected with DBP. The other goat was injected with substance 3.	
	Metric 5: Negative Control Response	Uninformative	Study authors did not report the use of a negative control, so a negative control response was not reported.	
	Metric 6: Randomized Allocation	Low	It was not reported how the 2 goats were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The preparation of the DBP for the injection was not reported. The exposure was via a subcutaneous injection.	
	Metric 8: Consistency of Exposure Administration	Low	Reporting omissions are likely to have a substantial impact on results. Injections appeared to be administered daily for 11 days, but no other information was provided on the administration.	
	Metric 9: Measurement of Test Substance Concentration	Low	Study authors did not report if the test doses were measured.	
	Metric 10: Exposure Duration and Frequency	Medium	The exposure duration was reported to be one injection a day for 11 days at doses ranging from 5cc to 50cc. This appeared to sufficient time to assess the outcomes of interest.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Low	There were several different doses administered over an 11 day period, but there was only one goat to which they were administered. It's possible the effect of the doses administered could have been cumulative.	
	Metric 12: Testing at or Below Solubility Limit	Low	Reporting omissions prevented determination of whether exposure concentrations exceeded the water solubility limit. Exposures were reported in ccs. The concentration of the DBP being dispensed in the injection was not reported.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the goats and the age of the goats were not reported. They were simply reported to be 15kg. The breed of the goat was not reported either.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the goats were acclimated in any way to test conditions.	
	Metric 15: Number of Organisms and Replicates per Group	Uninformative	There was only one goat exposed to DBP in this study. There was also no control, so no comparison could be statistically made.	
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Study Citation:	Cornell University, (1931). Report upon the toxicity of plasticizers.		
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days		
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection		
Taxa, Species, Age:	Vertebrate; Mammalian; <i>Capra hircus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Gastrointestinal		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	1332948		
Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	Low	No information on environmental conditions was provided.
	Metric 17: Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported. It was reported autopsy was performed, but it is unclear what was looked at for this.
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment limited. Little details on the autopsy protocol were reported other than autopsy was performed 1 day after the final injection.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. Environmental conditions were not reported, nor was it reported if the goat was acclimated.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Uninformative	There was only one organism that was exposed to DBP and no control, so statistical comparison was not possible.
	Metric 22: Reporting of Data	Low	Results were described in the text only.
	Metric 23: Explanation of Unexpected Outcomes	Low	Insufficient information was provided to determine if excessive variability or unexpected outcomes occurred. There was only one organism exposed, so variability or unexpected outcomes were uncertain.
Additional Comments:	This evaluation was on the effect of DBP administered via subcutaneous injection to a goat. This study received an unacceptable ranking due to the lack of negative control as well as because there was only one organism used in the study. No statistical analysis could be conducted because there was only one organism and no control. Autopsy was performed. It was reported the goat exposed to DBP did not have any visible injury in the autopsy. Little other information was reported. The goat exposed to substance 3 (not PECO relevant) reported effects in the gastrointestinal tract, lymph nodes, heart rate, lungs, liver, and ovaries as well as a change in body temperature. So it is inferred these systems were checked in the goat exposed to DBP.		

Overall Quality Determination**Uninformative**

Study Citation:	Cornell University, (1931). Report upon the toxicity of plasticizers.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection			
Taxa, Species, Age:	Vertebrate; Mammalian; <i>Capra hircus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Immune/Hematological			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332948			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The DBP was identified by name and CASRN.	
	Metric 2: Test Substance Source	Low	The source of the DBP was not reported.	
	Metric 3: Test Substance Purity	Low	The purity/grade of the DBP was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	Uninformative	Study authors did not report the use of a negative control. Only one goat was injected with DBP. The other goat was injected with substance 3.	
	Metric 5: Negative Control Response	Uninformative	Study authors did not report the use of a negative control, so a negative control response was not reported.	
	Metric 6: Randomized Allocation	Low	It was not reported how the 2 goats were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The preparation of the DBP for the injection was not reported. The exposure was via a subcutaneous injection.	
	Metric 8: Consistency of Exposure Administration	Low	Reporting omissions are likely to have a substantial impact on results. Injections appeared to be administered daily for 11 days, but no other information was provided on the administration.	
	Metric 9: Measurement of Test Substance Concentration	Low	Study authors did not report if the test doses were measured.	
	Metric 10: Exposure Duration and Frequency	Medium	The exposure duration was reported to be one injection a day for 11 days at doses ranging from 5cc to 50cc. This appeared to sufficient time to assess the outcomes of interest.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Low	There were several different doses administered over an 11 day period, but there was only one goat to which they were administered. It's possible the effect of the doses administered could have been cumulative.	
	Metric 12: Testing at or Below Solubility Limit	Low	Reporting omissions prevented determination of whether exposure concentrations exceeded the water solubility limit. Exposures were reported in ccs. The concentration of the DBP being dispensed in the injection was not reported.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the goats and the age of the goats were not reported. They were simply reported to be 15kg. The breed of the goat was not reported either.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the goats were acclimated in any way to test conditions.	
	Metric 15: Number of Organisms and Replicates per Group	Uninformative	There was only one goat exposed to DBP in this study. There was also no control, so no comparison could be statistically made.	
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Study Citation:	Cornell University, (1931). Report upon the toxicity of plasticizers.		
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days		
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection		
Taxa, Species, Age:	Vertebrate; Mammalian; <i>Capra hircus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Immune/Hematological		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	1332948		
Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	Low	No information on environmental conditions was provided.
	Metric 17: Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported. It was reported autopsy was performed, but it is unclear what was looked at for this.
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment limited. Little details on the autopsy protocol were reported other than autopsy was performed 1 day after the final injection.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. Environmental conditions were not reported, nor was it reported if the goat was acclimated.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Uninformative	There was only one organism that was exposed to DBP and no control, so statistical comparison was not possible.
	Metric 22: Reporting of Data	Low	Results were described in the text only.
	Metric 23: Explanation of Unexpected Outcomes	Low	Insufficient information was provided to determine if excessive variability or unexpected outcomes occurred. There was only one organism exposed, so variability or unexpected outcomes were uncertain.
Additional Comments:	This evaluation was on the effect of DBP administered via subcutaneous injection to a goat. This study received an unacceptable ranking due to the lack of negative control as well as because there was only one organism used in the study. No statistical analysis could be conducted because there was only one organism and no control. Autopsy was performed. It was reported the goat exposed to DBP did not have any visible injury in the autopsy. Little other information was reported. The goat exposed to substance 3 (not PECO relevant) reported effects in the gastrointestinal tract, lymph nodes, heart rate, lungs, liver, and ovaries as well as a change in body temperature. So it is inferred these systems were checked in the goat exposed to DBP.		

Overall Quality Determination**Uninformative**

Study Citation:	Cornell University, (1931). Report upon the toxicity of plasticizers.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection			
Taxa, Species, Age:	Vertebrate; Mammalian; <i>Capra hircus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Cardiovascular			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332948			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The DBP was identified by name and CASRN.	
	Metric 2: Test Substance Source	Low	The source of the DBP was not reported.	
	Metric 3: Test Substance Purity	Low	The purity/grade of the DBP was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	Uninformative	Study authors did not report the use of a negative control. Only one goat was injected with DBP. The other goat was injected with substance 3.	
	Metric 5: Negative Control Response	Uninformative	Study authors did not report the use of a negative control, so a negative control response was not reported.	
	Metric 6: Randomized Allocation	Low	It was not reported how the 2 goats were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The preparation of the DBP for the injection was not reported. The exposure was via a subcutaneous injection.	
	Metric 8: Consistency of Exposure Administration	Low	Reporting omissions are likely to have a substantial impact on results. Injections appeared to be administered daily for 11 days, but no other information was provided on the administration.	
	Metric 9: Measurement of Test Substance Concentration	Low	Study authors did not report if the test doses were measured.	
	Metric 10: Exposure Duration and Frequency	Medium	The exposure duration was reported to be one injection a day for 11 days at doses ranging from 5cc to 50cc. This appeared to sufficient time to assess the outcomes of interest.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Low	There were several different doses administered over an 11 day period, but there was only one goat to which they were administered. It's possible the effect of the doses administered could have been cumulative.	
	Metric 12: Testing at or Below Solubility Limit	Low	Reporting omissions prevented determination of whether exposure concentrations exceeded the water solubility limit. Exposures were reported in ccs. The concentration of the DBP being dispensed in the injection was not reported.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the goats and the age of the goats were not reported. They were simply reported to be 15kg. The breed of the goat was not reported either.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the goats were acclimated in any way to test conditions.	
	Metric 15: Number of Organisms and Replicates per Group	Uninformative	There was only one goat exposed to DBP in this study. There was also no control, so no comparison could be statistically made.	
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Study Citation:	Cornell University, (1931). Report upon the toxicity of plasticizers.		
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days		
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection		
Taxa, Species, Age:	Vertebrate; Mammalian; <i>Capra hircus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Cardiovascular		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	1332948		
Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
Metric 16:	Adequacy of Test Conditions	Low	No information on environmental conditions was provided.
Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported. It was reported autopsy was performed, but it is unclear what was looked at for this.
Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment limited. Little details on the autopsy protocol were reported other than autopsy was performed 1 day after the final injection.
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. Environmental conditions were not reported, nor was it reported if the goat was acclimated.
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	Uninformative	There was only one organism that was exposed to DBP and no control, so statistical comparison was not possible.
Metric 22:	Reporting of Data	Low	Results were described in the text only.
Metric 23:	Explanation of Unexpected Outcomes	Low	Insufficient information was provided to determine if excessive variability or unexpected outcomes occurred. There was only one organism exposed, so variability or unexpected outcomes were uncertain.
Additional Comments:	This evaluation was on the effect of DBP administered via subcutaneous injection to a goat. This study received an unacceptable ranking due to the lack of negative control as well as because there was only one organism used in the study. No statistical analysis could be conducted because there was only one organism and no control. Autopsy was performed. It was reported the goat exposed to DBP did not have any visible injury in the autopsy. Little other information was reported. The goat exposed to substance 3 (not PECO relevant) reported effects in the gastrointestinal tract, lymph nodes, heart rate, lungs, liver, and ovaries as well as a change in body temperature. So it is inferred these systems were checked in the goat exposed to DBP.		

Overall Quality Determination**Uninformative**

Study Citation:	Bello, U. M., Madekurozwa, M.,-C, Groenewald, H. B., Aire, T. A., Arukwe, A. (2014). The effects on steroidogenesis and histopathology of adult male Japanese quails (<i>Coturnix coturnix japonica</i>) testis following pre-pubertal exposure to di(n-butyl) phthalate (DBP). Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 166:24-33.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Vertebrate; Avian; <i>Coturnix coturnix</i> ; japonica; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2346127			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	Chemical was identified by name and CAS#.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity reported as 99%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	High	There were no abnormalities seen in the control testis tissue.	
	Metric 6: Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations. It was not reported whether solutions were prepared in glass or plastic containers.	
	Metric 8: Consistency of Exposure Administration	Medium	Only general methods of exposure administration were reported so assessment of consistency was difficult to determine.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Medium	The number of exposure groups and spacing of exposure levels were suitable, however no response was reported.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via gavage.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The number of test organisms were reported and sufficient to characterize toxicological effects.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.	
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Study Citation:	Bello, U. M., Madekurozwa, M.,-C, Groenewald, H. B., Aire, T. A., Arukwe, A. (2014). The effects on steroidogenesis and histopathology of adult male Japanese quails (<i>Coturnix coturnix japonica</i>) testis following pre-pubertal exposure to di(n-butyl) phthalate (DBP). Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 166:24-33.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Vertebrate; Avian; <i>Coturnix coturnix</i> ; japonica; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2346127			
Domain	Metric	Rating	Comments	
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described.
	Metric 22:	Reporting of Data	High	Histological changes were reported in section 3.3 and shown in Figure 5.
	Metric 23:	Explanation of Unexpected Outcomes	Medium	unexpected outcomes (low concentration effects) were not satisfactorily explained.
Additional Comments:	This evaluation is for the histological assessment of testis tissue.			

Overall Quality Determination**Medium**

Study Citation:	Bello, U. M., Madekurozwa, M.,-C, Groenewald, H. B., Aire, T. A., Arukwe, A. (2014). The effects on steroidogenesis and histopathology of adult male Japanese quails (<i>Coturnix coturnix japonica</i>) testis following pre-pubertal exposure to di(n-butyl) phthalate (DBP). Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 166:24-33.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Vertebrate; Avian; <i>Coturnix coturnix</i> ; japonica; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2346127			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	Chemical was identified by name and CAS#	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity reported as 99%	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group	
	Metric 5: Negative Control Response	Medium	The biological response of the negative control group was only reported in text	
	Metric 6: Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations. It was not reported whether solutions were prepared in glass or plastic containers.	
	Metric 8: Consistency of Exposure Administration	Medium	Only general methods of exposure administration were reported so assessment of consistency was difficult to determine	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Medium	The number of exposure groups and spacing of exposure levels were suitable, however no response was reported	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via gavage	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The number of test organisms were reported and sufficient to characterize toxicological effects	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest	
	Metric 18: Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups	
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Study Citation:	Bello, U. M., Madekurozwa, M.,-C, Groenewald, H. B., Aire, T. A., Arukwe, A. (2014). The effects on steroidogenesis and histopathology of adult male Japanese quails (<i>Coturnix coturnix japonica</i>) testis following pre-pubertal exposure to di(n-butyl) phthalate (DBP). Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 166:24-33.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Vertebrate; Avian; <i>Coturnix coturnix</i> ; japonica; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2346127			
Domain	Metric	Rating	Comments	
Domain 6: Confounding / Variable Control				
Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions	
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups	
Domain 7: Data Presentation and Analysis				
Metric 21:	Statistical Methods	High	Statistical methods were adequately described	
Metric 22:	Reporting of Data	Low	Data were only reported for GSI, other weight results were mentioned in text only	
Metric 23:	Explanation of Unexpected Outcomes	Medium	unexpected outcomes (low concentration effects) were not satisfactorily explained	
Additional Comments:	None			
Overall Quality Determination		Medium		

Study Citation:	Bello, U. M., Madekurozwa, M.,-C, Groenewald, H. B., Aire, T. A., Arukwe, A. (2014). The effects on steroidogenesis and histopathology of adult male Japanese quails (<i>Coturnix coturnix japonica</i>) testis following pre-pubertal exposure to di(n-butyl) phthalate (DBP). Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 166:24-33.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Vertebrate; Avian; <i>Coturnix coturnix</i> ; japonica; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Endocrine toxicity-Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2346127			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The chemical was identified by name and CAS#.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity was reported as 99%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable.	
	Metric 6: Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations. It was not reported whether solutions were prepared in glass or plastic containers.	
	Metric 8: Consistency of Exposure Administration	Medium	Only general methods of exposure administration were reported so assessment of consistency was difficult to determine.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type (30 days).	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Medium	The number of exposure groups and spacing of exposure levels were suitable to observe differences among groups.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via gavage	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions for two weeks.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The number of test organisms were reported and sufficient to characterize toxicological effects (90 birds divided into six groups inclusive of control, or 15 birds per group).	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcomes of interest (steroid analysis via immunoassay, DNA/mRNA analysis via rtPCR).	
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Study Citation:	Bello, U. M., Madekurozwa, M.,-C, Groenewald, H. B., Aire, T. A., Arukwe, A. (2014). The effects on steroidogenesis and histopathology of adult male Japanese quails (<i>Coturnix coturnix japonica</i>) testis following pre-pubertal exposure to di(n-butyl) phthalate (DBP). Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 166:24-33.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Vertebrate; Avian; <i>Coturnix coturnix</i> ; japonica; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Endocrine toxicity-Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2346127			
Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described (One-way ANOVA with Tukey's after testing for normality and homogeneity of variance).
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group in Figures 1-5.
	Metric 23:	Explanation of Unexpected Outcomes	Medium	Unexpected outcomes (non-monotonic response) were not satisfactorily explained.
Additional Comments: None				
Overall Quality Determination			Medium	

Study Citation:	DuPont, (1949). Toxicity of dibutyl phthalate.		
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 0 - 4 days (0-96h)		
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary		
Taxa, Species, Age:	Vertebrate; Avian; <i>Gallus domesticus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Mortality		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	1332945		
Domain	Metric	Rating	Comments
Domain 1: Test Substance			
Metric 1:	Test Substance Identity	Low	Chemical was identified by name. CAS # was provided on the cover sheet for this report by that is not part of the authors work.
Metric 2:	Test Substance Source	Low	No source was listed for this test product. Uncertainty is present with the actual verification of the DBP as a blinded study was conducted: "The Plasticizer samples were received identified only by numbers. Our interpretation of these numbers is based on verbal information only, but was checked up fairly successfully by a determination of the specific gravity of the samples."
Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design			
Metric 4:	Negative Controls	Uninformative	The stock colony of animals was used as a control but no control survival was reported and they did not perform sham treatments for the effects of handling and dose administration.
Metric 5:	Negative Control Response	Low	The biological response of the negative control groups was not reported other than they were reported to "appear normal"
Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization			
Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations
Metric 8:	Consistency of Exposure Administration	Low	One time dose but with few details provided
Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
Metric 12:	Testing at or Below Solubility Limit	N/A	exposure was via gavage
Domain 4: Test Organism			
Metric 13:	Test Organism Characteristics	Low	The source of the test animals was not reported
Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups as treated animals were removed from the stock culture

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Study Citation:	DuPont, (1949). Toxicity of dibutyl phthalate.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Vertebrate; Avian; <i>Gallus domesticus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332945			
Domain	Metric	Rating	Comments	
	Metric 15: Number of Organisms and Replicates per Group	Low	Individuals without replicates were used	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate	
	Metric 17: Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported	
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment (e.g., timing of assessment across groups) were limited	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions	
	Metric 20: Outcomes Unrelated to Exposure	Low	There was no information in the study to suggest differences among groups	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	N/A	Statistical analysis was not conducted as this was not the intent of the study.	
	Metric 22: Reporting of Data	Medium	Data for exposure-related findings were presented for each treatment but control group effects were not quantitated.	
	Metric 23: Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability.	
Additional Comments:	None			

Overall Quality Determination**Uninformative**

Study Citation:	DuPont, (1949). Toxicity of dibutyl phthalate.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection			
Taxa, Species, Age:	Vertebrate; Avian; <i>Gallus domesticus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332945			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The chemical was identified by name. The CAS# was provided on the cover sheet for this report, but that is not part of the author’s work.
	Metric 2:	Test Substance Source	Low	No source was listed for this test product. Uncertainty is present with the actual verification of the DBP as a blinded study was conducted: “The Plasticizer samples were received identified only by numbers. Our interpretation of these numbers is based on verbal information only, but was checked up fairly successfully by a determination of the specific gravity of the samples.”
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of the test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Uninformative	The stock colony of animals was used as a control but no control survival was reported and they did not perform sham treatments for the effects of handling and dose administration.
	Metric 5:	Negative Control Response	Low	The biological response of the negative control groups was not reported other than they were reported to “appear normal”
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.
	Metric 8:	Consistency of Exposure Administration	Low	A one-time dose was used, but with few details provided.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via injection.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test animals was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups as treated animals were removed from the stock culture.
	Metric 15:	Number of Organisms and Replicates per Group	Low	Individuals without replicates were used.

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Study Citation:	DuPont, (1949). Toxicity of dibutyl phthalate.		
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 0 - 4 days (0-96h)		
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection		
Taxa, Species, Age:	Vertebrate; Avian; <i>Gallus domesticus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Mortality		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	1332945		
Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.
	Metric 17: Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported.
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment (e.g., timing of assessment across groups) were limited.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Low	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	N/A	Statistical analysis was not conducted as this was not the intent of the study.
	Metric 22: Reporting of Data	Medium	Data for exposure-related findings were presented for each treatment, but control group effects were not quantitated.
	Metric 23: Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability.
Additional Comments: None			

Overall Quality Determination**Uninformative**

Study Citation:	Korhonen, A., Hemminki, K., Vainio, H. (1983). Embryotoxic effects of phtalic acid derivatives, phosphates and aromatic oils used in the manufacturing of rubber on three day chicken embryos. Drug and Chemical Toxicology 6(2):191-207.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection			
Taxa, Species, Age:	Vertebrate; Avian; <i>Gallus domesticus</i> ; White Leghorn; Embryo			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	94541			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Medium	Technical grade material was used.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.	
	Metric 8: Consistency of Exposure Administration	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and exposure frequency were reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Low	Only two concentrations were tested.	
	Metric 12: Testing at or Below Solubility Limit	Medium	The exposure was via injection that used acetone, but it was not reported what chemicals used acetone for dissolution.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups.	
	Metric 15: Number of Organisms and Replicates per Group	Low	There were 30 eggs without replicates. The number of replicates was not reported.	
Domain 5: Outcome Assessment				
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Study Citation:	Korhonen, A., Hemminki, K., Vainio, H. (1983). Embryotoxic effects of phthalic acid derivatives, phosphates and aromatic oils used in the manufacturing of rubber on three day chicken embryos. Drug and Chemical Toxicology 6(2):191-207.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection
Taxa, Species, Age:	Vertebrate; Avian; <i>Gallus domesticus</i> ; White Leghorn; Embryo
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	94541

Domain	Metric	Rating	Comments
	Metric 16: Adequacy of Test Conditions	Medium	Environmental conditions of the test system were conducive to maintenance of organism health.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18: Consistency of Outcome Assessment	Medium	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	There were no reported differences among the study groups in environmental conditions. "The temperature was kept at 37.7°C and the humidity between 66% and 71% throughout the incubation period." Any acclimation was not reported.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Sufficient data were provided to conduct an independent statistical analysis.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	Low	Variability was not reported.

Additional Comments: This evaluation is for the mortality outcome of chick embryos exposed to DBP.

Overall Quality Determination

Medium

Study Citation:	Korhonen, A., Hemminki, K., Vainio, H. (1983). Embryotoxic effects of phtalic acid derivatives, phosphates and aromatic oils used in the manufacturing of rubber on three day chicken embryos. Drug and Chemical Toxicology 6(2):191-207.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection			
Taxa, Species, Age:	Vertebrate; Avian; <i>Gallus domesticus</i> ; White Leghorn; Embryo			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	94541			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Medium	Technical grade material was used.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.	
	Metric 8: Consistency of Exposure Administration	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and exposure frequency were reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Low	Only two concentrations were tested.	
	Metric 12: Testing at or Below Solubility Limit	Medium	The exposure was via injection that used acetone, but it was not reported what chemicals used acetone for dissolution.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups.	
	Metric 15: Number of Organisms and Replicates per Group	Low	There were 30 eggs without replicates. The number of replicates was not reported.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Medium	Environmental conditions of the test system were conducive to maintenance of organism health.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	
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Study Citation:	Korhonen, A., Hemminki, K., Vainio, H. (1983). Embryotoxic effects of phthalic acid derivatives, phosphates and aromatic oils used in the manufacturing of rubber on three day chicken embryos. Drug and Chemical Toxicology 6(2):191-207.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection
Taxa, Species, Age:	Vertebrate; Avian; <i>Gallus domesticus</i> ; White Leghorn; Embryo
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	94541

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	There were no reported differences among the study groups in environmental conditions. "The temperature was kept at 37.7oC and the humidity between 66% and 71% throughout the incubation period. Any acclimation was not reported.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Sufficient data were provided to conduct an independent statistical analysis.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	Low	Variability was not reported.

Additional Comments: This evaluation is for the malformations reported in the chick embryos after exposure to DBP.

Overall Quality Determination

Medium

Study Citation:	Cornell University, (1931). Report upon the toxicity of plasticizers.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Vertebrate; Avian; <i>Gallus gallus</i> ; cockerels; Juvenile			
Health Outcome:	Ocular and Sensory			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332948			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The DBP was identified by name and CASRN.
	Metric 2:	Test Substance Source	Low	The source of the DBP was not reported.
	Metric 3:	Test Substance Purity	Low	The purity/grade of the DBP was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Uninformative	Study authors did not report the use of a negative control.
	Metric 5:	Negative Control Response	Uninformative	Study authors did not report the use of a negative control, so a negative control response was not reported.
	Metric 6:	Randomized Allocation	Low	It was not reported how the chickens were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The preparation of the DBP for the capsule that was administered was not reported.
	Metric 8:	Consistency of Exposure Administration	Low	Reporting omissions are likely to have a substantial impact on results. The cockerels were fed capsules daily for 10 days with dose increasing by 1.5cc per kg every other day.
	Metric 9:	Measurement of Test Substance Concentration	Low	Study authors did not report if the test doses were measured.
	Metric 10:	Exposure Duration and Frequency	Medium	The exposure duration was reported to 10 days with dosed capsules being fed every day with an increase in dose by 1.5cc per kg of body weight every other day.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	There were several different doses administered over an 10 day period, but there was only one cockerel to which they were administered. It's possible the effect of the doses administered could have been cumulative.
	Metric 12:	Testing at or Below Solubility Limit	N/A	The exposure was via diet.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the birds and the age were not reported. The chickens were reported to be cockerels that were still growing that weighed approximately 1.4kg. No other information was provided.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the birds were acclimated in any way to test conditions.
	Metric 15:	Number of Organisms and Replicates per Group	Uninformative	There was only one bird exposed in each chemical, which was insufficient for statistical analysis.

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Study Citation:	Cornell University, (1931). Report upon the toxicity of plasticizers.		
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days		
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary		
Taxa, Species, Age:	Vertebrate; Avian; <i>Gallus gallus</i> ; cockerels; Juvenile		
Health Outcome:	Ocular and Sensory		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	1332948		
Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	Low	No information on environmental conditions was provided.
	Metric 17: Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported. The chickens were examined after the study, but it was unclear what the protocols for this examination entailed.
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported. The cockerel was examined after a day 10 exposure period, but the protocol was not described in any way.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. Environmental conditions were not reported, nor was it reported if the cockerel was acclimated.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Uninformative	There was only one organism that was exposed to DBP and no control, so statistical comparison was not possible.
	Metric 22: Reporting of Data	Low	Results were described in the text only.
	Metric 23: Explanation of Unexpected Outcomes	Low	Insufficient information was provided to determine if excessive variability or unexpected outcomes occurred. There was only one organism exposed, so variability or unexpected outcomes were uncertain.
Additional Comments:	This evaluation was on the effect of DBP administered via capsule to a chicken for 10 days. The dose increased by 1.5cc per kg body weight every other day for this period. This study received an unacceptable ranking due to the lack of negative control as well as because there was only one organism used in the study. No statistical analysis could be conducted because there was only one organism and no control.		

Overall Quality Determination**Uninformative**

Study Citation:	Cornell University, (1931). Report upon the toxicity of plasticizers.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Vertebrate; Avian; <i>Gallus gallus</i> ; cockerels; Juvenile			
Health Outcome:	Skin & Connective Tissue			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332948			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The DBP was identified by name and CASRN.
	Metric 2:	Test Substance Source	Low	The source of the DBP was not reported.
	Metric 3:	Test Substance Purity	Low	The purity/grade of the DBP was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Uninformative	Study authors did not report the use of a negative control.
	Metric 5:	Negative Control Response	Uninformative	Study authors did not report the use of a negative control, so a negative control response was not reported.
	Metric 6:	Randomized Allocation	Low	It was not reported how the chickens were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The preparation of the DBP for the capsule that was administered was not reported.
	Metric 8:	Consistency of Exposure Administration	Low	Reporting omissions are likely to have a substantial impact on results. The cockerels were fed capsules daily for 10 days with dose increasing by 1.5cc per kg every other day.
	Metric 9:	Measurement of Test Substance Concentration	Low	Study authors did not report if the test doses were measured.
	Metric 10:	Exposure Duration and Frequency	Medium	The exposure duration was reported to 10 days with dosed capsules being fed every day with an increase in dose by 1.5cc per kg of body weight every other day.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	There were several different doses administered over an 10 day period, but there was only one cockerel to which they were administered. It's possible the effect of the doses administered could have been cumulative.
	Metric 12:	Testing at or Below Solubility Limit	N/A	The exposure was via diet.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the birds and the age were not reported. The chickens were reported to be cockerels that were still growing that weighed approximately 1.4kg. No other information was provided.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the birds were acclimated in any way to test conditions.
	Metric 15:	Number of Organisms and Replicates per Group	Uninformative	There was only one bird exposed in each chemical, which was insufficient for statistical analysis.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	No information on environmental conditions was provided.

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Study Citation:	Cornell University, (1931). Report upon the toxicity of plasticizers.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route,	Terrestrial; Food/Diet; Dietary			
Media, Path:				
Taxa, Species, Age:	Vertebrate; Avian; <i>Gallus gallus</i> ; cockerels; Juvenile			
Health Outcome:	Skin & Connective Tissue			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332948			
Domain	Metric	Rating	Comments	
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported. The chickens were examined after the study, but it was unclear what the protocols for this examination entailed. Details regarding the execution of the study protocol for outcome assessment were not reported. The cockerel was examined after a day 10 exposure period, but the protocol was not described in any way.
	Metric 18:	Consistency of Outcome Assessment	Low	
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. Environmental conditions were not reported, nor was it reported if the cockerel was acclimated. There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Uninformative	There was only one organism that was exposed to DBP and no control, so statistical comparison was not possible. Results were described in the text only. Insufficient information was provided to determine if excessive variability or unexpected outcomes occurred. There was only one organism exposed, so variability or unexpected outcomes were uncertain.
	Metric 22:	Reporting of Data	Low	
	Metric 23:	Explanation of Unexpected Outcomes	Low	
Additional Comments:	This evaluation was on the effect of DBP administered via capsule to a chicken for 10 days. The dose increased by 1.5cc per kg body weight every other day for this period. This study received an unacceptable ranking due to the lack of negative control as well as because there was only one organism used in the study. No statistical analysis could be conducted because there was only one organism and no control.			

Overall Quality Determination**Uninformative**

Study Citation:	Cornell University, (1931). Report upon the toxicity of plasticizers.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Vertebrate; Avian; <i>Gallus gallus</i> ; cockerels; Juvenile			
Health Outcome:	Musculoskeletal			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332948			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The DBP was identified by name and CASRN.
	Metric 2:	Test Substance Source	Low	The source of the DBP was not reported.
	Metric 3:	Test Substance Purity	Low	The purity/grade of the DBP was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Uninformative	Study authors did not report the use of a negative control.
	Metric 5:	Negative Control Response	Uninformative	Study authors did not report the use of a negative control, so a negative control response was not reported.
	Metric 6:	Randomized Allocation	Low	It was not reported how the chickens were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The preparation of the DBP for the capsule that was administered was not reported.
	Metric 8:	Consistency of Exposure Administration	Low	Reporting omissions are likely to have a substantial impact on results. The cockerels were fed capsules daily for 10 days with dose increasing by 1.5cc per kg every other day.
	Metric 9:	Measurement of Test Substance Concentration	Low	Study authors did not report if the test doses were measured.
	Metric 10:	Exposure Duration and Frequency	Medium	The exposure duration was reported to 10 days with dosed capsules being fed every day with an increase in dose by 1.5cc per kg of body weight every other day.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	There were several different doses administered over an 10 day period, but there was only one cockerel to which they were administered. It's possible the effect of the doses administered could have been cumulative.
	Metric 12:	Testing at or Below Solubility Limit	N/A	The exposure was via diet.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the birds and the age were not reported. The chickens were reported to be cockerels that were still growing that weighed approximately 1.4kg. No other information was provided.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the birds were acclimated in any way to test conditions.
	Metric 15:	Number of Organisms and Replicates per Group	Uninformative	There was only one bird exposed in each chemical, which was insufficient for statistical analysis.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	No information on environmental conditions was provided.

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Study Citation:	Cornell University, (1931). Report upon the toxicity of plasticizers.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route,	Terrestrial; Food/Diet; Dietary			
Media, Path:				
Taxa, Species, Age:	Vertebrate; Avian; <i>Gallus gallus</i> ; cockerels; Juvenile			
Health Outcome:	Musculoskeletal			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332948			
Domain	Metric	Rating	Comments	
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported. The chickens were examined after the study, but it was unclear what the protocols for this examination entailed.
	Metric 18:	Consistency of Outcome Assessment	Low	
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. Environmental conditions were not reported, nor was it reported if the cockerel was acclimated.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Uninformative	There was only one organism that was exposed to DBP and no control, so statistical comparison was not possible.
	Metric 22:	Reporting of Data	Low	Results were described in the text only.
	Metric 23:	Explanation of Unexpected Outcomes	Low	Insufficient information was provided to determine if excessive variability or unexpected outcomes occurred. There was only one organism exposed, so variability or unexpected outcomes were uncertain.
Additional Comments:	This evaluation was on the effect of DBP administered via capsule to a chicken for 10 days. The dose increased by 1.5cc per kg body weight every other day for this period. This study received an unacceptable ranking due to the lack of negative control as well as because there was only one organism used in the study. No statistical analysis could be conducted because there was only one organism and no control.			

Overall Quality Determination**Uninformative**

Study Citation:	Cornell University, (1931). Report upon the toxicity of plasticizers.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Vertebrate; Avian; <i>Gallus gallus</i> ; cockerels; Juvenile			
Health Outcome:	Gastrointestinal			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332948			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The DBP was identified by name and CASRN.
	Metric 2:	Test Substance Source	Low	The source of the DBP was not reported.
	Metric 3:	Test Substance Purity	Low	The purity/grade of the DBP was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Uninformative	Study authors did not report the use of a negative control.
	Metric 5:	Negative Control Response	Uninformative	Study authors did not report the use of a negative control, so a negative control response was not reported.
	Metric 6:	Randomized Allocation	Low	It was not reported how the chickens were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The preparation of the DBP for the capsule that was administered was not reported.
	Metric 8:	Consistency of Exposure Administration	Low	Reporting omissions are likely to have a substantial impact on results. The cockerels were fed capsules daily for 10 days with the dose increasing by 1.5cc per kg every other day.
	Metric 9:	Measurement of Test Substance Concentration	Low	Study authors did not report if the test doses were measured.
	Metric 10:	Exposure Duration and Frequency	Medium	The exposure duration was reported as 10 days with dosed capsules being fed every day with an increase in dose by 1.5cc per kg of body weight every other day.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	There were several different doses administered over a 10-day period, but there was only one cockerel to which they were administered. It's possible the effect of the doses administered could have been cumulative.
	Metric 12:	Testing at or Below Solubility Limit	N/A	The exposure was via diet.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the birds and the age were not reported. The chickens were reported to be cockerels that were still growing that weighed approximately 1.4kg. No other information was provided.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the birds were acclimated in any way to test conditions.
	Metric 15:	Number of Organisms and Replicates per Group	Uninformative	There was only one bird exposed in each chemical, which was insufficient for statistical analysis.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	No information on environmental conditions was provided.

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Study Citation:	Cornell University, (1931). Report upon the toxicity of plasticizers.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route,	Terrestrial; Food/Diet; Dietary			
Media, Path:				
Taxa, Species, Age:	Vertebrate; Avian; <i>Gallus gallus</i> ; cockerels; Juvenile			
Health Outcome:	Gastrointestinal			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332948			
Domain	Metric	Rating	Comments	
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported. The chickens were examined after the study, but it was unclear what the protocols for this examination entailed. Details regarding the execution of the study protocol for outcome assessment were not reported. The cockerel was examined after a day 10 exposure period, but the protocol was not described in any way.
	Metric 18:	Consistency of Outcome Assessment	Low	
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. Environmental conditions were not reported, nor was it reported if the cockerel was acclimated. There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Uninformative	There was only one organism that was exposed to DBP and no control, so statistical comparison was not possible. Results were described in the text only. Insufficient information was provided to determine if excessive variability or unexpected outcomes occurred. There was only one organism exposed, so variability or unexpected outcomes were uncertain.
	Metric 22:	Reporting of Data	Low	
	Metric 23:	Explanation of Unexpected Outcomes	Low	
Additional Comments:	This evaluation was on the effect of DBP administered via capsule to a chicken for 10 days. The dose increased by 1.5cc per kg body weight every other day for this period. This study received an unacceptable ranking due to the lack of negative control as well as because there was only one organism used in the study. No statistical analysis could be conducted because there was only one organism and no control. The study reported that the bird that received treatment one containing DBP, showed effects of a distended gall bladder and thickening of the small intestine.			

Overall Quality Determination**Uninformative**

Study Citation:	Cornell University, (1931). Report upon the toxicity of plasticizers.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Vertebrate; Avian; <i>Gallus gallus</i> ; cockerels; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332948			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The DBP was identified by name and CASRN.
	Metric 2:	Test Substance Source	Low	The source of the DBP was not reported.
	Metric 3:	Test Substance Purity	Low	The purity/grade of the DBP was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Uninformative	Study authors did not report the use of a negative control.
	Metric 5:	Negative Control Response	Uninformative	Study authors did not report the use of a negative control, so a negative control response was not reported.
	Metric 6:	Randomized Allocation	Low	It was not reported how the chickens were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The preparation of the DBP for the capsule that was administered was not reported.
	Metric 8:	Consistency of Exposure Administration	Low	Reporting omissions are likely to have a substantial impact on results. The cockerels were fed capsules daily for 10 days with dose increasing by 1.5cc per kg every other day.
	Metric 9:	Measurement of Test Substance Concentration	Low	Study authors did not report if the test doses were measured.
	Metric 10:	Exposure Duration and Frequency	Medium	The exposure duration was reported to 10 days with dosed capsules being fed every day with an increase in dose by 1.5cc per kg of body weight every other day.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	There were several different doses administered over an 10 day period, but there was only one cockerel to which they were administered. It's possible the effect of the doses administered could have been cumulative.
	Metric 12:	Testing at or Below Solubility Limit	N/A	The exposure was via diet.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the birds and the age were not reported. The chickens were reported to be cockerels that were still growing that weighed approximately 1.4kg. No other information was provided.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the birds were acclimated in any way to test conditions.
	Metric 15:	Number of Organisms and Replicates per Group	Uninformative	There was only one bird exposed in each chemical, which was insufficient for statistical analysis.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	No information on environmental conditions was provided.

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Study Citation:	Cornell University, (1931). Report upon the toxicity of plasticizers.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route,	Terrestrial; Food/Diet; Dietary			
Media, Path:				
Taxa, Species, Age:	Vertebrate; Avian; <i>Gallus gallus</i> ; cockerels; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332948			
Domain	Metric	Rating	Comments	
	Metric 17:	Outcome Assessment Methodology	High	The bird was monitored for mortality for 10 days.
	Metric 18:	Consistency of Outcome Assessment	High	The bird was monitored for mortality for 10 days.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. Environmental conditions were not reported, nor was it reported if the cockerel was acclimated.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Uninformative	There was only one organism that was exposed to DBP and no control, so statistical comparison was not possible.
	Metric 22:	Reporting of Data	Low	Results were described in the text only.
	Metric 23:	Explanation of Unexpected Outcomes	Low	Insufficient information was provided to determine if excessive variability or unexpected outcomes occurred. There was only one organism exposed, so variability or unexpected outcomes were uncertain.
Additional Comments:	This evaluation was on the effect of DBP administered via capsule to a chicken for 10 days. The dose increased by 1.5cc per kg body weight every other day for this period. This study received an unacceptable ranking due to the lack of negative control as well as because there was only one organism used in the study. No statistical analysis could be conducted because there was only one organism and no control.			

Overall Quality Determination**Uninformative**

Study Citation:	Cornell University, (1931). Report upon the toxicity of plasticizers.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Vertebrate; Avian; <i>Gallus gallus</i> ; cockerels; Juvenile			
Health Outcome:	Ocular and Sensory			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332948			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The DBP was identified by name and CASRN.
	Metric 2:	Test Substance Source	Low	The source of the DBP was not reported.
	Metric 3:	Test Substance Purity	Low	The purity/grade of the DBP was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Uninformative	Study authors did not report the use of a negative control.
	Metric 5:	Negative Control Response	Uninformative	Study authors did not report the use of a negative control, so a negative control response was not reported.
	Metric 6:	Randomized Allocation	Low	It was not reported how the chickens were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The preparation of the DBP for the capsule that was administered was not reported.
	Metric 8:	Consistency of Exposure Administration	Low	Reporting omissions are likely to have a substantial impact on results. The cockerels were fed capsules daily for 11 days (until death) with at a dose of 1.8cc per kg of body weight.
	Metric 9:	Measurement of Test Substance Concentration	Low	Study authors did not report if the test doses were measured.
	Metric 10:	Exposure Duration and Frequency	Medium	The exposure duration was reported to 11 days with a dose of 1.8cc/kg body weight. The exposure was conducted until death of the bird.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	There was only one exposure level. It did not appear that the goal was to observe a dose dependent effect.
	Metric 12:	Testing at or Below Solubility Limit	N/A	The exposure was via diet.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the birds and the age were not reported. The chickens were reported to be cockerels that were still growing that weighed approximately 1.4kg. No other information was provided.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the birds were acclimated in any way to test conditions.
	Metric 15:	Number of Organisms and Replicates per Group	Uninformative	There was only one bird exposed in each chemical, which was insufficient for statistical analysis.
Domain 5: Outcome Assessment				

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Study Citation:	Cornell University, (1931). Report upon the toxicity of plasticizers.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Vertebrate; Avian; <i>Gallus gallus</i> ; cockerels; Juvenile			
Health Outcome:	Ocular and Sensory			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332948			
Domain	Metric	Rating	Comments	
	Metric 16: Adequacy of Test Conditions	Low	No information on environmental conditions was provided.	
	Metric 17: Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported. The chickens were examined after the study, but it was unclear what the protocols for this examination entailed.	
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported. The cockerel was examined after death, which in the case of DBP, was an 11 day exposure period, but the protocol was not described in any way.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. Environmental conditions were not reported, nor was it reported if the cockerel was acclimated.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	Uninformative	There was only one organism that was exposed to DBP and no control, so statistical comparison was not possible.	
	Metric 22: Reporting of Data	Low	Results were described in the text only.	
	Metric 23: Explanation of Unexpected Outcomes	Low	Insufficient information was provided to determine if excessive variability or unexpected outcomes occurred. There was only one organism exposed, so variability or unexpected outcomes were uncertain.	
Additional Comments:	This evaluation was on the effect of DBP administered via capsule to a chicken for 10 days. The dose increased by 1.5cc per kg body weight every other day for this period. This study received an unacceptable ranking due to the lack of negative control as well as because there was only one organism used in the study. No statistical analysis could be conducted because there was only one organism and no control.			

Overall Quality Determination**Uninformative**

Study Citation:	Cornell University, (1931). Report upon the toxicity of plasticizers.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Vertebrate; Avian; <i>Gallus gallus</i> ; cockerels; Juvenile			
Health Outcome:	Skin & Connective Tissue			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332948			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The DBP was identified by name and CASRN.	
	Metric 2: Test Substance Source	Low	The source of the DBP was not reported.	
	Metric 3: Test Substance Purity	Low	The purity/grade of the DBP was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	Uninformative	Study authors did not report the use of a negative control.	
	Metric 5: Negative Control Response	Uninformative	Study authors did not report the use of a negative control, so a negative control response was not reported.	
	Metric 6: Randomized Allocation	Low	It was not reported how the chickens were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The preparation of the DBP for the capsule that was administered was not reported.	
	Metric 8: Consistency of Exposure Administration	Low	Reporting omissions are likely to have a substantial impact on results. The cockerels were fed capsules daily for 11 days (until death) with at a dose of 1.8cc per kg of body weight.	
	Metric 9: Measurement of Test Substance Concentration	Low	Study authors did not report if the test doses were measured.	
	Metric 10: Exposure Duration and Frequency	Medium	The exposure duration was reported to 11 days with a dose of 1.8cc/kg body weight. The exposure was conducted until death of the bird.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	N/A	There was only one exposure level. It did not appear that the goal was to observe a dose dependent effect.	
	Metric 12: Testing at or Below Solubility Limit	N/A	The exposure was via diet.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the birds and the age were not reported. The chickens were reported to be cockerels that were still growing that weighed approximately 1.4kg. No other information was provided.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the birds were acclimated in any way to test conditions.	
	Metric 15: Number of Organisms and Replicates per Group	Uninformative	There was only one bird exposed in each chemical, which was insufficient for statistical analysis.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	No information on environmental conditions was provided.	
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Study Citation:	Cornell University, (1931). Report upon the toxicity of plasticizers.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Vertebrate; Avian; <i>Gallus gallus</i> ; cockerels; Juvenile			
Health Outcome:	Skin & Connective Tissue			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332948			
Domain	Metric	Rating	Comments	
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported. The chickens were examined after the study, but it was unclear what the protocols for this examination entailed. Details regarding the execution of the study protocol for outcome assessment were not reported. The cockerel was examined after death, which in the case of DBP, was an 11 day exposure period, but the protocol was not described in any way.
	Metric 18:	Consistency of Outcome Assessment	Low	
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. Environmental conditions were not reported, nor was it reported if the cockerel was acclimated. There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Uninformative	There was only one organism that was exposed to DBP and no control, so statistical comparison was not possible. Results were described in the text only. Insufficient information was provided to determine if excessive variability or unexpected outcomes occurred. There was only one organism exposed, so variability or unexpected outcomes were uncertain.
	Metric 22:	Reporting of Data	Low	
	Metric 23:	Explanation of Unexpected Outcomes	Low	
Additional Comments:	This evaluation was on the effect of DBP administered via capsule to a chicken for 10 days. The dose increased by 1.5cc per kg body weight every other day for this period. This study received an unacceptable ranking due to the lack of negative control as well as because there was only one organism used in the study. No statistical analysis could be conducted because there was only one organism and no control.			

Overall Quality Determination**Uninformative**

Study Citation:	Cornell University, (1931). Report upon the toxicity of plasticizers.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Vertebrate; Avian; <i>Gallus gallus</i> ; cockerels; Juvenile			
Health Outcome:	Musculoskeletal			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332948			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The DBP was identified by name and CASRN.	
	Metric 2: Test Substance Source	Low	The source of the DBP was not reported.	
	Metric 3: Test Substance Purity	Low	The purity/grade of the DBP was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	Uninformative	Study authors did not report the use of a negative control.	
	Metric 5: Negative Control Response	Uninformative	Study authors did not report the use of a negative control, so a negative control response was not reported.	
	Metric 6: Randomized Allocation	Low	It was not reported how the chickens were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The preparation of the DBP for the capsule that was administered was not reported.	
	Metric 8: Consistency of Exposure Administration	Low	Reporting omissions are likely to have a substantial impact on results. The cockerels were fed capsules daily for 11 days (until death) with at a dose of 1.8cc per kg of body weight.	
	Metric 9: Measurement of Test Substance Concentration	Low	Study authors did not report if the test doses were measured.	
	Metric 10: Exposure Duration and Frequency	Medium	The exposure duration was reported to 11 days with a dose of 1.8cc/kg body weight. The exposure was conducted until death of the bird.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	N/A	There was only one exposure level. It did not appear that the goal was to observe a dose dependent effect.	
	Metric 12: Testing at or Below Solubility Limit	N/A	The exposure was via diet.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the birds and the age were not reported. The chickens were reported to be cockerels that were still growing that weighed approximately 1.4kg. No other information was provided.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the birds were acclimated in any way to test conditions.	
	Metric 15: Number of Organisms and Replicates per Group	Uninformative	There was only one bird exposed in each chemical, which was insufficient for statistical analysis.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	No information on environmental conditions was provided.	
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Study Citation:	Cornell University, (1931). Report upon the toxicity of plasticizers.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Vertebrate; Avian; <i>Gallus gallus</i> ; cockerels; Juvenile			
Health Outcome:	Musculoskeletal			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332948			
Domain	Metric	Rating	Comments	
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported. The chickens were examined after the study, but it was unclear what the protocols for this examination entailed.
	Metric 18:	Consistency of Outcome Assessment	Low	
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. Environmental conditions were not reported, nor was it reported if the cockerel was acclimated.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Uninformative	There was only one organism that was exposed to DBP and no control, so statistical comparison was not possible.
	Metric 22:	Reporting of Data	Low	
	Metric 23:	Explanation of Unexpected Outcomes	Low	
Insufficient information was provided to determine if excessive variability or unexpected outcomes occurred. There was only one organism exposed, so variability or unexpected outcomes were uncertain.				
Additional Comments:	This evaluation was on the effect of DBP administered via capsule to a chicken for 10 days. The dose increased by 1.5cc per kg body weight every other day for this period. This study received an unacceptable ranking due to the lack of negative control as well as because there was only one organism used in the study. No statistical analysis could be conducted because there was only one organism and no control.			

Overall Quality Determination**Uninformative**

Study Citation:	Cornell University, (1931). Report upon the toxicity of plasticizers.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Vertebrate; Avian; <i>Gallus gallus</i> ; cockerels; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332948			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The DBP was identified by name and CASRN.
	Metric 2:	Test Substance Source	Low	The source of the DBP was not reported.
	Metric 3:	Test Substance Purity	Low	The purity/grade of the DBP was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Uninformative	Study authors did not report the use of a negative control.
	Metric 5:	Negative Control Response	Uninformative	Study authors did not report the use of a negative control, so a negative control response was not reported.
	Metric 6:	Randomized Allocation	Low	It was not reported how the chickens were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The preparation of the DBP for the capsule that was administered was not reported.
	Metric 8:	Consistency of Exposure Administration	Low	Reporting omissions are likely to have a substantial impact on results. The cockerels were fed capsules daily until death, which was 11 days in the case of the DBP. The dosage was 1.8cc per kg administered daily.
	Metric 9:	Measurement of Test Substance Concentration	Low	Study authors did not report if the test doses were measured.
	Metric 10:	Exposure Duration and Frequency	Medium	The exposure duration was reported as 11 days (until death) with a dosage of 1.8cc DBP daily. This was sufficient for observations to be made.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	There was only one exposure level. It did not appear that the goal was to observe a dose dependent effect.
	Metric 12:	Testing at or Below Solubility Limit	N/A	The exposure was via diet.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the birds and the age were not reported. The chickens were reported to be cockerels that were still growing that weighed approximately 1.4kg. No other information was provided.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the birds were acclimated in any way to test conditions.
	Metric 15:	Number of Organisms and Replicates per Group	Uninformative	There was only one bird exposed in each chemical, which was insufficient for statistical analysis.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	No information on environmental conditions was provided.
	Metric 17:	Outcome Assessment Methodology	High	The bird was monitored for mortality for 11 days.

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Study Citation:	Cornell University, (1931). Report upon the toxicity of plasticizers.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Vertebrate; Avian; <i>Gallus gallus</i> ; cockerels; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332948			
Domain	Metric	Rating	Comments	
	Metric 18: Consistency of Outcome Assessment	High	The bird was monitored for mortality for 11 days (until death occurred).	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. Environmental conditions were not reported, nor was it reported if the cockerel was acclimated.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	Uninformative	There was only one organism that was exposed to DBP and no control, so statistical comparison was not possible.	
	Metric 22: Reporting of Data	Low	Results were described in the text only in an unlabeled table.	
	Metric 23: Explanation of Unexpected Outcomes	Low	Insufficient information was provided to determine if excessive variability or unexpected outcomes occurred. There was only one organism exposed, so variability or unexpected outcomes were uncertain.	
Additional Comments:	This evaluation was on the effect of DBP administered via capsule to a chicken for 11 days. The dose was reported to be 1.8cc per kg body weight until death. This study received an unacceptable ranking due to the lack of negative control as well as because there was only one organism used in the study. No statistical analysis could be conducted because there was only one organism and no control.			

Overall Quality Determination**Uninformative**

Study Citation:	Cornell University, (1931). Report upon the toxicity of plasticizers.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Vertebrate; Avian; <i>Gallus gallus</i> ; cockerels; Juvenile			
Health Outcome:	Renal/Kidney			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332948			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The DBP was identified by name and CASRN.	
	Metric 2: Test Substance Source	Low	The source of the DBP was not reported.	
	Metric 3: Test Substance Purity	Low	The purity/grade of the DBP was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	Uninformative	Study authors did not report the use of a negative control.	
	Metric 5: Negative Control Response	Uninformative	Study authors did not report the use of a negative control, so a negative control response was not reported.	
	Metric 6: Randomized Allocation	Low	It was not reported how the chickens were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The preparation of the DBP for the capsule that was administered was not reported.	
	Metric 8: Consistency of Exposure Administration	Low	Reporting omissions are likely to have a substantial impact on results. The cockerels were fed capsules daily for 11 days (until death) with at a dose of 1.8cc per kg of body weight.	
	Metric 9: Measurement of Test Substance Concentration	Low	Study authors did not report if the test doses were measured.	
	Metric 10: Exposure Duration and Frequency	Medium	The exposure duration was reported to 11 days with a dose of 1.8cc/kg body weight. The exposure was conducted until death of the bird.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	N/A	There was only one exposure level. It did not appear that the goal was to observe a dose dependent effect.	
	Metric 12: Testing at or Below Solubility Limit	N/A	The exposure was via diet.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the birds and the age were not reported. The chickens were reported to be cockerels that were still growing that weighed approximately 1.4kg. No other information was provided.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the birds were acclimated in any way to test conditions.	
	Metric 15: Number of Organisms and Replicates per Group	Uninformative	There was only one bird exposed in each chemical, which was insufficient for statistical analysis.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	No information on environmental conditions was provided.	
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Study Citation:	Cornell University, (1931). Report upon the toxicity of plasticizers.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Vertebrate; Avian; <i>Gallus gallus</i> ; cockerels; Juvenile			
Health Outcome:	Renal/Kidney			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332948			
Domain	Metric	Rating	Comments	
	Metric 17: Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported. The chickens were examined after the study, but it was unclear what the protocols for this examination entailed.	
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported. The cockerel was examined after death, which in the case of DBP, was an 11 day exposure period, but the protocol was not described in any way.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. Environmental conditions were not reported, nor was it reported if the cockerel was acclimated.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	Uninformative	There was only one organism that was exposed to DBP and no control, so statistical comparison was not possible.	
	Metric 22: Reporting of Data	Low	Results were described in the text only.	
	Metric 23: Explanation of Unexpected Outcomes	Low	Insufficient information was provided to determine if excessive variability or unexpected outcomes occurred. There was only one organism exposed, so variability or unexpected outcomes were uncertain.	
Additional Comments:	This evaluation was on the effect of DBP administered via capsule to a chicken for 10 days. The dose increased by 1.5cc per kg body weight every other day for this period. This study received an unacceptable ranking due to the lack of negative control as well as because there was only one organism used in the study. No statistical analysis could be conducted because there was only one organism and no control. The study reported that the affected bird showed effects on the gastrointestinal tract including the gizzard, stomach, small intestine, and gall bladder. Effects on other organ systems included the spleen, kidney, and liver.			

Overall Quality Determination**Uninformative**

Study Citation:	Cornell University, (1931). Report upon the toxicity of plasticizers.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Vertebrate; Avian; <i>Gallus gallus</i> ; cockerels; Juvenile			
Health Outcome:	Gastrointestinal			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332948			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The DBP was identified by name and CASRN.	
	Metric 2: Test Substance Source	Low	The source of the DBP was not reported.	
	Metric 3: Test Substance Purity	Low	The purity/grade of the DBP was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	Uninformative	Study authors did not report the use of a negative control.	
	Metric 5: Negative Control Response	Uninformative	Study authors did not report the use of a negative control, so a negative control response was not reported.	
	Metric 6: Randomized Allocation	Low	It was not reported how the chickens were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The preparation of the DBP for the capsule that was administered was not reported.	
	Metric 8: Consistency of Exposure Administration	Low	Reporting omissions are likely to have a substantial impact on results. The cockerels were fed capsules daily for 11 days (until death) with at a dose of 1.8cc per kg of body weight.	
	Metric 9: Measurement of Test Substance Concentration	Low	Study authors did not report if the test doses were measured.	
	Metric 10: Exposure Duration and Frequency	Medium	The exposure duration was reported to 11 days with a dose of 1.8cc/kg body weight. The exposure was conducted until death of the bird.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	N/A	There was only one exposure level. It did not appear that the goal was to observe a dose dependent effect.	
	Metric 12: Testing at or Below Solubility Limit	N/A	The exposure was via diet.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the birds and the age were not reported. The chickens were reported to be cockerels that were still growing that weighed approximately 1.4kg. No other information was provided.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the birds were acclimated in any way to test conditions.	
	Metric 15: Number of Organisms and Replicates per Group	Uninformative	There was only one bird exposed in each chemical, which was insufficient for statistical analysis.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	No information on environmental conditions was provided.	
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Study Citation:	Cornell University, (1931). Report upon the toxicity of plasticizers.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Vertebrate; Avian; <i>Gallus gallus</i> ; cockerels; Juvenile			
Health Outcome:	Gastrointestinal			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332948			
Domain	Metric	Rating	Comments	
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported. The chickens were examined after the study, but it was unclear what the protocols for this examination entailed.
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported. The cockerel was examined after death, which in the case of DBP, was an 11 day exposure period, but the protocol was not described in any way.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. Environmental conditions were not reported, nor was it reported if the cockerel was acclimated.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Uninformative	There was only one organism that was exposed to DBP and no control, so statistical comparison was not possible.
	Metric 22:	Reporting of Data	Low	Results were described in the text only.
	Metric 23:	Explanation of Unexpected Outcomes	Low	Insufficient information was provided to determine if excessive variability or unexpected outcomes occurred. There was only one organism exposed, so variability or unexpected outcomes were uncertain.
Additional Comments:	This evaluation was on the effect of DBP administered via capsule to a chicken for 10 days. The dose increased by 1.5cc per kg body weight every other day for this period. This study received an unacceptable ranking due to the lack of negative control as well as because there was only one organism used in the study. No statistical analysis could be conducted because there was only one organism and no control. The study reported that the affected bird showed effects on the gastrointestinal tract including the gizzard, stomach, small intestine, and gall bladder. Effects on other organ systems included the spleen, kidney, and liver.			

Overall Quality Determination**Uninformative**

Study Citation:	Cornell University, (1931). Report upon the toxicity of plasticizers.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Vertebrate; Avian; <i>Gallus gallus</i> ; cockerels; Juvenile			
Health Outcome:	Hepatic/Liver			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332948			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The DBP was identified by name and CASRN.	
	Metric 2: Test Substance Source	Low	The source of the DBP was not reported.	
	Metric 3: Test Substance Purity	Low	The purity/grade of the DBP was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	Uninformative	Study authors did not report the use of a negative control.	
	Metric 5: Negative Control Response	Uninformative	Study authors did not report the use of a negative control, so a negative control response was not reported.	
	Metric 6: Randomized Allocation	Low	It was not reported how the chickens were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The preparation of the DBP for the capsule that was administered was not reported.	
	Metric 8: Consistency of Exposure Administration	Low	Reporting omissions are likely to have a substantial impact on results. The cockerels were fed capsules daily for 11 days (until death) with at a dose of 1.8cc per kg of body weight.	
	Metric 9: Measurement of Test Substance Concentration	Low	Study authors did not report if the test doses were measured.	
	Metric 10: Exposure Duration and Frequency	Medium	The exposure duration was reported to 11 days with a dose of 1.8cc/kg body weight. The exposure was conducted until death of the bird.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	N/A	There was only one exposure level. It did not appear that the goal was to observe a dose dependent effect.	
	Metric 12: Testing at or Below Solubility Limit	N/A	The exposure was via diet.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the birds and the age were not reported. The chickens were reported to be cockerels that were still growing that weighed approximately 1.4kg. No other information was provided.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the birds were acclimated in any way to test conditions.	
	Metric 15: Number of Organisms and Replicates per Group	Uninformative	There was only one bird exposed in each chemical, which was insufficient for statistical analysis.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	No information on environmental conditions was provided.	
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Study Citation:	Cornell University, (1931). Report upon the toxicity of plasticizers.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Vertebrate; Avian; <i>Gallus gallus</i> ; cockerels; Juvenile			
Health Outcome:	Hepatic/Liver			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332948			
Domain	Metric	Rating	Comments	
	Metric 17: Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported. The chickens were examined after the study, but it was unclear what the protocols for this examination entailed.	
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported. The cockerel was examined after death, which in the case of DBP, was an 11 day exposure period, but the protocol was not described in any way.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. Environmental conditions were not reported, nor was it reported if the cockerel was acclimated.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	Uninformative	There was only one organism that was exposed to DBP and no control, so statistical comparison was not possible.	
	Metric 22: Reporting of Data	Low	Results were described in the text only.	
	Metric 23: Explanation of Unexpected Outcomes	Low	Insufficient information was provided to determine if excessive variability or unexpected outcomes occurred. There was only one organism exposed, so variability or unexpected outcomes were uncertain.	
Additional Comments:	This evaluation was on the effect of DBP administered via capsule to a chicken for 10 days. The dose increased by 1.5cc per kg body weight every other day for this period. This study received an unacceptable ranking due to the lack of negative control as well as because there was only one organism used in the study. No statistical analysis could be conducted because there was only one organism and no control. The study reported that the affected bird showed effects on the gastrointestinal tract including the gizzard, stomach, small intestine, and gall bladder. Effects on other organ systems included the spleen, kidney, and liver.			

Overall Quality Determination**Uninformative**

Study Citation:	Cornell University, (1931). Report upon the toxicity of plasticizers.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Vertebrate; Avian; <i>Gallus gallus</i> ; cockerels; Juvenile			
Health Outcome:	Immune/Hematological			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332948			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The DBP was identified by name and CASRN.	
	Metric 2: Test Substance Source	Low	The source of the DBP was not reported.	
	Metric 3: Test Substance Purity	Low	The purity/grade of the DBP was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	Uninformative	Study authors did not report the use of a negative control.	
	Metric 5: Negative Control Response	Uninformative	Study authors did not report the use of a negative control, so a negative control response was not reported.	
	Metric 6: Randomized Allocation	Low	It was not reported how the chickens were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The preparation of the DBP for the capsule that was administered was not reported.	
	Metric 8: Consistency of Exposure Administration	Low	Reporting omissions are likely to have a substantial impact on results. The cockerels were fed capsules daily for 11 days (until death) with at a dose of 1.8cc per kg of body weight.	
	Metric 9: Measurement of Test Substance Concentration	Low	Study authors did not report if the test doses were measured.	
	Metric 10: Exposure Duration and Frequency	Medium	The exposure duration was reported to 11 days with a dose of 1.8cc/kg body weight. The exposure was conducted until death of the bird.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	N/A	There was only one exposure level. It did not appear that the goal was to observe a dose dependent effect.	
	Metric 12: Testing at or Below Solubility Limit	N/A	The exposure was via diet.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the birds and the age were not reported. The chickens were reported to be cockerels that were still growing that weighed approximately 1.4kg. No other information was provided.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the birds were acclimated in any way to test conditions.	
	Metric 15: Number of Organisms and Replicates per Group	Uninformative	There was only one bird exposed in each chemical, which was insufficient for statistical analysis.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	No information on environmental conditions was provided.	
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Study Citation:	Cornell University, (1931). Report upon the toxicity of plasticizers.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Vertebrate; Avian; <i>Gallus gallus</i> ; cockerels; Juvenile			
Health Outcome:	Immune/Hematological			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332948			
Domain	Metric	Rating	Comments	
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported. The chickens were examined after the study, but it was unclear what the protocols for this examination entailed.
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported. The cockerel was examined after death, which in the case of DBP, was an 11 day exposure period, but the protocol was not described in any way.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. Environmental conditions were not reported, nor was it reported if the cockerel was acclimated.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Uninformative	There was only one organism that was exposed to DBP and no control, so statistical comparison was not possible.
	Metric 22:	Reporting of Data	Low	Results were described in the text only.
	Metric 23:	Explanation of Unexpected Outcomes	Low	Insufficient information was provided to determine if excessive variability or unexpected outcomes occurred. There was only one organism exposed, so variability or unexpected outcomes were uncertain.
Additional Comments:	This evaluation was on the effect of DBP administered via capsule to a chicken for 10 days. The dose increased by 1.5cc per kg body weight every other day for this period. This study received an unacceptable ranking due to the lack of negative control as well as because there was only one organism used in the study. No statistical analysis could be conducted because there was only one organism and no control. The study reported that the affected bird showed effects on the gastrointestinal tract including the gizzard, stomach, small intestine, and gall bladder. Effects on other organ systems included the spleen, kidney, and liver.			

Overall Quality Determination**Uninformative**

Study Citation:	Abdul-Ghani, S., Yanai, J., Abdul-Ghani, R., Pinkas, A., Abdeen, Z. (2012). The teratogenicity and behavioral teratogenicity of di(2-ethylhexyl) phthalate (DEHP) and di-butyl Phthalate (DBP) in a chick model. Neurotoxicology and Teratology 34(1):56-62.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection			
Taxa, Species, Age:	Vertebrate; Avian; <i>Gallus gallus domesticus</i> ; Cobb broiler strain; Embryo			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Genotox (including DNA repair)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1249807			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The chemical substance was identified by name.
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	The chemical purity was reported as 99.7%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was adequate.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured or measurements were not reported.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	Only one test concentration and a control were used.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via egg injection.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.

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Study Citation:	Abdul-Ghani, S., Yanai, J., Abdul-Ghani, R., Pinkas, A., Abdeen, Z. (2012). The teratogenicity and behavioral teratogenicity of di(2-ethylhexyl) phthalate (DEHP) and di-butyl Phthalate (DBP) in a chick model. Neurotoxicology and Teratology 34(1):56-62.
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection
Taxa, Species, Age:	Vertebrate; Avian; <i>Gallus gallus domesticus</i> ; Cobb broiler strain; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Genotox (including DNA repair)
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	1249807

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	High	There were no differences among groups reported.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: DNA damage

Overall Quality Determination**High**

Study Citation:	Abdul-Ghani, S., Yanai, J., Abdul-Ghani, R., Pinkas, A., Abdeen, Z. (2012). The teratogenicity and behavioral teratogenicity of di(2-ethylhexyl) phthalate (DEHP) and di-butyl Phthalate (DBP) in a chick model. Neurotoxicology and Teratology 34(1):56-62.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection			
Taxa, Species, Age:	Vertebrate; Avian; <i>Gallus gallus domesticus</i> ; Cobb broiler strain; Embryo			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1249807			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The chemical was identified by name.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity was reported as 99.7%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was adequate.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured or measurements were not reported.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	N/A	Only one test concentration and a control group were used.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via egg injection.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organism.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.	

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Study Citation:	Abdul-Ghani, S., Yanai, J., Abdul-Ghani, R., Pinkas, A., Abdeen, Z. (2012). The teratogenicity and behavioral teratogenicity of di(2-ethylhexyl) phthalate (DEHP) and di-butyl Phthalate (DBP) in a chick model. Neurotoxicology and Teratology 34(1):56-62.		
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported		
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection		
Taxa, Species, Age:	Vertebrate; Avian; <i>Gallus gallus domesticus</i> ; Cobb broiler strain; Embryo		
Health Outcome:	Development/Growth		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	1249807		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	High	There were no outcomes unrelated to exposures among test groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments: hatch, defects			
Overall Quality Determination		High	

Study Citation:	Abdul-Ghani, S., Yanai, J., Abdul-Ghani, R., Pinkas, A., Abdeen, Z. (2012). The teratogenicity and behavioral teratogenicity of di(2-ethylhexyl) phthalate (DEHP) and di-butyl Phthalate (DBP) in a chick model. Neurotoxicology and Teratology 34(1):56-62.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection			
Taxa, Species, Age:	Vertebrate; Avian; <i>Gallus gallus domesticus</i> ; Cobb broiler strain; Embryo			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1249807			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The chemical was identified by name.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity was reported as 99.7%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was adequate.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.	
	Metric 8: Consistency of Exposure	High	Exposures were administered consistently across study groups.	
	Metric 9: Administration Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured or measurements were not reported.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	N/A	Only one test concentration and a control group were used.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via egg injection.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.	

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Study Citation:	Abdul-Ghani, S., Yanai, J., Abdul-Ghani, R., Pinkas, A., Abdeen, Z. (2012). The teratogenicity and behavioral teratogenicity of di(2-ethylhexyl) phthalate (DEHP) and di-butyl Phthalate (DBP) in a chick model. Neurotoxicology and Teratology 34(1):56-62.		
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported		
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection		
Taxa, Species, Age:	Vertebrate; Avian; <i>Gallus gallus domesticus</i> ; Cobb broiler strain; Embryo		
Health Outcome:	Mortality		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	1249807		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	High	There were no outcomes unrelated to exposures among test groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments: Mortality added for hatching.			
Overall Quality Determination		High	

Study Citation:	Peakall, D. B. (1974). Effects of di-n-butyl and di-2-ethylhexyl phthalate on the eggs of ring doves. Bulletin of Environmental Contamination and Toxicology 12(6):698-702.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Vertebrate; Avian; <i>Streptopelia risoria</i> ; Adult			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	681729			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The test substance was identified by chemical name.
	Metric 2:	Test Substance Source	Low	The source was not reported and the test substance was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and grade of the test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	A concurrent negative control was included in the experiment.
	Metric 5:	Negative Control Response	High	The biological responses of the negative control group were reported.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Uninformative	The testing methods were not described in the paper and the references cited in the methods are only for maintenance of cages (HERO ID: 3061674) and for outcome assessment methods. No information was given on how the diets were dosed with DEHP, feeding schedule, etc. Concentration of the test substance was not measured during the study.
	Metric 8:	Consistency of Exposure Administration	Low	Details of exposure administration were not reported.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	Uninformative	The duration of exposure and exposure frequency were not reported.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Only one exposure group (10ppm DNB) was used to assess eggshell thickness, weight, rate of water loss, surface area and permeability. For DBP, eggshell thickness and weight were assessed for a group that were given 10 ppm DNB diet followed by clean food (Table 1).
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via diet.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study does not report whether pretreatment conditions were the same for control and exposed groups.
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Study Citation:	Peakall, D. B. (1974). Effects of di-n-butyl and di-2-ethylhexyl phthalate on the eggs of ring doves. Bulletin of Environmental Contamination and Toxicology 12(6):698-702.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Vertebrate; Avian; <i>Streptopelia risoria</i> ; Adult			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	681729			
Domain		Metric	Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of dove pairs and replicates used were not reported. But it was reported that a total of 34 eggs were used to determine eggshell thickness index, weight, rate of water loss, surface area and permeability (Table 1).
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Pairs of ring doves were maintained as described in Peakall,1970. Feeding schedule was not provided.
	Metric 17:	Outcome Assessment Methodology	Medium	The outcome assessment methodologies were briefly described. References were cited for eggshell index calculation (Ratcliffe, D.A., 1970) and permeability determination (HERO ID: 2180519; pdf not available).
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment (e.g., timing of assessment across groups) were not reported.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information on outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for the treatment and control group.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments:	No details given for the dietary toxicity testing methods. The duration of exposure and exposure frequency, and the number of pairs of doves used in the experiment were not reported.			

Overall Quality Determination**Uninformative**

Study Citation:	Shin, N., Cuenca, L., Karthikraj, R., Kannan, K., Colaiácovo, M. P. (2019). Assessing effects of germline exposure to environmental toxicants by high-throughput screening in <i>C. elegans</i> . <i>PLoS Genetics</i> 15(2):e1007975.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); <i>Caenorhabditis elegans</i> ; Larvae			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5043459			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The chemical was identified by name and structure.
	Metric 2:	Test Substance Source	High	The test substance identity was verified by GC-MS.
	Metric 3:	Test Substance Purity	Low	Purity or grade of the test substance was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups. Multiple strains were tested, but it is unclear which strain results were reported for various tests.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.
	Metric 8:	Consistency of Exposure Administration	Low	Only general methods of exposure administration were reported so assessment was difficult to determine.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Four concentrations over an adequate range were used.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The original source was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Medium	All pretreatment conditions appeared to be the same for control and exposed organisms but few details were provided. An external publication was cited for standard culture methods.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Three replicate exposures were reported.
Domain 5: Outcome Assessment				

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Study Citation:	Shin, N., Cuenca, L., Karthikraj, R., Kannan, K., Colaiácovo, M. P. (2019). Assessing effects of germline exposure to environmental toxicants by high-throughput screening in <i>C. elegans</i> . <i>Pl o S Genetics</i> 15(2):e1007975.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); <i>Caenorhabditis elegans</i> ; Larvae			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5043459			
Domain	Metric	Rating	Comments	
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate. The paper referred to "standard conditions" and cited an external source, and did not report conditions other than temperature (20C).
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest as described in the methods section, "Scoring embryonic lethality, larval lethality, and sterility"
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups as described in the methods section, "Scoring embryonic lethality, larval lethality, and sterility."
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments:	Multiple strains were tested, but it is unclear which strain results were reported. This form is for reporting embryonic lethality, larval lethality, and progeny counts after adults were exposed to DBP. The reproductive/teratogenic outcome was selected.			

Overall Quality Determination**Medium**

Study Citation:	Shin, N., Cuenca, L., Karthikraj, R., Kannan, K., Colaiácovo, M. P. (2019). Assessing effects of germline exposure to environmental toxicants by high-throughput screening in C. elegans. Pl o S Genetics 15(2):e1007975.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); <i>Caenorhabditis elegans</i> ; Larvae			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Genotox (including DNA repair)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5043459			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	Chemical identified by name and structure	
	Metric 2: Test Substance Source	High	The test substance identity was verified by GC-MS.	
	Metric 3: Test Substance Purity	Low	Purity or grade of test substance was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups. Multiple strains tested, unclear which strain results were reported for various tests	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations	
	Metric 8: Consistency of Exposure Administration	Low	Only general methods of exposure administration were reported so assessment was difficult to determine	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	Four concentrations over an adequate range	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The original source was not reported	
	Metric 14: Acclimatization and Pretreatment Conditions	Medium	all pretreatment conditions appeared to be the same for control and exposed organisms but few details were provided	
	Metric 15: Number of Organisms and Replicates per Group	Medium	Three replicate exposures were reported	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest	
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Study Citation:	Shin, N., Cuenca, L., Karthikraj, R., Kannan, K., Colaiácovo, M. P. (2019). Assessing effects of germline exposure to environmental toxicants by high-throughput screening in <i>C. elegans</i> . <i>PLoS Genetics</i> 15(2):e1007975.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); <i>Caenorhabditis elegans</i> ; Larvae
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Genotox (including DNA repair)
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	5043459

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described
	Metric 22: Reporting of Data	Medium	Data for exposure-related findings were presented for each treatment and control group in most cases. It was unclear if there were multiple tests or if only the results from one concentration were reported for some assessed endpoints
	Metric 23: Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained

Additional Comments: Multiple strains tested, unclear which strain results were reported

Overall Quality Determination

Medium

Study Citation:	Shin, N., Cuenca, L., Karthikraj, R., Kannan, K., Colaiácovo, M. P. (2019). Assessing effects of germline exposure to environmental toxicants by high-throughput screening in <i>C. elegans</i> . <i>PLoS Genetics</i> 15(2):e1007975.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); <i>Caenorhabditis elegans</i> ; Larvae
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	5043459

Domain	Metric	Rating	Comments
Domain 1: Test Substance			
	Metric 1: Test Substance Identity	High	Chemical identified by name and structure
	Metric 2: Test Substance Source	High	The test substance identity was verified by GC-MS.
	Metric 3: Test Substance Purity	Low	Purity or grade of test substance was not reported.
Domain 2: Test Design			
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups. Multiple strains tested, unclear which strain results were reported for various tests
Domain 3: Exposure Characterization			
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8: Consistency of Exposure Administration	Low	Only general methods of exposure administration were reported so assessment was difficult to determine
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	Four concentrations over an adequate range
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism			
	Metric 13: Test Organism Characteristics	Low	The original source was not reported
	Metric 14: Acclimatization and Pretreatment Conditions	Medium	All pretreatment conditions appeared to be the same for control and exposed organisms but few details were provided. An external publication was cited for standard culture methods.
	Metric 15: Number of Organisms and Replicates per Group	Medium	Three replicate exposures were reported.
Domain 5: Outcome Assessment			

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Study Citation:	Shin, N., Cuenca, L., Karthikraj, R., Kannan, K., Colaiácovo, M. P. (2019). Assessing effects of germline exposure to environmental toxicants by high-throughput screening in <i>C. elegans</i> . <i>PLoS Genetics</i> 15(2):e1007975.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); <i>Caenorhabditis elegans</i> ; Larvae
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	5043459

Domain	Metric	Rating	Comments
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate. The paper referred to "standard conditions" and cited an external source, and did not report conditions other than temperature (20C).
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest as described in the methods section, "Scoring embryonic lethality, larval lethality, and sterility"
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups as described in the methods section, "Scoring embryonic lethality, larval lethality, and sterility"
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: Multiple strains tested, unclear which strain results were reported

Overall Quality Determination

Medium

Study Citation:	Shin, N., Cuenca, L., Karthikraj, R., Kannan, K., Colaiácovo, M. P. (2019). Assessing effects of germline exposure to environmental toxicants by high-throughput screening in <i>C. elegans</i> . <i>PLoS Genetics</i> 15(2):e1007975.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); <i>Caenorhabditis elegans</i> ; Larvae
Health Outcome:	ADME (biotransformation)
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	5043459

Domain	Metric	Rating	Comments
Domain 1: Test Substance			
	Metric 1: Test Substance Identity	High	Chemical identified by name and structure
	Metric 2: Test Substance Source	High	The test substance identity was verified by GC-MS.
	Metric 3: Test Substance Purity	Low	Purity or grade of test substance was not reported.
Domain 2: Test Design			
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups. Multiple strains tested, unclear which strain results were reported for various tests
Domain 3: Exposure Characterization			
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8: Consistency of Exposure Administration	Low	Only general methods of exposure administration were reported so assessment was difficult to determine
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	Four concentrations over an adequate range
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism			
	Metric 13: Test Organism Characteristics	Low	The original source was not reported
	Metric 14: Acclimatization and Pretreatment Conditions	Medium	All pretreatment conditions appeared to be the same for control and exposed organisms but few details were provided. An external publication was cited for standard culture methods.
	Metric 15: Number of Organisms and Replicates per Group	Medium	Three replicate exposures were reported.
Domain 5: Outcome Assessment			

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Study Citation:	Shin, N., Cuenca, L., Karthikraj, R., Kannan, K., Colaiácovo, M. P. (2019). Assessing effects of germline exposure to environmental toxicants by high-throughput screening in <i>C. elegans</i> . <i>PLoS Genetics</i> 15(2):e1007975.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); <i>Caenorhabditis elegans</i> ; Larvae
Health Outcome:	ADME (biotransformation)
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	5043459

Domain	Metric	Rating	Comments
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate. The paper referred to "standard conditions" and cited an external source, and did not report conditions other than temperature (20C).
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest as described in the methods section, "Scoring embryonic lethality, larval lethality, and sterility"
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups as described in the methods section, "Scoring embryonic lethality, larval lethality, and sterility"
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: Multiple strains tested, unclear which strain results were reported. This form is for reporting the ADME outcome in Fig 7 of the reference.

Overall Quality Determination

Medium

Study Citation:	Tseng, I. L., Yang, Y. F., Yu, C. W., Li, W. H., Liao, C., V.H. (2013). Phthalates induce neurotoxicity affecting locomotor and thermotactic behaviors and AFD neurons through oxidative stress in <i>Caenorhabditis elegans</i> . PLoS ONE 8(12):e82657.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); <i>Caenorhabditis elegans</i> ; wild type; Larvae			
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2215375			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The chemical was identified by name.
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of the test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using a concurrent negative control.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was adequate.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	Low	The duration of exposure was reported and adequate for the study type, but there was "No Effect level."
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	The number of exposure groups and spacing of exposure levels were adequate for a dose response but there was "No Effect level"
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via culture medium.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of test organisms was not reported, but repeated tests were used as replicates (n=3).
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.

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Study Citation:	Tseng, I. L., Yang, Y. F., Yu, C. W., Li, W. H., Liao, C., V.H. (2013). Phthalates induce neurotoxicity affecting locomotor and thermotactic behaviors and AFD neurons through oxidative stress in <i>Caenorhabditis elegans</i> . PLoS ONE 8(12):e82657.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); <i>Caenorhabditis elegans</i> ; wild type; Larvae
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	2215375

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	High	There were no differences among groups that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group and were adequate to determine values for the endpoint(s) of interest.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments:	Ethanol pretreatment, reactive oxygen species		

Overall Quality Determination**High**

Study Citation:	Tseng, I. L., Yang, Y. F., Yu, C. W., Li, W. H., Liao, C., V.H. (2013). Phthalates induce neurotoxicity affecting locomotor and thermotactic behaviors and AFD neurons through oxidative stress in <i>Caenorhabditis elegans</i> . PLoS ONE 8(12):e82657.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); <i>Caenorhabditis elegans</i> ; wild type; Larvae			
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2215375			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using a concurrent negative control
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was adequate
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure	High	exposures were administered consistently across study groups
	Metric 9:	Administration Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	Low	The duration of exposure was reported and adequate for the study type, but there was no "No Effect level"
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	The number of exposure groups and spacing of exposure levels were adequate for a dose response but there was no "No Effect level"
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via culture medium
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of test organisms was not reported, repeated tests were used as replicates (n=3)
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups

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Study Citation:	Tseng, I. L., Yang, Y. F., Yu, C. W., Li, W. H., Liao, C., V.H. (2013). Phthalates induce neurotoxicity affecting locomotor and thermotactic behaviors and AFD neurons through oxidative stress in <i>Caenorhabditis elegans</i> . PLoS ONE 8(12):e82657.		
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)		
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); <i>Caenorhabditis elegans</i> ; wild type; Larvae		
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	2215375		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
	Metric 20: Outcomes Unrelated to Exposure	High	there were no differences among groups that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group and were adequate to determine values for the endpoint(s) of interest
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes
Additional Comments: Ascorbic acid pretreatment, reactive oxygen species			

Overall Quality Determination**High**

Study Citation:	Tseng, I. L., Yang, Y. F., Yu, C. W., Li, W. H., Liao, C., V.H. (2013). Phthalates induce neurotoxicity affecting locomotor and thermotactic behaviors and AFD neurons through oxidative stress in <i>Caenorhabditis elegans</i> . PLoS ONE 8(12):e82657.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); <i>Caenorhabditis elegans</i> ; DA 1267; Larvae			
Health Outcome:	Mechanistic-Neurotoxicology-Ocular and Sensory			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2215375			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using a concurrent negative control
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was adequate
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure	High	exposures were administered consistently across study groups
	Metric 9:	Administration Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	Low	The duration of exposure was reported and adequate for the study type, but there was no "No Effect level"
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	The number of exposure groups and spacing of exposure levels were adequate for a dose response but there was no "No Effect level"
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via culture medium
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of test organisms was not reported, repeated tests were used as replicates (n=3)
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups

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Study Citation:	Tseng, I. L., Yang, Y. F., Yu, C. W., Li, W. H., Liao, C., V.H. (2013). Phthalates induce neurotoxicity affecting locomotor and thermotactic behaviors and AFD neurons through oxidative stress in <i>Caenorhabditis elegans</i> . PLoS ONE 8(12):e82657.		
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)		
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); <i>Caenorhabditis elegans</i> ; DA 1267; Larvae		
Health Outcome:	Mechanistic-Neurotoxicology-Ocular and Sensory		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	2215375		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
	Metric 20: Outcomes Unrelated to Exposure	High	there were no differences among groups that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group and were adequate to determine values for the endpoint(s) of interest
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes
Additional Comments: AFD thermosensory neurons			

Overall Quality Determination**High**

Study Citation:	Tseng, I. L., Yang, Y. F., Yu, C. W., Li, W. H., Liao, C., V.H. (2013). Phthalates induce neurotoxicity affecting locomotor and thermotactic behaviors and AFD neurons through oxidative stress in <i>Caenorhabditis elegans</i> . PLoS ONE 8(12):e82657.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); <i>Caenorhabditis elegans</i> ; wild type; Larvae			
Health Outcome:	Behavioral			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2215375			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	Chemical was identified by name	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Low	Purity and/or grade of test substance were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was adequate	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail	
	Metric 8: Consistency of Exposure	High	exposures were administered consistently across study groups	
	Metric 9: Administration Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured	
	Metric 10: Exposure Duration and Frequency	Medium	The duration of exposure was reported and adequate for the study type	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were adequate for a dose response	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via culture medium	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms	
	Metric 15: Number of Organisms and Replicates per Group	Low	The number of test organisms was not reported, repeated tests were used as replicates (n=3)	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest	
	Metric 18: Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups	
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Study Citation:	Tseng, I. L., Yang, Y. F., Yu, C. W., Li, W. H., Liao, C., V.H. (2013). Phthalates induce neurotoxicity affecting locomotor and thermotactic behaviors and AFD neurons through oxidative stress in <i>Caenorhabditis elegans</i> . PLoS ONE 8(12):e82657.		
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)		
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); <i>Caenorhabditis elegans</i> ; wild type; Larvae		
Health Outcome:	Behavioral		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	2215375		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
	Metric 20: Outcomes Unrelated to Exposure	High	there were no differences among groups that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group and were adequate to determine values for the endpoint(s) of interest
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes
Additional Comments: locomotor and thermotaxis			
Overall Quality Determination		High	

Study Citation:	Kang, S. W., Kim, H. K., Lee, W. J., Ahn, Y. J. (2006). Toxicity of bisabolangelone from <i>Ostericum koreanum</i> roots to <i>Dermatophagoides farinae</i> and <i>Dermatophagoides pteronyssinus</i> (Acari : Pyroglyphidae). Journal of Agricultural and Food Chemistry 54(10):3547-3550.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Dermatophagoides farinae</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	485854			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Low	Purity and/or grade of the test substance were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	High	No mortalities were reported in the negative control group.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The experimental system and methods for preparation of test media were described in adequate detail. Petri dishes were used, but it was not mentioned whether they were made of glass or plastic.	
	Metric 8: Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	Six concentrations were used.	
	Metric 12: Testing at or Below Solubility Limit	N/A	The chemical was applied directly to cotton cloth.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Medium	Organisms were from a lab culture, but origin of the colony was not reported.	
	Metric 14: Acclimatization and Pretreatment Conditions	Medium	Culture and test conditions were similar.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	Three replicates of 30-40 adults were used.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health. Temperature and relative humidity were reported.	
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Study Citation:	Kang, S. W., Kim, H. K., Lee, W. J., Ahn, Y. J. (2006). Toxicity of bisabolangelone from <i>Ostericum koreanum</i> roots to <i>Dermatophagoides farinae</i> and <i>Dermatophagoides pteronyssinus</i> (Acari : Pyroglyphidae). <i>Journal of Agricultural and Food Chemistry</i> 54(10):3547-3550.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Dermatophagoides farinae</i> ; Adult
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	485854

Domain	Metric	Rating	Comments
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group, but results were described in tables.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: None

Overall Quality Determination**Medium**

Study Citation:	Kang, S. W., Kim, H. K., Lee, W. J., Ahn, Y. J. (2006). Toxicity of bisabolangelone from <i>Ostericum koreanum</i> roots to <i>Dermatophagoides farinae</i> and <i>Dermatophagoides pteronyssinus</i> (Acari : Pyroglyphidae). Journal of Agricultural and Food Chemistry 54(10):3547-3550.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Dermatophagoides farinae</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	485854			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The chemical was identified by name only.
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	High	No mortalities were reported.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The experimental system and methods for preparation of test media were described in adequate detail. Petri dishes were used, but it was not mentioned whether they were made of glass or plastic.
	Metric 8:	Consistency of Exposure Administration	Medium	Details of exposure administration were reported and exposures were administered consistently across study groups, but because it was a vapor exposure, consistency is difficult to judge.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	Only one concentration was used, although both open and closed systems were evaluated.
	Metric 12:	Testing at or Below Solubility Limit	N/A	The chemical was applied directly to cotton cloth, and exposure was via vapors.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	Organisms were from a lab culture, but origin of the colony was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Medium	Culture and test conditions were similar.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Three replicates of 30-40 adults were used.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health. Temperature and relative humidity were reported.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
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Study Citation:	Kang, S. W., Kim, H. K., Lee, W. J., Ahn, Y. J. (2006). Toxicity of bisabolangelone from <i>Ostericum koreanum</i> roots to <i>Dermatophagoides farinae</i> and <i>Dermatophagoides pteronyssinus</i> (Acari : Pyroglyphidae). <i>Journal of Agricultural and Food Chemistry</i> 54(10):3547-3550.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Dermatophagoides farinae</i> ; Adult
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	485854

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Statistical methods were adequately described but results were not compared to controls; they were only compared to other chemicals.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were shown for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments: None			

Overall Quality Determination**Medium**

Study Citation:	Kim, H. K., Tak, J. H., Ahn, Y. J. (2004). Acaricidal activity of Paeonia suffruticosa root bark-derived compounds against Dermatophagoides farinae and Dermatophagoides pteronyssinus (Acari: Pyroglyphidae). Journal of Agricultural and Food Chemistry 52(26):7857-7861.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Air, N/A (e.g., injection); Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Dermatophagoides farinae</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332803			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The chemical was identified by name only.
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of the test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	High	No control mortalities were reported.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.
	Metric 8:	Consistency of Exposure Administration	Medium	A one-time dose was used, but few details were provided.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Only one concentration was tested.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via fabric contact.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The test used 25-35 mites with three replicates.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Control and treatment mites were held in conditions similar to rearing conditions.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed the intended outcome of interest.
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Study Citation:	Kim, H. K., Tak, J. H., Ahn, Y. J. (2004). Acaricidal activity of <i>Paeonia suffruticosa</i> root bark-derived compounds against <i>Dermatophagoides farinae</i> and <i>Dermatophagoides pteronyssinus</i> (Acari: Pyroglyphidae). <i>Journal of Agricultural and Food Chemistry</i> 52(26):7857-7861.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Air, N/A (e.g., injection); Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Dermatophagoides farinae</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332803			
Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	N/A	Statistical analysis was not compared to control values.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments: closed container, method A				

Overall Quality Determination**Medium**

Study Citation:	Kim, H. K., Tak, J. H., Ahn, Y. J. (2004). Acaricidal activity of <i>Paeonia suffruticosa</i> root bark-derived compounds against <i>Dermatophagoides farinae</i> and <i>Dermatophagoides pteronyssinus</i> (Acari: Pyroglyphidae). <i>Journal of Agricultural and Food Chemistry</i> 52(26):7857-7861.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Dermatophagoides farinae</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332803			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Low	Purity and/or grade of the test substance were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	Medium	A biological response by a control group was reported in Table 1, but it's unclear if this was the response of the control mentioned in the methods section.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.	
	Metric 8: Consistency of Exposure Administration	Medium	A one-time dose was used, but few details were provided.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	Eight concentrations over a suitable range were used.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via fabric contact.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The test used 25-30 mites with 3-5 replicates.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Medium	Control and treatment mites were held in conditions similar to rearing conditions.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed the intended outcome of interest.	
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.	

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Study Citation:	Kim, H. K., Tak, J. H., Ahn, Y. J. (2004). Acaricidal activity of <i>Paeonia suffruticosa</i> root bark-derived compounds against <i>Dermatophagoides farinae</i> and <i>Dermatophagoides pteronyssinus</i> (Acari: Pyroglyphidae). <i>Journal of Agricultural and Food Chemistry</i> 52(26):7857-7861.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Dermatophagoides farinae</i> ; Adult
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	1332803

Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group, but results were described in the tables.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments: None			

Overall Quality Determination**Medium**

Study Citation:	Kim, H. K., Yun, Y. K., Ahn, Y. J. (2008). Fumigant toxicity of cassia bark and cassia and cinnamon oil compounds to <i>Dermatophagoides farinae</i> and <i>Dermatophagoides pteronyssinus</i> (Acari: Pyroglyphidae). <i>Experimental and Applied Acarology</i> 44(1):1-9.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Dermatophagoides farinae</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1323180			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Low	Purity or grade of the test substance were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group.	
	Metric 5: Negative Control Response	Low	The biological response of the negative control group was not reported.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.	
	Metric 8: Consistency of Exposure Administration	Medium	A one-time dose was used, but few details were reported.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Low	Four to six treatments were used, but exposure levels were not reported.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via contact to treated fabric.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from an eight-year-old lab colony.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 25 mites/treatment with four replicates.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	

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Study Citation:	Kim, H. K., Yun, Y. K., Ahn, Y. J. (2008). Fumigant toxicity of cassia bark and cassia and cinnamon oil compounds to <i>Dermatophagoides farinae</i> and <i>Dermatophagoides pteronyssinus</i> (Acari: Pyroglyphidae). <i>Experimental and Applied Acarology</i> 44(1):1-9.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Dermatophagoides farinae</i> ; Adult
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	1323180

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	Low	Only resultant LD50s were reported.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This study gives dose in units of mg/m3 of felt. This may not be usable for terrestrial tox in the RE depending on exposure data available.

Overall Quality Determination

Medium

Study Citation:	Kim, H. K., Yun, Y. K., Ahn, Y. J. (2007). Toxicity of atractylon and atractylenolide III identified in Atractylodes ovata rhizome to Dermatophagoides farinae and Dermatophagoides pteronyssinus. Journal of Agricultural and Food Chemistry 55(15):6027-6031.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Dermatophagoides farinae</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1341977			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	High	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Low	Purity or grade of the test substance were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group.	
	Metric 5: Negative Control Response	High	In the Results section it is stated that "there was no mortality in the ethanol-treated controls."	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.	
	Metric 8: Consistency of Exposure Administration	Medium	Nothing in the study suggested that exposures were not administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via substrate contact.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	Mites were kept in similar conditions to the lab colony.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 25 mites with 4-6 replicates used.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	
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Study Citation:	Kim, H. K., Yun, Y. K., Ahn, Y. J. (2007). Toxicity of atractylon and atractylenolide III identified in Atractylodes ovata rhizome to Dermatophagoides farinae and Dermatophagoides pteronyssinus. Journal of Agricultural and Food Chemistry 55(15):6027-6031.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Dermatophagoides farinae</i> ; Adult
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	1341977

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group, but results were described in the tables.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments: None			

Overall Quality Determination**Medium**

Study Citation:	Wang, Z., Kim, H. K., Tao, W., Wang, M., Ahn, Y. J. (2011). Contact and fumigant toxicity of cinnamaldehyde and cinnamic acid and related compounds to <i>Dermatophagoides farinae</i> and <i>Dermatophagoides pteronyssinus</i> (Acari: Pyroglyphidae). <i>Journal of Medical Entomology</i> 48(2):366-371.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Air, N/A (e.g., injection); Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Dermatophagoides farinae</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	788260			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity was reported as 99%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	Low	The biological response of the negative control group was not reported.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.	
	Metric 8: Consistency of Exposure Administration	Medium	Only general methods of exposure administration were reported so assessment was difficult to determine.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type (24h acute).	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Four to six concentrations were used, but the range was not specified.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via treated fabric.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The organisms were sourced from a long-standing lab culture.	
	Metric 14: Acclimatization and Pretreatment Conditions	Medium	All pretreatment conditions were the same for control and exposed organisms, but few details were provided.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms was reported and sufficient to characterize toxicological effects (25 organisms, replicated 4 times per treatment).	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Medium	Temperature and relative humidity were reported and adequate.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	

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Study Citation:	Wang, Z., Kim, H. K., Tao, W., Wang, M., Ahn, Y. J. (2011). Contact and fumigant toxicity of cinnamaldehyde and cinnamic acid and related compounds to <i>Dermatophagoides farinae</i> and <i>Dermatophagoides pteronyssinus</i> (Acari: Pyroglyphidae). <i>Journal of Medical Entomology</i> 48(2):366-371.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; Air, N/A (e.g., injection); Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Dermatophagoides farinae</i> ; Adult
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	788260

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions across treatments.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group, but results were described in the table as LC50 values.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments: None			

Overall Quality Determination**Medium**

Study Citation:	Kang, S. W., Kim, H. K., Lee, W. J., Ahn, Y. J. (2006). Toxicity of bisabolangelone from <i>Ostericum koreanum</i> roots to <i>Dermatophagoides farinae</i> and <i>Dermatophagoides pteronyssinus</i> (Acari : Pyroglyphidae). Journal of Agricultural and Food Chemistry 54(10):3547-3550.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Dermatophagoides pteronyssinus</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	485854			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Low	Purity and/or grade of the test substance were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	High	No mortalities were reported.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The experimental system and methods for preparation of test media were described in adequate detail. Petri dishes were used, but it was not mentioned whether they were made of glass or plastic.	
	Metric 8: Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	Six concentrations were used.	
	Metric 12: Testing at or Below Solubility Limit	N/A	The chemical was applied directly to cotton cloth.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Medium	Organisms were from a lab culture, but the origin of colony was not reported.	
	Metric 14: Acclimatization and Pretreatment Conditions	Medium	Culture and test conditions were similar.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	Three replicates of 30-40 adults were used.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health. Temperature and relative humidity were reported.	
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Study Citation:	Kang, S. W., Kim, H. K., Lee, W. J., Ahn, Y. J. (2006). Toxicity of bisabolangelone from <i>Ostericum koreanum</i> roots to <i>Dermatophagoides farinae</i> and <i>Dermatophagoides pteronyssinus</i> (Acari : Pyroglyphidae). <i>Journal of Agricultural and Food Chemistry</i> 54(10):3547-3550.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Dermatophagoides pteronyssinus</i> ; Adult
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	485854

Domain	Metric	Rating	Comments
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group, but results were described in the tables.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: None

Overall Quality Determination**Medium**

Study Citation:	Kang, S. W., Kim, H. K., Lee, W. J., Ahn, Y. J. (2006). Toxicity of bisabolangelone from <i>Ostericum koreanum</i> roots to <i>Dermatophagoides farinae</i> and <i>Dermatophagoides pteronyssinus</i> (Acari : Pyroglyphidae). Journal of Agricultural and Food Chemistry 54(10):3547-3550.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Dermatophagoides pteronyssinus</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	485854			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The chemical was identified by name only.
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of the test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	High	No mortalities were reported.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail. Petri dishes were used, but it was not mentioned whether they were made of glass or plastic.
	Metric 8:	Consistency of Exposure Administration	Medium	Details of exposure administration were reported and exposures were administered consistently across study groups but because it was a vapor exposure, consistency is difficult to judge.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	Only one concentration was used, although both open and closed systems were evaluated.
	Metric 12:	Testing at or Below Solubility Limit	N/A	The chemical was applied directly to cotton cloth, and exposure was via vapors.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	Organisms were from a lab culture, but the origin of colony was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Medium	Culture and test conditions were similar.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Three replicates of 30-40 adults were used.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health. Temperature and relative humidity were reported.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
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Study Citation:	Kang, S. W., Kim, H. K., Lee, W. J., Ahn, Y. J. (2006). Toxicity of bisabolangelone from <i>Ostericum koreanum</i> roots to <i>Dermatophagoides farinae</i> and <i>Dermatophagoides pteronyssinus</i> (Acari : Pyroglyphidae). <i>Journal of Agricultural and Food Chemistry</i> 54(10):3547-3550.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Dermatophagoides pteronyssinus</i> ; Adult
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	485854

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Statistical methods were adequately described but results were not compared to controls; they were only compared to other chemicals.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were shown for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments: None			

Overall Quality Determination**Medium**

Study Citation:	Kim, H. K., Tak, J. H., Ahn, Y. J. (2004). Acaricidal activity of Paeonia suffruticosa root bark-derived compounds against Dermatophagoides farinae and Dermatophagoides pteronyssinus (Acari: Pyroglyphidae). Journal of Agricultural and Food Chemistry 52(26):7857-7861.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Dermatophagoides pteronyssinus</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332803			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Low	Purity and/or grade of the test substance were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	Medium	A biological response by a control group was reported in Table 1, but it is unclear if this was the response of the control mentioned in the methods section.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.	
	Metric 8: Consistency of Exposure Administration	Medium	A one-time dose was used, but few details were provided.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	Eight concentrations over a suitable range were used.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via fabric contact.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 25-30 mites with 3-5 replicates used.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Medium	Control and treatment mites were held in conditions similar to rearing conditions.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed the intended outcome of interest.	

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Study Citation:	Kim, H. K., Tak, J. H., Ahn, Y. J. (2004). Acaricidal activity of <i>Paeonia suffruticosa</i> root bark-derived compounds against <i>Dermatophagoides farinae</i> and <i>Dermatophagoides pteronyssinus</i> (Acari: Pyroglyphidae). <i>Journal of Agricultural and Food Chemistry</i> 52(26):7857-7861.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Dermatophagoides pteronyssinus</i> ; Adult
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	1332803

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group, but results were described in the tables.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments: None			

Overall Quality Determination**Medium**

Study Citation:	Kim, H. K., Tak, J. H., Ahn, Y. J. (2004). Acaricidal activity of Paeonia suffruticosa root bark-derived compounds against Dermatophagoides farinae and Dermatophagoides pteronyssinus (Acari: Pyroglyphidae). Journal of Agricultural and Food Chemistry 52(26):7857-7861.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Air, N/A (e.g., injection); Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Dermatophagoides pteronyssinus</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1332803			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Low	Purity and/or grade of the test substance were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	High	No control mortalities were reported.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.	
	Metric 8: Consistency of Exposure Administration	Medium	A one-time dose was used, but few details were provided.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	Only one concentration was tested.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via fabric contact.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 25-35 mites with three replicates used.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Medium	Control and treatment mites were held in conditions similar to rearing conditions.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed the intended outcome of interest.	
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.	
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Study Citation:	Kim, H. K., Tak, J. H., Ahn, Y. J. (2004). Acaricidal activity of Paeonia suffruticosa root bark-derived compounds against Dermatophagoides farinae and Dermatophagoides pteronyssinus (Acari: Pyroglyphidae). Journal of Agricultural and Food Chemistry 52(26):7857-7861.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; Air, N/A (e.g., injection); Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Dermatophagoides pteronyssinus</i> ; Adult
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	1332803

Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	N/A	Statistical analysis was not compared to control values.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: closed container, method A

Overall Quality Determination**Medium**

Study Citation:	Kim, H. K., Yun, Y. K., Ahn, Y. J. (2008). Fumigant toxicity of cassia bark and cassia and cinnamon oil compounds to Dermatophagoides farinae and Dermatophagoides pteronyssinus (Acari: Pyroglyphidae). Experimental and Applied Acarology 44(1):1-9.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Dermatophagoides pteronyssinus</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1323180			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Low	Purity or grade of the test substance were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group.	
	Metric 5: Negative Control Response	Low	The biological response of the negative control group was not reported.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.	
	Metric 8: Consistency of Exposure Administration	Medium	A one-time dose was used, but few details were reported.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Low	Four to six treatments were used, but exposure levels were not reported.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via contact to treated fabric.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from an eight-year-old lab colony.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 25 mites/treatment with four replicates used.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	
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Study Citation:	Kim, H. K., Yun, Y. K., Ahn, Y. J. (2008). Fumigant toxicity of cassia bark and cassia and cinnamon oil compounds to <i>Dermatophagoides farinae</i> and <i>Dermatophagoides pteronyssinus</i> (Acari: Pyroglyphidae). <i>Experimental and Applied Acarology</i> 44(1):1-9.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Dermatophagoides pteronyssinus</i> ; Adult
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	1323180

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	Low	Only resultant LD50s were reported.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This study gives dose in units of mg/m3 of felt. This may not be usable for terrestrial tox in the RE depending on exposure data available.

Overall Quality Determination

Medium

Study Citation:	Kim, H. K., Yun, Y. K., Ahn, Y. J. (2007). Toxicity of atractylon and atractylenolide III identified in Atractylodes ovata rhizome to Dermatophagoides farinae and Dermatophagoides pteronyssinus. Journal of Agricultural and Food Chemistry 55(15):6027-6031.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Dermatophagoides pteronyssinus</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1341977			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	High	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Low	Purity or grade of the test substance were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group.	
	Metric 5: Negative Control Response	High	In the Results section it is stated that "there was no mortality in the ethanol-treated controls."	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.	
	Metric 8: Consistency of Exposure Administration	Medium	Nothing in the study suggested that exposures were not administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via substrate contact.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	Mites were kept in similar conditions to the lab colony.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 25 mites with 4-6 replicates used.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	
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Study Citation:	Kim, H. K., Yun, Y. K., Ahn, Y. J. (2007). Toxicity of atractylon and atractylenolide III identified in Atractylodes ovata rhizome to Dermatophagoides farinae and Dermatophagoides pteronyssinus. Journal of Agricultural and Food Chemistry 55(15):6027-6031.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Dermatophagoides pteronyssinus</i> ; Adult
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	1341977

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group, but results were described in the tables.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments: None			

Overall Quality Determination**Medium**

Study Citation:	Wang, Z., Kim, H. K., Tao, W., Wang, M., Ahn, Y. J. (2011). Contact and fumigant toxicity of cinnamaldehyde and cinnamic acid and related compounds to <i>Dermatophagoides farinae</i> and <i>Dermatophagoides pteronyssinus</i> (Acari: Pyroglyphidae). <i>Journal of Medical Entomology</i> 48(2):366-371.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Air, N/A (e.g., injection); Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Dermatophagoides pteronyssinus</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	788260			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity was reported as 99%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	Low	The biological response of the negative control group was not reported.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.	
	Metric 8: Consistency of Exposure Administration	Medium	Only general methods of exposure administration were reported so assessment was difficult to determine.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Four to six concentrations were used, but a range was not specified.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via treated fabric.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	Organisms were sourced from a long-standing lab culture.	
	Metric 14: Acclimatization and Pretreatment Conditions	Medium	All pretreatment conditions were the same for control and exposed organisms, but few details were provided.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms was reported and sufficient to characterize toxicological effects (25 organisms, replicated 4 times per treatment).	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Medium	Temperature and relative humidity were reported and adequate.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	

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Study Citation:	Wang, Z., Kim, H. K., Tao, W., Wang, M., Ahn, Y. J. (2011). Contact and fumigant toxicity of cinnamaldehyde and cinnamic acid and related compounds to <i>Dermatophagoides farinae</i> and <i>Dermatophagoides pteronyssinus</i> (Acari: Pyroglyphidae). <i>Journal of Medical Entomology</i> 48(2):366-371.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; Air, N/A (e.g., injection); Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Dermatophagoides pteronyssinus</i> ; Adult
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	788260

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions across treatments.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group, but results were described in the table as LC50 values.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments: None			

Overall Quality Determination**Medium**

Study Citation:	Misra, S., Singh, A., Ch, R., Sharma, V., Mudiam, R., M.K., Ram, K. R. (2014). Identification of Drosophila-based endpoints for the assessment and understanding of xenobiotic-mediated male reproductive adversities. Toxicological Sciences 141(1):278-291.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Drosophila melanogaster</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2510760			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	Chemical was identified by name only	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Low	Purity or grade of test substance was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group	
	Metric 5: Negative Control Response	Low	The biological response of the negative control group was not reported	
	Metric 6: Randomized Allocation	Medium	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The study provided only limited details on the measures taken to appropriately prepare test concentrations although the test system was described in adequate detail. Glass vials were used.	
	Metric 8: Consistency of Exposure Administration	Medium	One time dose to the food, few other details provided	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured	
	Metric 10: Exposure Duration and Frequency	Low	It was unclear what the exposure duration was, it was based on eclosion time	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Low	No information is provided on the number of exposure groups and spacing of exposure levels although the range was 10 mM to 2 M	
	Metric 12: Testing at or Below Solubility Limit	N/A	exposure was via diet	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized	
	Metric 15: Number of Organisms and Replicates per Group	Low	The number of test organisms (100) was adequate.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate	
	Metric 17: Outcome Assessment Methodology	Medium	The outcome assessment methodology was not reported	
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Study Citation:	Misra, S., Singh, A., Ch, R., Sharma, V., Mudiam, R., M.K., Ram, K. R. (2014). Identification of <i>Drosophila</i> -based endpoints for the assessment and understanding of xenobiotic-mediated male reproductive adversities. <i>Toxicological Sciences</i> 141(1):278-291.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Drosophila melanogaster</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2510760			
Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described
	Metric 22:	Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group, but results were described in the text.
	Metric 23:	Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability
Additional Comments: None				
Overall Quality Determination			Low	

Study Citation:	Misra, S., Singh, A., Ch, R., Sharma, V., Mudiam, R., M.K., Ram, K. R. (2014). Identification of <i>Drosophila</i> -based endpoints for the assessment and understanding of xenobiotic-mediated male reproductive adversities. <i>Toxicological Sciences</i> 141(1):278-291.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Drosophila melanogaster</i> ; Embryo			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2510760			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Low	Purity or grade of the test substance was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group.	
	Metric 5: Negative Control Response	Low	The biological response of the negative control group was reported and reasonable.	
	Metric 6: Randomized Allocation	Medium	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The study provided only limited details on the measures taken to appropriately prepare test concentrations although the test system was described in adequate detail. Glass vials were used.	
	Metric 8: Consistency of Exposure Administration	Medium	A one-time dose was used to the food, but few other details were provided.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	Low	It was unclear what the exact exposure duration was, but it was based on eclosion time (>10 days).	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Medium	There were two exposure groups that were evenly spaced.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via diet.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The number of test organisms (100) was adequate.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.	
	Metric 17: Outcome Assessment Methodology	Medium	The outcome assessment methodology was not reported.	
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported.	

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Study Citation:	Misra, S., Singh, A., Ch, R., Sharma, V., Mudiam, R., M.K., Ram, K. R. (2014). Identification of <i>Drosophila</i> -based endpoints for the assessment and understanding of xenobiotic-mediated male reproductive adversities. <i>Toxicological Sciences</i> 141(1):278-291.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Drosophila melanogaster</i> ; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Reproductive/Teratogenic
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	2510760

Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	High	Statistical methods were adequately described.
Metric 22:	Reporting of Data	High	Data for exposure-related findings were shown for each treatment and control group.
Metric 23:	Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.
Additional Comments: Multiple studies were reported, which led to a lack of clarity regarding test set up and conditions.			

Overall Quality Determination**Medium**

Study Citation:	Misra, S., Singh, A., Ch, R., Sharma, V., Mudiam, R., M.K., Ram, K. R. (2014). Identification of <i>Drosophila</i> -based endpoints for the assessment and understanding of xenobiotic-mediated male reproductive adversities. <i>Toxicological Sciences</i> 141(1):278-291.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Drosophila melanogaster</i> ; Embryo			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2510760			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Low	Purity or grade of the test substance was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group.	
	Metric 5: Negative Control Response	Low	The biological response of the negative control group was reported and reasonable.	
	Metric 6: Randomized Allocation	Medium	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The study provided only limited details on the measures taken to appropriately prepare test concentrations although the test system was described in adequate detail. Glass vials were used.	
	Metric 8: Consistency of Exposure Administration	Medium	A one-time dose was used to the food, but few other details were provided.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	Low	It was unclear what the exact exposure duration was, but it was based on eclosion time (>10 days).	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Medium	Two exposure groups were used and they were adequately spaced apart.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via diet.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The number of test organisms (100) was adequate.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.	
	Metric 17: Outcome Assessment Methodology	Medium	The outcome assessment methodology was not reported.	

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Study Citation:	Misra, S., Singh, A., Ch, R., Sharma, V., Mudiam, R., M.K., Ram, K. R. (2014). Identification of <i>Drosophila</i> -based endpoints for the assessment and understanding of xenobiotic-mediated male reproductive adversities. <i>Toxicological Sciences</i> 141(1):278-291.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Drosophila melanogaster</i> ; Embryo
Health Outcome:	Reproductive/Teratogenic
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	2510760

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were shown for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.

Additional Comments: Multiple studies were reported, which led to a lack of clarity regarding test set up and conditions.

Overall Quality Determination

Medium

Study Citation:	Williams, M. J., Wiemerslage, L., Gohel, P., Kheder, S., Kothegala, L. V., Schioth, H. B. (2016). Dibutyl Phthalate Exposure Disrupts Evolutionarily Conserved Insulin and Glucagon-Like Signaling in <i>Drosophila</i> Males. <i>Endocrinology</i> 157(6):2309-2321.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Drosophila melanogaster</i> ; CSORC wild type lab strain; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	3350270

Domain	Metric	Rating	Comments
Domain 1: Test Substance			
	Metric 1: Test Substance Identity	Low	Chemical was identified by name only
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3: Test Substance Purity	Low	Purity or grade of test substance was not reported
Domain 2: Test Design			
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes, control response was set to 100%
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization			
	Metric 7: Experimental System/Test Media Preparation	Medium	The experimental system and/or test media preparation methods were adequately reported but there is some question as to the stability of DBP in food over time
	Metric 8: Consistency of Exposure Administration	Medium	Some details of exposure administration were reported and exposures seemed to be administered consistently across study groups
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	N/A	Only one concentration was tested
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via diet
Domain 4: Test Organism			
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14: Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15: Number of Organisms and Replicates per Group	Medium	10 replicates is adequate
Domain 5: Outcome Assessment			

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Study Citation:	Williams, M. J., Wiemerslage, L., Gohel, P., Kheder, S., Kothegala, L. V., Schioth, H. B. (2016). Dibutyl Phthalate Exposure Disrupts Evolutionarily Conserved Insulin and Glucagon-Like Signaling in Drosophila Males. Endocrinology 157(6):2309-2321.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Drosophila melanogaster</i> ; CSORC wild type lab strain; Embryo			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3350270			
Domain	Metric	Rating	Comments	
	Metric 16:	Adequacy of Test Conditions	Medium	Environmental conditions of test system were most likely conducive to maintenance of organism health but actual measured condition values for control and exposed vessels was not reported
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained
Additional Comments:	The outcome was expression of metabolic and xenobiotic-related genes.			
Overall Quality Determination		Medium		

Study Citation:	Williams, M. J., Wiemerslage, L., Gohel, P., Kheder, S., Kothegala, L. V., Schioth, H. B. (2016). Dibutyl Phthalate Exposure Disrupts Evolutionarily Conserved Insulin and Glucagon-Like Signaling in Drosophila Males. Endocrinology 157(6):2309-2321.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Drosophila melanogaster</i> ; CSORC wild type lab strain; Embryo			
Health Outcome:	Behavioral			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3350270			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	Chemical was identified by name only	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Low	Purity or grade of test substance was not reported	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The experimental system and/or test media preparation methods were adequately reported but there is some question as to the stability of DBP in food over time	
	Metric 8: Consistency of Exposure Administration	Medium	Some details of exposure administration were reported and exposures seemed to be administered consistently across study groups	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Medium	Three concentrations and a control were sufficient to elicit a response	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via diet	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source	
	Metric 14: Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms	
	Metric 15: Number of Organisms and Replicates per Group	Medium	20 flies replicated 10 times is adequate	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Medium	Environmental conditions of test system were most likely conducive to maintenance of organism health but actual measured condition values for control and exposed vessels was not reported	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest	
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Study Citation:	Williams, M. J., Wiemerslage, L., Gohel, P., Kheder, S., Kothegala, L. V., Schioth, H. B. (2016). Dibutyl Phthalate Exposure Disrupts Evolutionarily Conserved Insulin and Glucagon-Like Signaling in <i>Drosophila</i> Males. <i>Endocrinology</i> 157(6):2309-2321.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Drosophila melanogaster</i> ; CSORC wild type lab strain; Embryo
Health Outcome:	Behavioral
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	3350270

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23: Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained

Additional Comments: locomotor, sleep, Fig 4, feeding behavior

Overall Quality Determination**Medium**

Study Citation:	Williams, M. J., Wiemerslage, L., Gohel, P., Kheder, S., Kothegala, L. V., Schioth, H. B. (2016). Dibutyl Phthalate Exposure Disrupts Evolutionarily Conserved Insulin and Glucagon-Like Signaling in Drosophila Males. Endocrinology 157(6):2309-2321.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Drosophila melanogaster</i> ; CSORC wild type lab strain; Adult			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3350270			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	Chemical was identified by name only	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Low	Purity or grade of test substance was not reported	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes, control response was set to 100%	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The experimental system and/or test media preparation methods were adequately reported but there is some question as to the stability of DBP in food over time	
	Metric 8: Consistency of Exposure Administration	Medium	Some details of exposure administration were reported and exposures seemed to be administered consistently across study groups	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	N/A	Only one concentration was tested	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via diet	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source	
	Metric 14: Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms	
	Metric 15: Number of Organisms and Replicates per Group	Medium	10 replicates is adequate	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Medium	Environmental conditions of test system were most likely conducive to maintenance of organism health but actual measured condition values for control and exposed vessels was not reported	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest	
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Study Citation:	Williams, M. J., Wiemerslage, L., Gohel, P., Kheder, S., Kothegala, L. V., Schioth, H. B. (2016). Dibutyl Phthalate Exposure Disrupts Evolutionarily Conserved Insulin and Glucagon-Like Signaling in <i>Drosophila</i> Males. <i>Endocrinology</i> 157(6):2309-2321.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Drosophila melanogaster</i> ; CSORC wild type lab strain; Adult
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	3350270

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23: Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained

Additional Comments: The outcome was expression of metabolic and xenobiotic-related genes.

Overall Quality Determination

Medium

Study Citation:	Williams, M. J., Wiemerslage, L., Gohel, P., Kheder, S., Kothegala, L. V., Schioth, H. B. (2016). Dibutyl Phthalate Exposure Disrupts Evolutionarily Conserved Insulin and Glucagon-Like Signaling in Drosophila Males. Endocrinology 157(6):2309-2321.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Drosophila melanogaster</i> ; CSORC wild type lab strain; Embryo			
Health Outcome:	Nutritional & Metabolic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3350270			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Low	Purity or grade of the test substance was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The experimental system and/or test media preparation methods were adequately reported but there is some question as to the stability of DBP in food over time.	
	Metric 8: Consistency of Exposure Administration	Medium	Some details of exposure administration were reported and exposures seemed to be administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Three concentrations and a control were sufficient to elicit a response.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via diet.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	Twenty flies with ten replicates were used.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Medium	Environmental conditions of the test system were most likely conducive to maintenance of organism health but actual measured condition values for control and exposed vessels were not reported.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	
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Study Citation:	Williams, M. J., Wiemerslage, L., Gohel, P., Kheder, S., Kothegala, L. V., Schioth, H. B. (2016). Dibutyl Phthalate Exposure Disrupts Evolutionarily Conserved Insulin and Glucagon-Like Signaling in <i>Drosophila</i> Males. <i>Endocrinology</i> 157(6):2309-2321.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Drosophila melanogaster</i> ; CSORC wild type lab strain; Embryo			
Health Outcome:	Nutritional & Metabolic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3350270			
Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23:	Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.
Additional Comments: Glucose, glycogen, trehalose, lipid content				

Overall Quality Determination**Medium**

Study Citation:	Williams, M. J., Wiemerslage, L., Gohel, P., Kheder, S., Kothegala, L. V., Schioth, H. B. (2016). Dibutyl Phthalate Exposure Disrupts Evolutionarily Conserved Insulin and Glucagon-Like Signaling in <i>Drosophila</i> Males. <i>Endocrinology</i> 157(6):2309-2321.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Drosophila melanogaster</i> ; CSORC wild type lab strain; Embryo			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3350270			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	Chemical was identified by name only	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Low	Purity or grade of test substance was not reported	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes, control response was set to 100%	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The experimental system and/or test media preparation methods were adequately reported but there is some question as to the stability of DBP in food over time	
	Metric 8: Consistency of Exposure Administration	Medium	Some details of exposure administration were reported and exposures seemed to be administered consistently across study groups	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	N/A	Only one concentration was tested	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via diet	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source	
	Metric 14: Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms	
	Metric 15: Number of Organisms and Replicates per Group	Medium	10 replicates is adequate	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Medium	Environmental conditions of test system were most likely conducive to maintenance of organism health but actual measured condition values for control and exposed vessels was not reported	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest	
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Study Citation:	Williams, M. J., Wiemerslage, L., Gohel, P., Kheder, S., Kothegala, L. V., Schioth, H. B. (2016). Dibutyl Phthalate Exposure Disrupts Evolutionarily Conserved Insulin and Glucagon-Like Signaling in <i>Drosophila</i> Males. <i>Endocrinology</i> 157(6):2309-2321.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Drosophila melanogaster</i> ; CSORC wild type lab strain; Embryo
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	3350270

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23: Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained

Additional Comments: Mortality Fig 1

Overall Quality Determination**Medium**

Study Citation:	Misra, S., Singh, A., Ch, R., Sharma, V., Mudiam, R., M.K., Ram, K. R. (2014). Identification of <i>Drosophila</i> -based endpoints for the assessment and understanding of xenobiotic-mediated male reproductive adversities. <i>Toxicological Sciences</i> 141(1):278-291.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Drosophila melanogaster</i> ; Embryo			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2510760			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	Chemical was identified by name only	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Low	Purity or grade of test substance was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group	
	Metric 5: Negative Control Response	Low	The biological response of the negative control group was reported and reasonable	
	Metric 6: Randomized Allocation	Medium	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The study provided only limited details on the measures taken to appropriately prepare test concentrations although the test system was described in adequate detail. Glass vials were used.	
	Metric 8: Consistency of Exposure Administration	Medium	One time dose to the food, few other details provided	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured	
	Metric 10: Exposure Duration and Frequency	Low	It was unclear what the exact exposure duration was, it was based on eclosion time (>10 days)	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Two exposure groups, suitably spaced	
	Metric 12: Testing at or Below Solubility Limit	N/A	exposure was via diet	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The number of test organisms (100) was adequate.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate	
	Metric 17: Outcome Assessment Methodology	Medium	The outcome assessment methodology was not reported	

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Study Citation:	Misra, S., Singh, A., Ch, R., Sharma, V., Mudiam, R., M.K., Ram, K. R. (2014). Identification of <i>Drosophila</i> -based endpoints for the assessment and understanding of xenobiotic-mediated male reproductive adversities. Toxicological Sciences 141(1):278-291.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Drosophila melanogaster</i> ; Embryo
Health Outcome:	Reproductive/Teratogenic
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	2510760

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described
	Metric 22: Reporting of Data	High	Data for exposure-related findings were shown for each treatment and control group
	Metric 23: Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained.
Additional Comments: Multiple studies reported, this led to a lack of clarity regarding test set up and conditions			

Overall Quality Determination**Medium**

Study Citation:	Misra, S., Singh, A., Ch, R., Sharma, V., Mudiam, R., M.K., Ram, K. R. (2014). Identification of <i>Drosophila</i> -based endpoints for the assessment and understanding of xenobiotic-mediated male reproductive adversities. <i>Toxicological Sciences</i> 141(1):278-291.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Drosophila melanogaster</i> ; Embryo			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2510760			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Low	Purity or grade of the test substance was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group.	
	Metric 5: Negative Control Response	Low	The biological response of the negative control group was not reported and reasonable for assessed outcomes.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The study provided only limited details on the measures taken to appropriately prepare test concentrations although the test system was described in adequate detail. Glass vials were used.	
	Metric 8: Consistency of Exposure Administration	Medium	A one-time dose was used to the food, but few other details were provided.	
	Metric 9: Measurement of Test Substance Concentration	Medium	Exposure concentrations were not measured, but body burdens were.	
	Metric 10: Exposure Duration and Frequency	Low	The exact duration was not clear. Eggs were exposed until eclosion, and the text did state that flies were three days old when assessed. The total duration was reported as 12-13 days.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	N/A	Only one level was tested.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via diet.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.	
	Metric 15: Number of Organisms and Replicates per Group	Low	The number of test organisms or replicates was not reported.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed the intended outcome of interest.	

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Study Citation:	Misra, S., Singh, A., Ch, R., Sharma, V., Mudiam, R., M.K., Ram, K. R. (2014). Identification of <i>Drosophila</i> -based endpoints for the assessment and understanding of xenobiotic-mediated male reproductive adversities. <i>Toxicological Sciences</i> 141(1):278-291.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Drosophila melanogaster</i> ; Embryo
Health Outcome:	ADME (biotransformation)
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	2510760

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	N/A	Statistical analysis is not typical for this type of outcome.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.

Additional Comments: None

Overall Quality Determination**Medium**

Study Citation:	Misra, S., Singh, A., Ch, R., Sharma, V., Mudiam, R., M.K., Ram, K. R. (2014). Identification of <i>Drosophila</i> -based endpoints for the assessment and understanding of xenobiotic-mediated male reproductive adversities. <i>Toxicological Sciences</i> 141(1):278-291.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Drosophila melanogaster</i> ; Adult			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2510760			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	Chemical was identified by name only	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Low	Purity or grade of test substance was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group	
	Metric 5: Negative Control Response	Low	The biological response of the negative control group was reported and reasonable	
	Metric 6: Randomized Allocation	Medium	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The study provided only limited details on the measures taken to appropriately prepare test concentrations although the test system was described in adequate detail. Glass vials were used.	
	Metric 8: Consistency of Exposure Administration	Medium	One time dose to the food, few other details provided	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured	
	Metric 10: Exposure Duration and Frequency	Low	It was unclear what the exact exposure duration was, it was based on eclosion time (>10 days)	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Two exposure groups, suitably spaced	
	Metric 12: Testing at or Below Solubility Limit	N/A	exposure was via diet	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The number of test organisms (100) was adequate.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate	
	Metric 17: Outcome Assessment Methodology	Medium	The outcome assessment methodology was not reported	
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported	

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Study Citation:	Misra, S., Singh, A., Ch, R., Sharma, V., Mudiam, R., M.K., Ram, K. R. (2014). Identification of <i>Drosophila</i> -based endpoints for the assessment and understanding of xenobiotic-mediated male reproductive adversities. <i>Toxicological Sciences</i> 141(1):278-291.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Drosophila melanogaster</i> ; Adult
Health Outcome:	Reproductive/Teratogenic
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	2510760

Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described
	Metric 22: Reporting of Data	High	Data for exposure-related findings were shown for each treatment and control group
	Metric 23: Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained.
Additional Comments: Multiple studies reported, this led to a lack of clarity regarding test set up and conditions			

Overall Quality Determination**Medium**

Study Citation:	Misra, S., Singh, A., Ch, R., Sharma, V., Mudiam, R., M.K., Ram, K. R. (2014). Identification of <i>Drosophila</i> -based endpoints for the assessment and understanding of xenobiotic-mediated male reproductive adversities. <i>Toxicological Sciences</i> 141(1):278-291.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Drosophila melanogaster</i> ; Embryo			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2510760			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	Chemical was identified by name only	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Low	Purity or grade of test substance was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group	
	Metric 5: Negative Control Response	Low	The biological response of the negative control group was reported and reasonable	
	Metric 6: Randomized Allocation	Medium	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The study provided only limited details on the measures taken to appropriately prepare test concentrations although the test system was described in adequate detail. Glass vials were used.	
	Metric 8: Consistency of Exposure Administration	Medium	One time dose to the food, few other details provided	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured	
	Metric 10: Exposure Duration and Frequency	Low	It was unclear what the exact exposure duration was, it was based on eclosion time (>10 days)	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Two exposure groups, suitably spaced	
	Metric 12: Testing at or Below Solubility Limit	N/A	exposure was via diet	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The number of test organisms (100) was adequate.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate	
	Metric 17: Outcome Assessment Methodology	Medium	The outcome assessment methodology was not reported	
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported	

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Study Citation:	Misra, S., Singh, A., Ch, R., Sharma, V., Mudiam, R., M.K., Ram, K. R. (2014). Identification of <i>Drosophila</i> -based endpoints for the assessment and understanding of xenobiotic-mediated male reproductive adversities. <i>Toxicological Sciences</i> 141(1):278-291.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Drosophila melanogaster</i> ; Embryo
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	2510760

Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described
	Metric 22: Reporting of Data	High	Data for exposure-related findings were shown for each treatment and control group
	Metric 23: Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained.
Additional Comments: Multiple studies reported, this led to a lack of clarity regarding test set up and conditions			

Overall Quality Determination**Medium**

Study Citation:	Misra, S., Singh, A., Ch, R., Sharma, V., Mudiam, R., M.K., Ram, K. R. (2014). Identification of <i>Drosophila</i> -based endpoints for the assessment and understanding of xenobiotic-mediated male reproductive adversities. <i>Toxicological Sciences</i> 141(1):278-291.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Drosophila melanogaster</i> ; Embryo			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2510760			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	Chemical was identified by name only	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Low	Purity or grade of test substance was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group	
	Metric 5: Negative Control Response	Low	The biological response of the negative control group was not reported	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The study provided only limited details on the measures taken to appropriately prepare test concentrations although the test system was described in adequate detail. glass vials were used.	
	Metric 8: Consistency of Exposure Administration	Medium	One time dose to the food, few other details provided	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured	
	Metric 10: Exposure Duration and Frequency	High	72 hour exposure duration	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Low	No information is provided on the number of exposure groups and spacing of exposure levels although the range was 10 mM to 2 M	
	Metric 12: Testing at or Below Solubility Limit	N/A	exposure was via diet	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized	
	Metric 15: Number of Organisms and Replicates per Group	Low	The number of test organisms was not reported.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate	
	Metric 17: Outcome Assessment Methodology	Low	The outcome assessment methodology was not reported	
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Study Citation:	Misra, S., Singh, A., Ch, R., Sharma, V., Mudiam, R., M.K., Ram, K. R. (2014). Identification of <i>Drosophila</i> -based endpoints for the assessment and understanding of xenobiotic-mediated male reproductive adversities. <i>Toxicological Sciences</i> 141(1):278-291.
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Drosophila melanogaster</i> ; Embryo
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	2510760

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described
	Metric 22: Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group, but results were described in the text.
	Metric 23: Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability

Additional Comments: None

Overall Quality Determination**Low**

Study Citation:	Misra, S., Singh, A., Ch, R., Sharma, V., Mudiam, R., M.K., Ram, K. R. (2014). Identification of <i>Drosophila</i> -based endpoints for the assessment and understanding of xenobiotic-mediated male reproductive adversities. <i>Toxicological Sciences</i> 141(1):278-291.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Drosophila melanogaster</i> ; Adult			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2510760			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Low	Purity or grade of the test substance were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group.	
	Metric 5: Negative Control Response	Low	The biological response of the negative control group was reported and reasonable.	
	Metric 6: Randomized Allocation	Medium	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The study provided only limited details on the measures taken to appropriately prepare test concentrations although the test system was described in adequate detail. Glass vials were used.	
	Metric 8: Consistency of Exposure Administration	Medium	A one-time dose was used to the food, but few other details were provided.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	Low	It was unclear what the exact exposure duration was, but it was based on eclosion time (>10 days).	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Medium	There were two exposure groups that were evenly spaced.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via diet.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The number of test organisms (100) was adequate.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.	
	Metric 17: Outcome Assessment Methodology	Medium	The outcome assessment methodology was not reported.	
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported.	

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Study Citation:	Misra, S., Singh, A., Ch, R., Sharma, V., Mudiam, R., M.K., Ram, K. R. (2014). Identification of <i>Drosophila</i> -based endpoints for the assessment and understanding of xenobiotic-mediated male reproductive adversities. <i>Toxicological Sciences</i> 141(1):278-291.
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Drosophila melanogaster</i> ; Adult
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Reproductive/Teratogenic
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	2510760

Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were shown for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.

Additional Comments: Multiple studies were reported, which led to a lack of clarity regarding test set up and conditions.

Overall Quality Determination

Medium

Study Citation:	Du, L., Li, G., Liu, M., Li, Y., Yin, S., Zhao, J. (2015). Biomarker responses in earthworms (<i>Eisenia fetida</i>) to soils contaminated with di-n-butyl phthalates. Environmental Science and Pollution Research 22(6):4660-4669.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Sediment; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); <i>Eisenia fetida</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2816887			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The test substance was identified by name only.	
	Metric 2: Test Substance Source	Low	The source of the DBP was reported to be Shandong Jingbo Agricultural Chemical Co. Ltd. It was not reported if it was analytically verified.	
	Metric 3: Test Substance Purity	High	The purity of the DBP was reported to be 98%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	Low	A control group is mentioned, but results are not shown and it is not reported whether it is a true negative control or a vehicle/solvent control.	
	Metric 5: Negative Control Response	Low	The negative control response was not reported.	
	Metric 6: Randomized Allocation	Low	It was not reported how the worms were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	Limited details were provided on the preparation of the test media. The filter paper test was used for this portion of the study. Glass culture dishes were used and washed in chromic acid, and dried at 300C overnight.	
	Metric 8: Consistency of Exposure Administration	Low	All exposures were for 48h at 20C with a 12L:12D photoperiod, but little other information was provided to determine consistency.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if the DBP was measured at any point in the study.	
	Metric 10: Exposure Duration and Frequency	High	The exposure duration was 48h, which is typical of an acute toxicity test.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	There were seven exposure levels and spacing appeared adequate for a response.	
	Metric 12: Testing at or Below Solubility Limit	N/A	The exposure was via "soil," which was filter paper in this case.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The worms were from the Shandong Agricultural University and were reported to be adults.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	A 24h depuration period was provided in the same conditions as the test (Petri dish lined with filter paper at 20C and 12/12h photoperiod).	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were ten organisms per test chamber and there were three replicates per treatment level.	
Domain 5: Outcome Assessment				

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Study Citation:	Du, L., Li, G., Liu, M., Li, Y., Yin, S., Zhao, J. (2015). Biomarker responses in earthworms (<i>Eisenia fetida</i>) to soils contaminated with di-n-butyl phthalates. Environmental Science and Pollution Research 22(6):4660-4669.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Sediment; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); <i>Eisenia fetida</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2816887			
Domain	Metric	Rating	Comments	
	Metric 16: Adequacy of Test Conditions	Medium	Organisms were reported to be kept at 20C with a 12L:12D photoperiod. It was reported cow dung was used as a substrate and as the food source, but how much was used was not reported. Worms were allowed to empty their guts prior to the start of the test.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest–LC50 values.	
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. Worms were assessed for mortalities every 24h.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical methods were reported in the "Statistical Analysis" section of the paper.	
	Metric 22: Reporting of Data	Low	Only LC50 values were reported in the text. No mortality or control data was reported.	
	Metric 23: Explanation of Unexpected Outcomes	High	The study authors did not report any unexpected outcomes. 95% confidence intervals were reported in the text.	
Additional Comments:	This portion of the evaluation was on the 24 and 48h acute toxicity of DBP on <i>E. fetida</i> using the filter paper test. Mortality was selected as the outcome of interest.			

Overall Quality Determination**Medium**

Study Citation:	Neuhauser, E. F., Loehr, R. C., Malecki, M. R., Milligan, D. L., Durkin, P. R. (1985). The toxicity of selected organic chemicals to the earthworm <i>Eisenia fetida</i> . Journal of Environmental Quality 14(3):383-388.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Eisenia fetida</i> ; Savigny; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3625226			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Test substance was listed by name and CAS number.
	Metric 2:	Test Substance Source	High	"The chemicals were purchased from the Aldrich Chemical Co., Milwaukee, WI, Eastman Kodak Co., Rochester, NY, and Fisher Scientific Co., Fairlawn, NJ."
	Metric 3:	Test Substance Purity	High	A minimum purity of 98% was stated.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Controls were utilized.
	Metric 5:	Negative Control Response	Low	Negative control response was not reported.
	Metric 6:	Randomized Allocation	Low	Random allocation was not stated.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	It is unclear whether DBP was dissolved in a solvent prior to addition to filter paper.
	Metric 8:	Consistency of Exposure Administration	Medium	It is unclear if solvent was utilized and if same amount of solvent was used across the different treatment groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	This study utilized a 48-hr contact exposure duration to establish an LC50 value.
	Metric 11:	Number of Exposure Groups/Spacing of Exposure Levels	Low	Exposure concentrations were not stated. At least five concentrations were utilized for the definitive test (exact number of exposure groups not specified); unclear whether one of these concentrations is a control (blank).
	Metric 12:	Testing at or Below Solubility Limit	Low	Test substance was applied to filter paper using water or other solvent. Unclear which was used and if concentrations exceeded solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	Source of the test organisms not stated.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	An acclimation process/procedure was not reported.
	Metric 15:	Number of Organisms and Replicates per Group	Low	There were ten replicate worms per test concentration (one worm per vial).
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	It was unclear whether worms had adequate air flow during the exposure period.

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Study Citation:	Neuhauser, E. F., Loehr, R. C., Malecki, M. R., Milligan, D. L., Durkin, P. R. (1985). The toxicity of selected organic chemicals to the earthworm <i>Eisenia fetida</i> . Journal of Environmental Quality 14(3):383-388.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Eisenia fetida</i> ; Savigny; Adult
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	3625226

Domain	Metric	Rating	Comments
	Metric 17: Outcome Assessment Methodology	High	Mortality was assessed using a "gentle mechanical touch".
	Metric 18: Consistency of Outcome Assessment	High	Mortality was assessed in study groups after the 48 hour exposure duration.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	Attrition in each treatment group was not reported; however there was no information to suggest differences in attrition among the study groups unrelated to exposure.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	"The LC50 value for each chemical tested was calculated using the method of Litchfield and Wilcoxon (1949)."
	Metric 22: Reporting of Data	Low	The LC50 value with CI was reported in Table 1, but data for each individual treatment group was not shown.
	Metric 23: Explanation of Unexpected Outcomes	High	Confidence intervals were reported.

Additional Comments: This form is for the contact test. DEHP and DBP were not selected for the artificial soil test.

Overall Quality Determination

Medium

Study Citation:	Du, L., Li, G., Liu, M., Li, Y., Yin, S., Zhao, J. (2015). Biomarker responses in earthworms (<i>Eisenia fetida</i>) to soils contaminated with di-n-butyl phthalates. Environmental Science and Pollution Research 22(6):4660-4669.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Sediment; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); <i>Eisenia fetida</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2816887			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The test substance was identified by name only.
	Metric 2:	Test Substance Source	Low	The source of the DBP was reported to be Shandong Jingbo Agricultural Chemical Co. Ltd. It was not reported if it was analytically verified.
	Metric 3:	Test Substance Purity	High	The purity of the DBP was reported to be 98%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported that a concurrent negative control was used for this part of the study.
	Metric 5:	Negative Control Response	Low	The negative control response was not reported. Only LC50 values were reported.
	Metric 6:	Randomized Allocation	Medium	The worms were reported to be randomly allocated to treatment groups for this portion of the study.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	Limited details were provided on the preparation of the test media for each concentration. Artificial soil was used as the substrate/test media for this test. Glassware was used and washed in chromic acid, then dried at 300C overnight.
	Metric 8:	Consistency of Exposure Administration	Low	All exposures took place in 1000mL containers with 500g of treated soil with 10 worms each. Experiments were conducted at 20C with a 12L:12D photoperiod. All containers were sealed with plastic film.
	Metric 9:	Measurement of Test Substance Concentration	Low	It was not reported if the DBP was measured at any point in the study.
	Metric 10:	Exposure Duration and Frequency	Medium	The exposure duration was reported to be 14d. This is longer than is typical for an acute toxicity test, but it was effective to obtain results pertinent to the outcome of interest.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	There were five exposure levels and spacing appeared adequate for a response.
	Metric 12:	Testing at or Below Solubility Limit	N/A	The exposure was via artificial soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The worms were from the Shandong Agricultural University and were reported to be adults.
	Metric 14:	Acclimatization and Pretreatment Conditions	Medium	Worms were acclimated for 24h in similar conditions to those in the study (glass Petri dishes lined with filter paper at a 12/12h photoperiod and 20C).
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Study Citation:	Du, L., Li, G., Liu, M., Li, Y., Yin, S., Zhao, J. (2015). Biomarker responses in earthworms (<i>Eisenia fetida</i>) to soils contaminated with di-n-butyl phthalates. Environmental Science and Pollution Research 22(6):4660-4669.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Sediment; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); <i>Eisenia fetida</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2816887			
Domain	Metric	Rating	Comments	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were ten organisms per test chamber and there were three replicates per treatment level.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Medium	Organisms were reported to be kept at 20C with a 12L:12D photoperiod. It was reported cow dung was used as a substrate and as the food source, but how much was used was not reported. Worms were allowed to empty their guts prior to the start of the test. It is unclear what they were fed for the duration of this study.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest–LC50 values.	
	Metric 18: Consistency of Outcome Assessment	High	Details regarding the execution of the study protocol for outcome assessment were limited. Worms were assessed for mortality at 14d, but it is unclear if they were assessed at any other point.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical methods were reported in the “Statistical Analysis” section of the paper.	
	Metric 22: Reporting of Data	Low	Only LC50 values were reported in the text. No mortality or control data was reported.	
	Metric 23: Explanation of Unexpected Outcomes	High	The study authors did not report any unexpected outcomes. 95% confidence intervals were reported in the text.	
Additional Comments:	This portion of the evaluation was on 14d acute toxicity of DBP on <i>E. fetida</i> using artificial soil. Mortality was selected as the outcome of interest.			

Overall Quality Determination**Medium**

Study Citation:	Du, L., Li, G., Liu, M., Li, Y., Yin, S., Zhao, J. (2015). Biomarker responses in earthworms (<i>Eisenia fetida</i>) to soils contaminated with di-n-butyl phthalates. Environmental Science and Pollution Research 22(6):4660-4669.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); <i>Eisenia fetida</i> ; Adult			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Oxidative stress (including redox biology)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2816887			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The test substance was identified by name only.	
	Metric 2: Test Substance Source	Low	The source of the DBP was reported to be Shandong Jingbo Agricultural Chemical Co. Ltd. It was not reported if it was analytically verified.	
	Metric 3: Test Substance Purity	High	The purity of the DBP was reported to be 98%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported that a concurrent negative control was used for this part of the study.	
	Metric 5: Negative Control Response	High	The negative control responses were reported in Figures 1-6 and were adequate for the outcomes of interest.	
	Metric 6: Randomized Allocation	Medium	The worms were reported to be randomly allocated to treatment groups for this portion of the study.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	Limited details were provided on the preparation of the test media for each concentration. Artificial soil was used as the substrate/test media for this test. Glassware was washed in chromic acid and dried at 300C overnight.	
	Metric 8: Consistency of Exposure Administration	Low	All exposures took place in 1000mL containers with 500g of treated soil with 10 worms each. Experiments were conducted at 20C with a 12L:12D photoperiod. All containers were sealed with plastic film.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if the DBP was measured at any point in the study.	
	Metric 10: Exposure Duration and Frequency	High	The exposure duration was reported to be 28d. This was adequate for the outcomes of interest.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	There were five exposure levels and spacing appeared adequate for a response.	
	Metric 12: Testing at or Below Solubility Limit	N/A	The exposure was via artificial soil.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The worms were from the Shandong Agricultural University and were reported to be adults.	
	Metric 14: Acclimatization and Pretreatment Conditions	Medium	Acclimation was for 24h in glass Petri dishes at 20C with a 12/12h photoperiod, which was similar to the study conditions.	

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Study Citation:	Du, L., Li, G., Liu, M., Li, Y., Yin, S., Zhao, J. (2015). Biomarker responses in earthworms (<i>Eisenia fetida</i>) to soils contaminated with di-n-butyl phthalates. Environmental Science and Pollution Research 22(6):4660-4669.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); <i>Eisenia fetida</i> ; Adult			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Oxidative stress (including redox biology)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2816887			
Domain	Metric	Rating	Comments	
	Metric 15:	Number of Organisms and Replicates per Group	Medium	There were ten organisms per test chamber and there were three replicates per treatment level.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Organisms were reported to be kept at 20C with a 12L:12D photoperiod. It was reported cow dung was used as a substrate and as the food source, but how much was used was not reported. Worms were allowed to empty their guts prior to the start of the test. It is unclear what they were fed for the duration of this study.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest–SOD, CAT, POD, GST, GSH, MDA.
	Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. Organisms were sampled at days 7, 14, 21, and 28 and assessed for SOD, CAT, POD, GST, GSH, and MDA.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. It was not reported if the worms were acclimated.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were reported in the "Statistical Analysis" section of the paper.
	Metric 22:	Reporting of Data	High	Exposure and control responses were reported in Figures 1-6. They were adequate for the outcomes of interest.
	Metric 23:	Explanation of Unexpected Outcomes	High	The study authors did not report any unexpected outcomes. Variability was reported in the figures.
Additional Comments:	This portion of the evaluation was on the effect of DBP on SOD, CAT, POD, GSH, GST, and MDA in <i>E. fetida</i> . Mechanistic outcomes of biomarkers and oxidative stress were chosen as the outcomes of interest.			
Overall Quality Determination		Medium		

Study Citation:	Frances, S. P. (1994). Response of a chigger, eutrombicula-hirsti (acari, trombiculidae) to repellent and toxicant compounds in the laboratory. Journal of Medical Entomology 31(4):628-630.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Eutrombicula hirsti</i> ; Larvae			
Health Outcome:	Behavioral			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1341925			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The DBP was identified by name only.	
	Metric 2: Test Substance Source	Low	The source of the DBP was reported at Colbar in Melbourne, Australia. It was not reported if it was analytically verified.	
	Metric 3: Test Substance Purity	High	The purity of the DBP was reported to be 100%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of a concurrent negative control in which ethanol was used.	
	Metric 5: Negative Control Response	Low	The biological response of the negative control was not reported.	
	Metric 6: Randomized Allocation	Low	It was not reported how the chiggers were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	Minimal details were provided on the preparation of the test solutions. Serial dilutions were reported to be used, but little other information was provided.	
	Metric 8: Consistency of Exposure Administration	Medium	Exposures were administered via 10 x 10cm cotton fabric. The test substance (0.3mL) was added and allowed to dry for 20h before 10-20 chigger larvae were placed in the test system. Chiggers were checked every three minutes until the mite could no longer move the length of its body. The duration was not reported.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if the test substance was measured at any point in the study.	
	Metric 10: Exposure Duration and Frequency	Uninformative	The exposure duration was not reported. It was reported that the organisms were observed every three minutes, but it was unclear how long this continued for.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Uninformative	The number and the spacing of the exposure groups was not reported. Only endpoint values were reported.	
	Metric 12: Testing at or Below Solubility Limit	High	Study authors reported the use of an appropriate solvent control.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	Organisms were obtained from a culture colony originally obtained from larvae collected at Cowley Beach near Innisfail, northern Queensland, Australia.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the organisms were acclimated in any way.	
	Metric 15: Number of Organisms and Replicates per Group	Low	There were 10-20 chiggers tested at once. Tests were repeated three times at each concentration.	

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Study Citation:	Frances, S. P. (1994). Response of a chigger, eutrombicula-hirsti (acari, trombiculidae) to repellent and toxicant compounds in the laboratory. Journal of Medical Entomology 31(4):628-630.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Eutrombicula hirsti</i> ; Larvae			
Health Outcome:	Behavioral			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1341925			
Domain	Metric	Rating	Comments	
Domain 5: Outcome Assessment				
Metric 16:	Adequacy of Test Conditions	Medium	Feeding regimen was reported for the larvae, and they were kept at 27C in 75-80% relative humidity.	
Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—behavior changes due to DBP exposure reported as ET50 and ET95 in terms of time to knock out.	
Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. Organisms were monitored for behavioral changes every three minutes until they no longer can move the length of their body.	
Domain 6: Confounding / Variable Control				
Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. It was not reported if any acclimation occurred.	
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
Metric 21:	Statistical Methods	High	Probit analysis was reported to be used to determine ET50 and ET95 values.	
Metric 22:	Reporting of Data	Low	Only ET50 and ET95 values were reported in Table 2. Control response and any raw data were not reported.	
Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in Table 2.	
Additional Comments:	This study was on the effect of DBP on repellent behavior in chigger E. hirsti. Organisms were observed after being placed in the test chamber with the 10 x 10cm cotton cloth. The study received an unacceptable rank due to the lack of information regarding exposure concentrations and duration.			

Overall Quality Determination**Uninformative**

Study Citation:	Frances, S. P. (1994). Response of a chigger, eutrombicula-hirsti (acari, trombiculidae) to repellent and toxicant compounds in the laboratory. Journal of Medical Entomology 31(4):628-630.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Eutrombicula hirsti</i> ; Larvae			
Health Outcome:	Behavioral			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1341925			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The DBP was identified by name only.
	Metric 2:	Test Substance Source	Low	The source of the DBP was reported at Colbar in Melbourne, Australia. It was not reported if it was analytically verified.
	Metric 3:	Test Substance Purity	High	
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of a concurrent negative control in which ethanol was used.
	Metric 5:	Negative Control Response	Low	The biological response of the negative control was not reported.
	Metric 6:	Randomized Allocation	Low	It was not reported how the chiggers were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	Minimal details were provided on the preparation of the test solutions. Serial dilutions were reported to be used, but little other information was provided.
	Metric 8:	Consistency of Exposure Administration	Medium	Exposures were administered via filter paper. The test substance (0.5mL) was added and allowed to dry before five chigger larvae were placed in the test system. Chiggers were monitored for 10 minutes for changes in behavior due to the repellent.
	Metric 9:	Measurement of Test Substance Concentration	Low	It was not reported if the test substance was measured at any point in the study.
	Metric 10:	Exposure Duration and Frequency	Medium	The exposure duration was reported to be 10 minutes, and this was adequate to see a response.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Uninformative	The number and the spacing of the exposure groups was not reported. Only endpoint values were reported.
	Metric 12:	Testing at or Below Solubility Limit	High	Study authors reported the use of an appropriate solvent control.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Organisms were obtained from a culture colony originally obtained from larvae collected at Cowley Beach near Innisfail, northern Queensland, Australia.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the organisms were acclimated in any way.
	Metric 15:	Number of Organisms and Replicates per Group	Low	There were five chiggers tested at once. Tests were repeated at least seven times at each concentration.
Domain 5: Outcome Assessment				
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Study Citation:	Frances, S. P. (1994). Response of a chigger, eutrombicula-hirsti (acari, trombiculidae) to repellent and toxicant compounds in the laboratory. Journal of Medical Entomology 31(4):628-630.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Eutrombicula hirsti</i> ; Larvae			
Health Outcome:	Behavioral			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1341925			
Domain	Metric	Rating	Comments	
	Metric 16: Adequacy of Test Conditions	Medium	Feeding regimen was reported for the larvae, and they were kept at 27C in 75-80% relative humidity.	
	Metric 17: Outcome Assessment Methodology	Uninformative	Repellency and mortality was combined to evaluate ED50 (effective dose) values - and the study did not specify what fraction died and what fraction were repelled.	
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. Organisms were monitored for behavioral changes for 10 minutes to determine if the repellent was effective.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. It was not reported if any acclimation occurred.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Probit analysis was reported to be used to determine ED50 values.	
	Metric 22: Reporting of Data	Low	Only ED50 values were reported in Table 1. Control response and any raw data were not reported.	
	Metric 23: Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in Table 1.	
Additional Comments:	This study was on the effect of DBP on repellent behavior in chigger E. hirsti. Organisms were observed after being placed in the test chamber treated filter paper. If the organisms remained in place, the repellent was successful. If they moved off the paper, the repellent did not work. The study received an unacceptable rank due to the lack of information regarding exposure concentrations and because the outcome evaluated was unclear (mortality combined with repellency).			

Overall Quality Determination**Uninformative**

Study Citation:	Jensen, J., Langevelde, van, J., Pritzl, G., Krogh, P. H. (2001). Effects of di(2-ethylhexyl) phthalate and dibutyl phthalate on the collembolan <i>Folsomia fimetaria</i> . <i>Environmental Toxicology and Chemistry</i> 20(5):1085-1091.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Folsomia fimetaria</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	789786			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The test substance was identified only by nomenclature. No other information (CASRN, structure etc.) was provided.	
	Metric 2: Test Substance Source	Low	"The chemicals used (DBP, DEHP, and acetone) were obtained from Merck Schuchardt, Darmstadt, Germany." The test substance identity was NOT analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Low	Purity and/or grade of the test substance were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	A negative control was used (acetone-water).	
	Metric 5: Negative Control Response	Low	The biological response of the negative control groups was not reported.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	Experiments were conducted in small multidish chamber vessels with spiked soil. The exposure duration for this experiment was one week, and the experimental results for the degradation test (Figure 1) show that degradation after one week was greater than 20%. This may have an impact on results, though not as great as the impact for the longer experiments reported.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	Low	DBP concentrations were not measured throughout the study, and may be expected to degrade in soil. This may have a substantial impact on results.	
	Metric 10: Exposure Duration and Frequency	High	Exposure duration and frequency were acceptable (preparation of spiked soil at beginning of experiment).	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	"Test concentrations were DBP at 0, 1, 5, 10, and 25 mg/kg dry weight."	
	Metric 12: Testing at or Below Solubility Limit	N/A	This was a spiked soil exposure.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	"Ten juveniles (0–1 d old) were added to each of two replicated microcosms."	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	There was no reported acclimatization, but there was no evidence to suggest results were impacted.	
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Study Citation:	Jensen, J., Langevelde, van, J., Pritzl, G., Krogh, P. H. (2001). Effects of di(2-ethylhexyl) phthalate and dibutyl phthalate on the collembolan <i>Folsomia fimetaria</i> . Environmental Toxicology and Chemistry 20(5):1085-1091.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Folsomia fimetaria</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	789786			
Domain	Metric	Rating	Comments	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 10 juveniles per concentration with two replications.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Reporting of housing and/or environmental conditions and/or food, water, and nutrients and/or biomass loading was not sufficiently reported to evaluate if adequate and whether differences occurred between control and exposed populations.	
	Metric 17: Outcome Assessment Methodology	High	Juveniles were assessed for survival at the end of the one-week exposure.	
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Details of statistical methods were reported in the methods and are appropriate for the study.	
	Metric 22: Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group, but results were described in the text. "Juvenile mortality was similar when exposed in multidisheswith a hard soil surface and in microcosms with loose soil(data not shown)."	
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.	
Additional Comments:	None			
Overall Quality Determination		Medium		

Study Citation:	Jensen, J., Langevelde, van, J., Pritzl, G., Krogh, P. H. (2001). Effects of di(2-ethylhexyl) phthalate and dibutyl phthalate on the collembolan <i>Folsomia fimetaria</i> . Environmental Toxicology and Chemistry 20(5):1085-1091.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Folsomia fimetaria</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	789786			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The test substance was identified only by nomenclature. No other information (CASRN, structure etc.) was provided.	
	Metric 2: Test Substance Source	Low	"The chemicals used (DBP, DEHP, and acetone) were obtained from Merck Schuchardt, Darmstadt, Germany." The test substance identity was NOT analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Low	Purity and/or grade of the test substance were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	A negative control was used (acetone-water).	
	Metric 5: Negative Control Response	Medium	Biological responses in the control were reported but there were limitations. "In some of the tests, mortality in the control samples exceeded 20%, which lessened the certainty of the test results".	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	Experiments were conducted in microcosm vessels with spiked soil. Because of the length of the exposure (22 days) loss of test substance is to be expected due to degradation, but no effort was made to replenish the phthalate in soil. Concentrations of test substance were not measured during the study.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	Low	DBP concentrations were not measured throughout the study, and may be expected to degrade in soil. This may have a substantial impact on results.	
	Metric 10: Exposure Duration and Frequency	High	Exposure duration and frequency were acceptable (preparation of spiked soil at beginning of experiment).	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	"Test concentrations used were... DBP at 0, 100, 250, 500, 750, and 1,000 mg/kg dry weight."	
	Metric 12: Testing at or Below Solubility Limit	N/A	This was a spiked soil exposure.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	Male & female adult collembolans were used.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	There was no reported acclimatization, but there was no evidence to suggest results were impacted.	

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Study Citation:	Jensen, J., Langevelde, van, J., Pritzl, G., Krogh, P. H. (2001). Effects of di(2-ethylhexyl) phthalate and dibutyl phthalate on the collembolan <i>Folsomia fimetaria</i> . Environmental Toxicology and Chemistry 20(5):1085-1091.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Folsomia fimetaria</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	789786			
Domain	Metric	Rating	Comments	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 10 male & 10 female per microcosm with four replicates per concentration.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Organism housing and conditions were acceptable. "Experiments were conducted at a constant temperature (20oC), with a 12:12 h light: dark regime. Animals were fed dried baker's yeast (15mg dry weight) at day 0 and day 14."	
	Metric 17: Outcome Assessment Methodology	High	Mortality was assessed using digital image processing.	
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups (at the end of the experiment).	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Details of statistical methods were reported in the methods and are appropriate for the study.	
	Metric 22: Reporting of Data	High	The EC10/LC10/EC50/LC50 are reported for all endpoints.	
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.	
Additional Comments:	Concentration of DBP in soil was not measured during or at the end of the experiment. Environmental conditions (moisture content, pH, etc.) were not reported. Control mortality exceeded 20% (Fig 2).			

Overall Quality Determination**Medium**

Study Citation:	Jensen, J., Langevelde, van, J., Pritzl, G., Krogh, P. H. (2001). Effects of di(2-ethylhexyl) phthalate and dibutyl phthalate on the collembolan <i>Folsomia fimetaria</i> . Environmental Toxicology and Chemistry 20(5):1085-1091.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Folsomia fimetaria</i> ; Adult			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	789786			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The test substance was identified only by nomenclature. No other information (CASRN, structure etc.) was provided.	
	Metric 2: Test Substance Source	Low	"The chemicals used (DBP, DEHP, and acetone) were obtained from Merck Schuchardt, Darmstadt, Germany." The test substance identity was NOT analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Low	Purity and/or grade of the test substance were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	A negative control was used (acetone-water).	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was adequate.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	Experiments were conducted in microcosm vessels with spiked soil. Because of the length of the exposure (22 days) loss of test substance is to be expected due to degradation, but no effort was made to replenish the phthalate in soil. Concentrations of test substance were not measured during the study.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	Low	DBP concentrations were not measured throughout the study, and may be expected to degrade in soil. This may have a substantial impact on results.	
	Metric 10: Exposure Duration and Frequency	High	Exposure duration and frequency were acceptable (preparation of spiked soil at beginning of experiment).	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	"Test concentrations used were... DBP at 0, 100, 250, 500, 750,and 1,000 mg/kg dry weight."	
	Metric 12: Testing at or Below Solubility Limit	N/A	This was a spiked soil exposure.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	Male & female adult collembolans were used.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	There was no reported acclimatization, but there was no evidence to suggest results were impacted.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 10 male & 10 female per microcosm, with four replicates per concentration.	
Domain 5: Outcome Assessment				

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Study Citation:	Jensen, J., Langevelde, van, J., Pritzl, G., Krogh, P. H. (2001). Effects of di(2-ethylhexyl) phthalate and dibutyl phthalate on the collembolan <i>Folsomia fimetaria</i> . Environmental Toxicology and Chemistry 20(5):1085-1091.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Folsomia fimetaria</i> ; Adult			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	789786			
Domain	Metric	Rating	Comments	
	Metric 16: Adequacy of Test Conditions	High	Organism housing and conditions were acceptable. "Experiments were conducted at constant temperature (20°C), with a 12:12 h light: dark regime. Animals were fed dried baker's yeast (15 mg dry weight) at day 0 and day 14".	
	Metric 17: Outcome Assessment Methodology	Medium	Reproduction was assessed by counting surviving juveniles at the end of study using digital image processing. However, juvenile mortality during the test period was not assessed, and was only counted at the end of the experiment.	
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups (at the end of the experiment).	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Details of statistical methods were reported in the methods and are appropriate for the study.	
	Metric 22: Reporting of Data	High	Adult reproduction data is given in Fig 2. The EC10/LC10/EC50/LC50 are reported for all endpoints (Table 1).	
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.	
Additional Comments:	None			
Overall Quality Determination		Medium		

Study Citation:	Jensen, J., Langevelde, van, J., Pritzl, G., Krogh, P. H. (2001). Effects of di(2-ethylhexyl) phthalate and dibutyl phthalate on the collembolan <i>Folsomia fimetaria</i> . Environmental Toxicology and Chemistry 20(5):1085-1091.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Folsomia fimetaria</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	789786			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	Test substance identified only by nomenclature. No other information (CASRN, structure etc.) was provided.	
	Metric 2: Test Substance Source	Low	"The chemicals used (DBP, DEHP, and acetone) were obtained from Merck Schuchardt, Darmstadt, Germany."The test substance identity was NOT analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Low	Purity and/or grade of test substance were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	A negative control was used (acetone-water).	
	Metric 5: Negative Control Response	Low	The biological response of the negative control groups was not reported.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	Experiments were conducted in small multidish chamber vessels with spiked soil. Because of the length of the exposure (60 days) loss of test substance is to be expected due to degradation, but no effort was made to replenish the phthalate in soil. Concentrations of test substance were not measured during the study.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	Low	DBP concentrations were not measured throughout the study, and may be expected to degrade in soil. This may have a substantial impact on results.	
	Metric 10: Exposure Duration and Frequency	High	Exposure duration and frequency were acceptable (preparation of spiked soil at beginning of experiment).	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	"Test concentrations were DBP at 0, 1, 5, 10, and 25 mg/kg dry weight"	
	Metric 12: Testing at or Below Solubility Limit	N/A	Spiked soil exposure.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	A single 0 to 1-d old juvenile springtail was added to each chamber.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	No reported acclimatization, but no evidence to suggest results impacted.	
	Metric 15: Number of Organisms and Replicates per Group	Low	20 organisms per exposure concentration, no replicates.	

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Study Citation:	Jensen, J., Langevelde, van, J., Pritzl, G., Krogh, P. H. (2001). Effects of di(2-ethylhexyl) phthalate and dibutyl phthalate on the collembolan <i>Folsomia fimetaria</i> . Environmental Toxicology and Chemistry 20(5):1085-1091.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Folsomia fimetaria</i> ; Juvenile
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	789786

Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Organism housing & conditions acceptable. "Experiments were run at constant temperature (20oC), with a12:12 h light : dark regime".
	Metric 17: Outcome Assessment Methodology	Medium	Juveniles were assessed twice a week for six weeks.
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups (twice a week for six weeks).
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Details of statistical methods reported in methods & are appropriate for the study.
	Metric 22: Reporting of Data	Low	EC10/LC10/EC50/LC50 are reported for all endpoints. But percent survival data was not given for each treatment group and control.
	Metric 23: Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability (e.g., SE, SD, confidence intervals) and/or insufficient information was provided to determine if excessive variability or unexpected outcomes occurred.

Additional Comments: None

Overall Quality Determination**Medium**

Study Citation:	Jensen, J., Langevelde, van, J., Pritzl, G., Krogh, P. H. (2001). Effects of di(2-ethylhexyl) phthalate and dibutyl phthalate on the collembolan <i>Folsomia fimetaria</i> . Environmental Toxicology and Chemistry 20(5):1085-1091.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Folsomia fimetaria</i> ; Juvenile			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	789786			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	Test substance identified only by nomenclature. No other information (CASRN, structure etc.) was provided.	
	Metric 2: Test Substance Source	Low	"The chemicals used (DBP, DEHP, and acetone) were obtained from Merck Schuchardt, Darmstadt, Germany." The test substance identity was NOT analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Low	Purity and/or grade of test substance were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	A negative control was used (acetone-water).	
	Metric 5: Negative Control Response	High	Biological responses in control were normal.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	Experiments were conducted in small multidish chamber vessels with spiked soil. Because of the length of the exposure (60 days) loss of test substance is to be expected due to degradation, but no effort was made to replenish the phthalate in soil. Concentrations of test substance were not measured during the study.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	Low	DBP concentrations were not measured throughout the study, and may be expected to degrade in soil. This may have a substantial impact on results.	
	Metric 10: Exposure Duration and Frequency	High	Exposure duration and frequency were acceptable (preparation of spiked soil at beginning of experiment).	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	"Test concentrations were DBP at 0, 1, 5, 10, and 25 mg/kg dry weight"	
	Metric 12: Testing at or Below Solubility Limit	N/A	Spiked soil exposure.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	A single 0 to 1-d old juvenile springtail was added to each chamber.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	No reported acclimatization, but no evidence to suggest results impacted.	
	Metric 15: Number of Organisms and Replicates per Group	Low	20 organisms per exposure concentration, no replicates.	
Domain 5: Outcome Assessment				
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Study Citation:	Jensen, J., Langevelde, van, J., Pritzl, G., Krogh, P. H. (2001). Effects of di(2-ethylhexyl) phthalate and dibutyl phthalate on the collembolan <i>Folsomia fimetaria</i> . Environmental Toxicology and Chemistry 20(5):1085-1091.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Folsomia fimetaria</i> ; Juvenile
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	789786

Domain	Metric	Rating	Comments
	Metric 16: Adequacy of Test Conditions	High	Organism housing & conditions acceptable. "Experiments were run at constant temperature (208C), with a 12:12 h light : dark regime."
	Metric 17: Outcome Assessment Methodology	Medium	"During the first three weeks, covering an entire <i>F. fimetaria</i> life cycle, exuviae of growing juveniles were recorded every second day and removed if present." "Growth of the animals was determined manually at the screen by measuring the length from the posterior end of the abdomen to the anterior end of the head."
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Details of statistical methods reported in methods & are appropriate for the study.
	Metric 22: Reporting of Data	High	EC10/LC10/EC50/LC50 are reported for all endpoints. DBP at the concentrations tested did not affect growth of juveniles (Fig 2).
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: None

Overall Quality Determination**Medium**

Study Citation:	Lenoir, A., Touchard, A., Devers, S., Christidès, J. P., Boulay, R., Cuvillier-Hot, V. (2014). Ant cuticular response to phthalate pollution. Environmental Science and Pollution Research 21(23):13446-13451.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Dermal (topical application)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Lasius niger</i> ; Adult			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2347468			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The chemical was identified by name and CAS#.
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	The chemical was 99% pure.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent solvent control group.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was suitable.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8:	Consistency of Exposure Administration	Medium	Exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were measured after administration and presented as time 0 within Figure 1 on page 4/6.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/Spacing of Exposure Levels	N/A	Only one dose was used.
	Metric 12:	Testing at or Below Solubility Limit	N/A	The test substance was solubilized in methanol prior to topical application.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	There are minor reservations about the source of test organisms.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Minor uncertainties were identified regarding environmental conditions of the test system due to few details reported.
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Study Citation:	Lenoir, A., Touchard, A., Devers, S., Christidès, J. P., Boulay, R., Cuvillier-Hot, V. (2014). Ant cuticular response to phthalate pollution. Environmental Science and Pollution Research 21(23):13446-13451.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Dermal (topical application)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Lasius niger</i> ; Adult
Health Outcome:	ADME (biotransformation)
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	2347468

Domain	Metric	Rating	Comments
	Metric 17: Outcome Assessment Methodology	Medium	The outcome assessment methodology reported the intended outcomes of interest with some uncertainty.
	Metric 18: Consistency of Outcome Assessment	Medium	There was incomplete reporting of minor details of outcome assessment protocol execution.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There were no differences among groups, but few details were provided to confirm that.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	Medium	Minor uncertainties or limitations were identified in how the study characterized unexpected outcomes.

Additional Comments: None

Overall Quality Determination**Medium**

Study Citation:	Yang, G., Zhou, B., Zhang, X., Zhang, Z., Wu, Y., Zhang, Y., Lü, S., Zou, Q., Gao, Y., Teng, L. (2016). Effects of Tomato Root Exudates on <i>Meloidogyne incognita</i> . PLoS ONE 11(4):e0154675.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); <i>Meloidogyne incognita</i> ; Juvenile			
Health Outcome:	Behavioral			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3350275			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The chemical was identified by name only.
	Metric 2:	Test Substance Source	Low	The source was not reported.
	Metric 3:	Test Substance Purity	Low	Purity or grade of the test substance was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	Low	The biological response of the negative control group was not reported other than to say control response was set at 100%.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.
	Metric 8:	Consistency of Exposure Administration	Medium	Nothing in the study suggested that exposures were not administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	Three concentrations were used. For this experiment, a dose-response relationship was observed, though repellancy was not significantly different among any treatment. A wider range of concentrations should have been used.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Three replicates were used.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.

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Study Citation:	Yang, G., Zhou, B., Zhang, X., Zhang, Z., Wu, Y., Zhang, Y., Lü, S., Zou, Q., Gao, Y., Teng, L. (2016). Effects of Tomato Root Exudates on <i>Meloidogyne incognita</i> . PLoS ONE 11(4):e0154675.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); <i>Meloidogyne incognita</i> ; Juvenile			
Health Outcome:	Behavioral			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3350275			
Domain	Metric	Rating	Comments	
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18:	Consistency of Outcome Assessment	Medium	Outcomes were assessed consistently across study groups, but few details were provided.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described.
	Metric 22:	Reporting of Data	Low	Data for exposure-related findings were shown for each treatment but not the control group,
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments:	This is for chemotaxis. For this outcome, though only three concentrations were used and there were no statistically significant differences among them, at least a linear dose-response relationship was observed. Therefore this paper was not downgraded.			
Overall Quality Determination		Medium		

Study Citation:	Yang, G., Zhou, B., Zhang, X., Zhang, Z., Wu, Y., Zhang, Y., Lü, S., Zou, Q., Gao, Y., Teng, L. (2016). Effects of Tomato Root Exudates on <i>Meloidogyne incognita</i> . PLoS ONE 11(4):e0154675.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); <i>Meloidogyne incognita</i> ; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3350275			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The chemical was identified by name only.
	Metric 2:	Test Substance Source	Low	The source was not reported.
	Metric 3:	Test Substance Purity	Low	Purity or grade of the test substance was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	Low	The biological response of the negative control group was not reported other than to say control response was set at 100%.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.
	Metric 8:	Consistency of Exposure Administration	Medium	Nothing in the study suggested that exposures were not administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Three concentrations were tested. A dose-response effect was not observed, and in fact mortality was highest in the middle concentration, and lowest in the lower and higher concentrations.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Three replicates were used.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
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Study Citation:	Yang, G., Zhou, B., Zhang, X., Zhang, Z., Wu, Y., Zhang, Y., Lü, S., Zou, Q., Gao, Y., Teng, L. (2016). Effects of Tomato Root Exudates on <i>Meloidogyne incognita</i> . PLoS ONE 11(4):e0154675.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); <i>Meloidogyne incognita</i> ; Juvenile
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	3350275

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	Medium	Outcomes were assessed consistently across study groups, but few details were provided.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	Low	Data for exposure-related findings were shown for each treatment but not the control group.
	Metric 23: Explanation of Unexpected Outcomes	Low	The nonlinearity in responses and lack of a dose-response relationship is not discussed.

Additional Comments: This form is for mortality.

Overall Quality Determination

Low

Study Citation:	Yang, G., Zhou, B., Zhang, X., Zhang, Z., Wu, Y., Zhang, Y., Lü, S., Zou, Q., Gao, Y., Teng, L. (2016). Effects of Tomato Root Exudates on Meloidogyne incognita. PLoS ONE 11(4):e0154675.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); <i>Meloidogyne incognita</i> ; Embryo			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3350275			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The chemical was identified by name only.
	Metric 2:	Test Substance Source	Low	The source was not reported.
	Metric 3:	Test Substance Purity	Low	Purity or grade of the test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	Low	The biological response of the negative control group was not reported other than to say control response was set at 100%.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.
	Metric 8:	Consistency of Exposure Administration	Medium	Nothing in the study suggested that exposures were not administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	Three concentrations were used. A dose-response relationship was not observed. Hatch suppression was not significantly different in any treatment group. All three concentrations exhibited hatch suppression relative to control.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Three replicates were used.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.

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Study Citation:	Yang, G., Zhou, B., Zhang, X., Zhang, Z., Wu, Y., Zhang, Y., Lü, S., Zou, Q., Gao, Y., Teng, L. (2016). Effects of Tomato Root Exudates on <i>Meloidogyne incognita</i> . PLoS ONE 11(4):e0154675.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); <i>Meloidogyne incognita</i> ; Embryo			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3350275			
Domain	Metric	Rating	Comments	
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18:	Consistency of Outcome Assessment	Medium	Outcomes were assessed consistently across study groups, but few details were provided.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described.
	Metric 22:	Reporting of Data	Low	Data for exposure-related findings were shown for each treatment but not the control group.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments:	This form is for the population abundance of the 8-day test.			
Overall Quality Determination		Low		

Study Citation:	Yang, G., Zhou, B., Zhang, X., Zhang, Z., Wu, Y., Zhang, Y., Lü, S., Zou, Q., Gao, Y., Teng, L. (2016). Effects of Tomato Root Exudates on <i>Meloidogyne incognita</i> . PLoS ONE 11(4):e0154675.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); <i>Meloidogyne incognita</i> ; Juvenile			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3350275			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	Low	The source was not reported.
	Metric 3:	Test Substance Purity	Low	Purity or grade of test substance was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	Low	The biological response of the negative control groups was not reported other than to say control response was set at 100%
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Medium	Nothing in the study suggested that exposures were not administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	Three concentrations were used. A dose-response relationship was not observed - hatch suppression was not significantly different in any treatment group. All three concentrations exhibited hatch suppression relative to control.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Three replicates were used
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate

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Study Citation:	Yang, G., Zhou, B., Zhang, X., Zhang, Z., Wu, Y., Zhang, Y., Lü, S., Zou, Q., Gao, Y., Teng, L. (2016). Effects of Tomato Root Exudates on <i>Meloidogyne incognita</i> . PLoS ONE 11(4):e0154675.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); <i>Meloidogyne incognita</i> ; Juvenile
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	3350275

Domain	Metric	Rating	Comments
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18: Consistency of Outcome Assessment	Medium	outcomes were assessed consistently across study groups but few details were provided
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described
	Metric 22: Reporting of Data	Low	Data for exposure-related findings were shown for each treatment but not the control group.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes
Additional Comments:	Population abundance of 8 day test.		

Overall Quality Determination**Low**

Study Citation:	Filho, D.N., I., Vieceli, N. C., Cardoso, E. M., Lovatel, E. R., Gonzatti, C. F., Marzotto, J. A., Montezano, D. G., Specht, A. (2013). Two generations of fall armyworm (Lepidoptera: Noctuidae) contamination by di-n-butylphthalate. Journal of Toxicology and Environmental Health, Part A: Current Issues 76(16):973-977.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Spodoptera frugiperda</i> ; F1 generation; Larvae			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2219889			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The test substance was identified by name only.
	Metric 2:	Test Substance Source	Low	The source of the DBP was not reported.
	Metric 3:	Test Substance Purity	High	The purity was reported to be >99%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of a concurrent negative control.
	Metric 5:	Negative Control Response	Low	The biological response of the negative control was not reported.
	Metric 6:	Randomized Allocation	Low	It was not reported how the armyworms were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The test system used 150mL plastic cups with plastic caps. The DBP was dissolved into n-hexane at 1000mg/L. This was added to the food at 1mg/g. More information is needed on the test system.
	Metric 8:	Consistency of Exposure Administration	Low	Plastic cups were used as the test chambers, so there is concern about the consistency of the exposure due to potential leaching and sorbing of the test substance.
	Metric 9:	Measurement of Test Substance Concentration	Low	It is unclear if the DBP concentration in the food was measured at any point in the study. Analysis was done on the worms themselves.
	Metric 10:	Exposure Duration and Frequency	Uninformative	The exposure duration was reported to be until death in the adult phase. Authors also reported the use of the F1 moths to produce the F2 larvae. It is unclear how long the exposure actually was.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	There was only one exposure level, as the goal was not to observe a dose response.
	Metric 12:	Testing at or Below Solubility Limit	N/A	The exposure was via diet.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The organisms were reported to be from an in-house colony at Laboratory of Pest Control and Insect Maintenance at the University of Craxia do Sol.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the organisms were acclimated.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	There were 100 larvae placed individually into 150mL plastic cups.

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Study Citation:	Filho, D.N., I., Vieceli, N. C., Cardoso, E. M., Lovatel, E. R., Gonzatti, C. F., Marzotto, J. A., Montezano, D. G., Specht, A. (2013). Two generations of fall armyworm (Lepidoptera: Noctuidae) contamination by di-n-butylphthalate. Journal of Toxicology and Environmental Health, Part A: Current Issues 76(16):973-977.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Spodoptera frugiperda</i> ; F1 generation; Larvae			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2219889			
Domain	Metric	Rating	Comments	
Domain 5: Outcome Assessment				
Metric 16:	Adequacy of Test Conditions	Medium	Organisms were kept at 25c with a relative humidity of 70% with a 14L:10D photoperiod. Organisms were fed, as the exposure was via diet, but the feeding regimen was not reported.	
Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest-mortality.	
Metric 18:	Consistency of Outcome Assessment	Low	It was unclear when the organisms were assessed for mortalities.	
Domain 6: Confounding / Variable Control				
Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. It was unclear if the organisms were acclimated to test conditions.	
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
Metric 21:	Statistical Methods	High	The statistical methods were described in the "Statistical Analysis " section of the paper.	
Metric 22:	Reporting of Data	Low	Results were only described in the text.	
Metric 23:	Explanation of Unexpected Outcomes	Low	Variability was not reported.	
Additional Comments:	This portion of the evaluation was on the effect of DBP on armyworm mortality after exposure via diet. This is for the F1 generation. Mortality was selected as the outcome of interest. The study received an unacceptable ranking due to the lack of information on the study duration.			

Overall Quality Determination**Uninformative**

Study Citation:	Filho, D.N., I., Vieceli, N. C., Cardoso, E. M., Lovatel, E. R., Gonzatti, C. F., Marzotto, J. A., Montezano, D. G., Specht, A. (2013). Two generations of fall armyworm (Lepidoptera: Noctuidae) contamination by di-n-butylphthalate. Journal of Toxicology and Environmental Health, Part A: Current Issues 76(16):973-977.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Spodoptera frugiperda</i> ; F1 generation; Larvae			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2219889			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The test substance was identified by name only.	
	Metric 2: Test Substance Source	Low	The source of the DBP was not reported.	
	Metric 3: Test Substance Purity	High	The purity was reported to be >99%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of a concurrent negative control.	
	Metric 5: Negative Control Response	High	The negative control response was reported in Figure 1 and was adequate for the outcome of interest.	
	Metric 6: Randomized Allocation	Low	It was not reported how the armyworms were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The test system used 150mL plastic cups with plastic caps. The DBP was dissolved into n-hexane at 1000mg/L. This was added to the food at 1mg/g. More information is needed on the test system.	
	Metric 8: Consistency of Exposure Administration	Low	Plastic cups were used as the test chambers, so there is concern about the consistency of the exposure due to potential leaching and sorbing of the test substance.	
	Metric 9: Measurement of Test Substance Concentration	Low	It is unclear if the DBP concentration in the food was measured at any point in the study. Analysis was done on the worms themselves.	
	Metric 10: Exposure Duration and Frequency	Uninformative	The exposure duration was reported to be until death in the adult phase. Authors also reported the use of the F1 moths to produce the F2 larvae. It is unclear how long the exposure actually was.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	N/A	There was only one exposure level, as the goal was not to observe a dose response.	
	Metric 12: Testing at or Below Solubility Limit	N/A	The exposure was via diet.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The organisms were reported to be from an in-house colony at Laboratory of Pest Control and Insect Maintenance at the University of Craxia do Sol.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the organisms were acclimated.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 100 larvae placed individually into 150mL plastic cups.	
Domain 5: Outcome Assessment				

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Study Citation:	Filho, D.N., I., Vieceli, N. C., Cardoso, E. M., Lovatel, E. R., Gonzatti, C. F., Marzotto, J. A., Montezano, D. G., Specht, A. (2013). Two generations of fall armyworm (Lepidoptera: Noctuidae) contamination by di-n-butylphthalate. Journal of Toxicology and Environmental Health, Part A: Current Issues 76(16):973-977.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Spodoptera frugiperda</i> ; F1 generation; Larvae			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2219889			
Domain		Metric	Rating	Comments
	Metric 16:	Adequacy of Test Conditions	Medium	Organisms were kept at 25c with a relative humidity of 70% with a 14L:10D photoperiod. Organism were fed, as the exposure was via diet, but the feeding regimen was not reported.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest–accumulation of DBP in armyworm tissue.
	Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. Male and female pupa were ground and an extraction was done. GC was used to analyzed DBP levels.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. It was unclear if the organisms were acclimated to test conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	The statistical methods were described in the ”Statistical Analysis ” section of the paper.
	Metric 22:	Reporting of Data	High	Data for the exposure and control responses was presented in Figure 1 and was adequate for the outcome of interest.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was presented in figure 1.
Additional Comments:	This portion of the evaluation was on accumulation of DBP in armyworm tissue after an exposure via diet. This is for the F1 generation. ADME was selected as the outcome of interest. The study received an unacceptable ranking due to the lack of information on the study duration.			

Overall Quality Determination**Uninformative**

Study Citation:	Filho, D.N., I., Viecei, N. C., Cardoso, E. M., Lovatel, E. R., Gonzatti, C. F., Marzotto, J. A., Montezano, D. G., Specht, A. (2013). Two generations of fall armyworm (Lepidoptera: Noctuidae) contamination by di-n-butylphthalate. Journal of Toxicology and Environmental Health, Part A: Current Issues 76(16):973-977.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Spodoptera frugiperda</i> ; F1 generation; Larvae			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2219889			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The test substance was identified by name only.	
	Metric 2: Test Substance Source	Low	The source of the DBP was not reported.	
	Metric 3: Test Substance Purity	High	The purity was reported to be >99%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of a concurrent negative control.	
	Metric 5: Negative Control Response	High	The negative control response was reported in Table 1 and was adequate for the outcome of interest.	
	Metric 6: Randomized Allocation	Low	It was not reported how the armyworms were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The test system used 150mL plastic cups with plastic caps. The DBP was dissolved into n-hexane at 1000mg/L. This was added to the food at 1mg/g. More information is needed on the test system.	
	Metric 8: Consistency of Exposure Administration	Low	Plastic cups were used as the test chambers, so there is concern about the consistency of the exposure due to potential leaching and sorbing of the test substance.	
	Metric 9: Measurement of Test Substance Concentration	Low	It is unclear if the DBP concentration in the food was measured at any point in the study. Analysis was done on the worms themselves.	
	Metric 10: Exposure Duration and Frequency	Uninformative	The exposure duration was reported to be until death in the adult phase. Authors also reported the use of the F1 moths to produce the F2 larvae. It is unclear how long the exposure actually was.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	N/A	There was only one exposure level, as the goal was not to observe a dose response.	
	Metric 12: Testing at or Below Solubility Limit	N/A	The exposure was via diet.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The organisms were reported to be from an in-house colony at Laboratory of Pest Control and Insect Maintenance at the University of Craxia do Sol.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the organisms were acclimated.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 100 larvae placed individually into 150mL plastic cups.	
Domain 5: Outcome Assessment				

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Study Citation:	Filho, D.N., I., Vieceli, N. C., Cardoso, E. M., Lovatel, E. R., Gonzatti, C. F., Marzotto, J. A., Montezano, D. G., Specht, A. (2013). Two generations of fall armyworm (Lepidoptera: Noctuidae) contamination by di-n-butylphthalate. Journal of Toxicology and Environmental Health, Part A: Current Issues 76(16):973-977.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Spodoptera frugiperda</i> ; F1 generation; Larvae			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2219889			
Domain	Metric	Rating	Comments	
	Metric 16: Adequacy of Test Conditions	Medium	Organisms were kept at 25c with a relative humidity of 70% with a 14L:10D photoperiod. Organism were fed, as the exposure was via diet, but the feeding regimen was not reported.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—weight, length, and width of the organisms.	
	Metric 18: Consistency of Outcome Assessment	Medium	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. Male and female pupae were measured and weighed, but details regarding this were limited.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. It was unclear if the organisms were acclimated to test conditions.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	The statistical methods were described in the "Statistical Analysis " section of the paper.	
	Metric 22: Reporting of Data	High	Data for the exposure and control responses was presented in Table 1 and was adequate for the outcome of interest.	
	Metric 23: Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was presented in Table 1.	
Additional Comments:	This portion of the evaluation was on the effect of DBP on armyworm growth after exposure via diet. This is for the F1 generation. Development/growth was selected as the outcome of interest. The study received an unacceptable ranking due to the lack of information on the study duration.			

Overall Quality Determination**Uninformative**

Study Citation:	Filho, D.N., I., Vieceli, N. C., Cardoso, E. M., Lovatel, E. R., Gonzatti, C. F., Marzotto, J. A., Montezano, D. G., Specht, A. (2013). Two generations of fall armyworm (Lepidoptera: Noctuidae) contamination by di-n-butylphthalate. Journal of Toxicology and Environmental Health, Part A: Current Issues 76(16):973-977.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Spodoptera frugiperda</i> ; F2 generation; Larvae			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2219889			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The test substance was identified by name only.	
	Metric 2: Test Substance Source	Low	The source of the DBP was not reported.	
	Metric 3: Test Substance Purity	High	The purity was reported to be >99%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of a concurrent negative control.	
	Metric 5: Negative Control Response	High	The negative control response was reported in Table 1 and was adequate for the outcome of interest.	
	Metric 6: Randomized Allocation	Low	It was not reported how the armyworms were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The test system used 150mL plastic cups with plastic caps. The DBP was dissolved into n-hexane at 1000mg/L. This was added to the food at 1mg/g. More information is needed on the test system.	
	Metric 8: Consistency of Exposure Administration	Low	Plastic cups were used as the test chambers, so there is concern about the consistency of the exposure due to potential leaching and sorbing of the test substance.	
	Metric 9: Measurement of Test Substance Concentration	Low	It is unclear if the DBP concentration in the food was measured at any point in the study. Analysis was done on the worms themselves.	
	Metric 10: Exposure Duration and Frequency	Uninformative	The exposure duration was reported to be until death in the adult phase. Authors also reported the use of the F1 moths to produce the F2 larvae. It is unclear how long the exposure actually was.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	N/A	There was only one exposure level, as the goal was not to observe a dose response.	
	Metric 12: Testing at or Below Solubility Limit	N/A	The exposure was via diet.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The organisms were reported to be from an in-house colony at Laboratory of Pest Control and Insect Maintenance at the University of Craxia do Sol.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the organisms were acclimated.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 100 larvae placed individually into 150mL plastic cups.	
Domain 5: Outcome Assessment				

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Study Citation:	Filho, D.N., I., Vieceli, N. C., Cardoso, E. M., Lovatel, E. R., Gonzatti, C. F., Marzotto, J. A., Montezano, D. G., Specht, A. (2013). Two generations of fall armyworm (Lepidoptera: Noctuidae) contamination by di-n-butylphthalate. Journal of Toxicology and Environmental Health, Part A: Current Issues 76(16):973-977.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Spodoptera frugiperda</i> ; F2 generation; Larvae			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2219889			
Domain	Metric	Rating	Comments	
	Metric 16: Adequacy of Test Conditions	Medium	Organisms were kept at 25c with a relative humidity of 70% with a 14L:10D photoperiod. Organism were fed, as the exposure was via diet, but the feeding regimen was not reported.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest–weight, length, and width of the organisms.	
	Metric 18: Consistency of Outcome Assessment	Medium	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. Male and female pupae were measured and weighed, but details regarding this were limited.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. It was unclear if the organisms were acclimated to test conditions.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	The statistical methods were described in the "Statistical Analysis " section of the paper.	
	Metric 22: Reporting of Data	High	Data for the exposure and control responses was presented in Table 1 and was adequate for the outcome of interest.	
	Metric 23: Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was presented in Table 1.	
Additional Comments:	This portion of the evaluation was on the effect of DBP on armyworm growth after exposure via diet. This is for the F2 generation. Development/growth was selected as the outcome of interest. The study received an unacceptable ranking due to the lack of information on the study duration.			

Overall Quality Determination**Uninformative**

Study Citation:	Filho, D.N., I., Vieceli, N. C., Cardoso, E. M., Lovatel, E. R., Gonzatti, C. F., Marzotto, J. A., Montezano, D. G., Specht, A. (2013). Two generations of fall armyworm (Lepidoptera: Noctuidae) contamination by di-n-butylphthalate. Journal of Toxicology and Environmental Health, Part A: Current Issues 76(16):973-977.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Spodoptera frugiperda</i> ; F2 generation; Larvae			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2219889			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The test substance was identified by name only.
	Metric 2:	Test Substance Source	Low	The source of the DBP was not reported.
	Metric 3:	Test Substance Purity	High	The purity was reported to be >99%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of a concurrent negative control.
	Metric 5:	Negative Control Response	High	The negative control response was reported in Figure 1 and was adequate for the outcome of interest.
	Metric 6:	Randomized Allocation	Low	It was not reported how the armyworms were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The test system used 150mL plastic cups with plastic caps. The DBP was dissolved into n-hexane at 1000mg/L. This was added to the food at 1mg/g. More information is needed on the test system.
	Metric 8:	Consistency of Exposure Administration	Low	Plastic cups were used as the test chambers, so there is concern about the consistency of the exposure due to potential leaching and sorbing of the test substance.
	Metric 9:	Measurement of Test Substance Concentration	Low	It is unclear if the DBP concentration in the food was measured at any point in the study. Analysis was done on the worms themselves.
	Metric 10:	Exposure Duration and Frequency	Uninformative	The exposure duration was reported to be until death in the adult phase. Authors also reported the use of the F1 moths to produce the F2 larvae. It is unclear how long the exposure actually was.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	There was only one exposure level, as the goal was not to observe a dose response.
	Metric 12:	Testing at or Below Solubility Limit	N/A	The exposure was via diet.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The organisms were reported to be from an in-house colony at Laboratory of Pest Control and Insect Maintenance at the University of Craxia do Sol.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the organisms were acclimated.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	There were 100 larvae placed individually into 150mL plastic cups.
Domain 5: Outcome Assessment				

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Study Citation:	Filho, D.N., I., Vieceli, N. C., Cardoso, E. M., Lovatel, E. R., Gonzatti, C. F., Marzotto, J. A., Montezano, D. G., Specht, A. (2013). Two generations of fall armyworm (Lepidoptera: Noctuidae) contamination by di-n-butylphthalate. Journal of Toxicology and Environmental Health, Part A: Current Issues 76(16):973-977.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Spodoptera frugiperda</i> ; F2 generation; Larvae			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2219889			
Domain		Metric	Rating	Comments
	Metric 16:	Adequacy of Test Conditions	Medium	Organisms were kept at 25c with a relative humidity of 70% with a 14L:10D photoperiod. Organism were fed, as the exposure was via diet, but the feeding regimen was not reported.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest–accumulation of DBP in armyworm tissue.
	Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. Male and female pupa were ground and an extraction was done. GC was used to analyzed DBP levels.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. It was unclear if the organisms were acclimated to test conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	The statistical methods were described in the "Statistical Analysis " section of the paper.
	Metric 22:	Reporting of Data	High	Data for the exposure and control responses was presented in Figure 1 and was adequate for the outcome of interest.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was presented in Figure 1.
Additional Comments:	This portion of the evaluation was on accumulation of DBP in armyworm tissue after an exposure via diet. This is for the F2 generation. ADME was selected as the outcome of interest. The study received an unacceptable ranking due to the lack of information on the study duration.			

Overall Quality Determination**Uninformative**

Study Citation:	Tak, J. H., Kim, H. K., Lee, S. H., Ahn, Y. J. (2006). Acaricidal activities of paeonol and benzoic acid from <i>Paeonia suffruticosa</i> root bark and monoterpenoids against <i>Tyrophagus putrescentiae</i> (Acari: Acaridae). <i>Pest Management Science</i> 62(6):551-557.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Tyrophagus putrescentiae</i> ; Adult
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	1323221

Domain	Metric	Rating	Comments
Domain 1: Test Substance			
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3: Test Substance Purity	Low	Purity or grade of the test substance were not reported.
Domain 2: Test Design			
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5: Negative Control Response	Low	The biological response of the negative control groups was not reported for the vapor phase tests.
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization			
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8: Consistency of Exposure Administration	Medium	A one-time dose was used, but few details were provided.
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Medium	Eight concentrations were tested, but it is not clear if this was for all compounds.
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via contact.
Domain 4: Test Organism			
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 25-30 mites with 3-6 replicates.
Domain 5: Outcome Assessment			

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Study Citation:	Tak, J. H., Kim, H. K., Lee, S. H., Ahn, Y. J. (2006). Acaricidal activities of paeonol and benzoic acid from <i>Paeonia suffruticosa</i> root bark and monoterpenoids against <i>Tyrophagus putrescentiae</i> (Acari: Acaridae). Pest Management Science 62(6):551-557.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Tyrophagus putrescentiae</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1323221			
Domain	Metric	Rating	Comments	
	Metric 16:	Adequacy of Test Conditions	Medium	Environmental conditions of the test system were most likely conducive to maintenance of organism health, but actual measured condition values for control and exposed vessels were not reported.
	Metric 17:	Outcome Assessment Methodology	High	Mites were considered dead if limbs did not move upon being prodded with a wooden dowel.
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described.
	Metric 22:	Reporting of Data	Low	Only slope and LD50s were reported.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments: table 3				
Overall Quality Determination			Medium	

Study Citation:	Tak, J. H., Kim, H. K., Lee, S. H., Ahn, Y. J. (2006). Acaricidal activities of paeonol and benzoic acid from <i>Paeonia suffruticosa</i> root bark and monoterpenoids against <i>Tyrophagus putrescentiae</i> (Acari: Acaridae). <i>Pest Management Science</i> 62(6):551-557.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Tyrophagus putrescentiae</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1323221			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Low	Purity or grade of the test substance were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	Low	The biological response of the negative control group was not reported for the vapor phase tests.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.	
	Metric 8: Consistency of Exposure Administration	Medium	A one-time dose was used, but few details were provided.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Medium	Eight concentrations were tested, but it is not clear if this was for all compounds.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via contact.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 25-30 mites with 3-6 replicates.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Medium	Environmental conditions of the test system were most likely conducive to maintenance of organism health, but actual measured condition values for control and exposed vessels were not reported.	
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Study Citation:	Tak, J. H., Kim, H. K., Lee, S. H., Ahn, Y. J. (2006). Acaricidal activities of paeonol and benzoic acid from <i>Paeonia suffruticosa</i> root bark and monoterpenoids against <i>Tyrophagus putrescentiae</i> (Acari: Acaridae). Pest Management Science 62(6):551-557.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Tyrophagus putrescentiae</i> ; Adult
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	1323221

Domain	Metric	Rating	Comments
	Metric 17: Outcome Assessment Methodology	High	Mites were considered dead if limbs did not move upon being prodded with a wooden dowel.
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	Low	Only slope and LD50s were reported.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: table 4

Overall Quality Determination**Medium**

Study Citation:	Tak, J. H., Kim, H. K., Lee, S. H., Ahn, Y. J. (2006). Acaricidal activities of paeonol and benzoic acid from <i>Paeonia suffruticosa</i> root bark and monoterpenoids against <i>Tyrophagus putrescentiae</i> (Acari: Acaridae). <i>Pest Management Science</i> 62(6):551-557.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Tyrophagus putrescentiae</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1323221			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Low	Purity or grade of the test substance were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	High	In section 3.2 it is reported that "no mortality was observed in the ethanol-treated controls."	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.	
	Metric 8: Consistency of Exposure Administration	Medium	A one-time dose was used, but few details were provided.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Medium	Eight concentrations were tested, but it is not clear if this was for all compounds.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via contact.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 25-30 mites with 3-6 replicates.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Medium	Environmental conditions of the test system were most likely conducive to maintenance of organism health but actual measured condition values for control and exposed vessels were not reported.	
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Study Citation:	Tak, J. H., Kim, H. K., Lee, S. H., Ahn, Y. J. (2006). Acaricidal activities of paeonol and benzoic acid from <i>Paeonia suffruticosa</i> root bark and monoterpenoids against <i>Tyrophagus putrescentiae</i> (Acari: Acaridae). <i>Pest Management Science</i> 62(6):551-557.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Tyrophagus putrescentiae</i> ; Adult
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	1323221

Domain	Metric	Rating	Comments
	Metric 17: Outcome Assessment Methodology	High	Mites were considered dead if limbs did not move upon being prodded with a wooden dowel.
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	Low	Only slope and LD50s were reported.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: table 2

Overall Quality Determination**Medium**

Study Citation:	Tak, J. H., Kim, H. K., Lee, S. H., Ahn, Y. J. (2006). Acaricidal activities of paeonol and benzoic acid from <i>Paeonia suffruticosa</i> root bark and monoterpenoids against <i>Tyrophagus putrescentiae</i> (Acari: Acaridae). <i>Pest Management Science</i> 62(6):551-557.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Tyrophagus putrescentiae</i> ; Adult			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1323221			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Low	Purity or grade of the test substance were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	Low	The biological response of the negative control group was not reported for the vapor phase tests.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.	
	Metric 8: Consistency of Exposure Administration	Medium	A one-time dose was used, but few details were provided.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Medium	Eight concentrations were tested, but it is not clear if this was for all compounds.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via contact.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 25-30 mites with 3-6 replicates.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Medium	Environmental conditions of the test system were most likely conducive to maintenance of organism health, but actual measured condition values for control and exposed vessels was not reported.	
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Study Citation:	Tak, J. H., Kim, H. K., Lee, S. H., Ahn, Y. J. (2006). Acaricidal activities of paeonol and benzoic acid from <i>Paeonia suffruticosa</i> root bark and monoterpenoids against <i>Tyrophagus putrescentiae</i> (Acari: Acaridae). <i>Pest Management Science</i> 62(6):551-557.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; <i>Tyrophagus putrescentiae</i> ; Adult
Health Outcome:	Mortality
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	1323221

Domain	Metric	Rating	Comments
	Metric 17: Outcome Assessment Methodology	High	Mites were considered dead if limbs did not move upon being prodded with a wooden dowel.
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	Low	Only slope and LD50s were reported.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: table 3

Overall Quality Determination**Medium**

Study Citation:	LãKke, H., Rasmussen, L. (1983). Phytotoxicological effects of Di-(2-ethyl hexyl)-phthalate and Di-n-butyl-phthalate on higher plants in laboratory and field experiments. Environmental Pollution Series A: Ecological and Biological 32(3):179-199.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Dermal (topical application)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Achilla millefolium</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Skin & Connective Tissue			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	9430481			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemicals were identified as di-(2-ethyl hexyl)-phthalate (DEHP) and di-(n-butyl)-phthalate (DBP). No CASRN was provided.
	Metric 2:	Test Substance Source	High	DBP was sourced from Fluka, Switzerland, and DEHP was sourced from Scandiflex, Ltd., Denmark. Analytical verification was conducted.
	Metric 3:	Test Substance Purity	High	DPB purity was >99.5%. DEHP purity was >99%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Negative controls contained untreated and solvent-treated (acetone (5%) + water (95%) + Tween 20 (0.1%)).
	Metric 5:	Negative Control Response	Low	The biological response of the negative control group was not reported.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The test material was prepared consistently.
	Metric 8:	Consistency of Exposure Administration	Low	Spraying procedure was not well-defined.
	Metric 9:	Measurement of Test Substance Concentration	High	The test concentrations were measured in the leaves immediately following treatment (day 0) and 1, 3, 7, 15 days after treatment.
	Metric 10:	Exposure Duration and Frequency	High	The one-time application followed by 15 days of monitoring seems adequate to determine chemical uptake by plants.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	There was a single exposure concentration.
	Metric 12:	Testing at or Below Solubility Limit	Low	Reporting omissions prevented determination of whether exposure concentrations exceeded the water solubility limit in the original application. Application concentration was presented in terms of ug/cm^2. A solvent was used, so this likely enhanced the solubility of the test material.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The test species were thin-cuticle species (<i>Sinapis alba</i> L.), a thick cuticle species (<i>Brasica napus</i>), and a common, wild-growing species (<i>Achillea millefolium</i> L.). No source was provided for the seeds.
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Study Citation:	LäKke, H., Rasmussen, L. (1983). Phytotoxicological effects of Di-(2-ethyl hexyl)-phthalate and Di-n-butyl-phthalate on higher plants in laboratory and field experiments. Environmental Pollution Series A: Ecological and Biological 32(3):179-199.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Dermal (topical application)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Achilla millefolium</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Skin & Connective Tissue			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	9430481			
Domain	Metric	Rating	Comments	
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions and all pretreatment conditions were the same for control and exposed organisms, such that the only difference was exposure to test substance.
	Metric 15:	Number of Organisms and Replicates per Group	Low	Twelve plants per species were utilized for the laboratory experiment. It was unclear whether there was a single replicate or multiple replicates with smaller numbers of plants.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Organism housing, environmental conditions, food, water, and nutrients were conducive to maintenance of health and biomass loading was appropriate as shown in Table 1. Biomass loading was not described.
	Metric 17:	Outcome Assessment Methodology	Low	Chlorotic spot assessment was not described in the methods section. However, this is an observational assessment.
	Metric 18:	Consistency of Outcome Assessment	Low	Since methods were not described, it is uncertain whether assessment was consistent.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure (e.g., infection) that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis for chlorophyll assessment was not adequately described in the methods.
	Metric 22:	Reporting of Data	Medium	No data was shown for chlorotic spots. Results were described in the text on pages 8 and 13 of the pdf.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes, or unexpected outcomes were satisfactorily explained.
Additional Comments:	This form applies to all three plant species and both chemicals. Note that results for Brassica treated with DEHP in this laboratory experiment were not found in the publication. The study reported the absorption of the test materials through leaf tissue following spray application to leaves. The authors conducted the test as a timecourse of a single dose (therefore no information about whether the exposure concentration affects the uptake). Evaporation from plants also characterized for some species and data from some species specified amount absorbed vs amount on surface of the plant. The authors tested the components of the test chamber (carbon air filter) as well as the plant.			

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Study Citation:	LäKke, H., Rasmussen, L. (1983). Phytotoxicological effects of Di-(2-ethyl hexyl)-phthalate and Di-n-butyl-phthalate on higher plants in laboratory and field experiments. Environmental Pollution Series A: Ecological and Biological 32(3):179-199.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Dermal (topical application)
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Achilla millefolium</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Skin & Connective Tissue
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	9430481

Domain	Metric	Rating	Comments
Overall Quality Determination		Medium	

Study Citation:	LãKke, H., Rasmussen, L. (1983). Phytotoxicological effects of Di-(2-ethyl hexyl)-phthalate and Di-n-butyl-phthalate on higher plants in laboratory and field experiments. Environmental Pollution Series A: Ecological and Biological 32(3):179-199.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Dermal (topical application)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Achilla millefolium</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	9430481			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
Metric 1:	Test Substance Identity	Low	Chemicals were identified as di-(2-ethyl hexyl)-phthalate (DEHP) and di-(n-butyl)-phthalate (DBP). No CASRN provided..	
Metric 2:	Test Substance Source	High	DBP was sourced from Fluka, Switzerland, and DEHP was sourced from Scandiflex, Ltd., Denmark. Analytical verification was conducted.	
Metric 3:	Test Substance Purity	High	DPB Purity >99.5%; DEHP Purity >99%.	
Domain 2: Test Design				
Metric 4:	Negative Controls	High	Negative controls contained untreated and solvent-treated (acetone (5%) + water (95%) + Tween 20 (0.1%)).	
Metric 5:	Negative Control Response	Low	The biological response of the negative control groups was not reported.	
Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
Metric 7:	Experimental System/Test Media Preparation	Medium	The test material was prepared consistently.	
Metric 8:	Consistency of Exposure Administration	Low	Spraying procedure not well-defined.	
Metric 9:	Measurement of Test Substance Concentration	High	The test concentrations were measured in the leaves immediately following treatment (day 0) and 1, 3, 7, 15 days after treatment.	
Metric 10:	Exposure Duration and Frequency	High	One-time application followed by 15 day monitoring seems adequate to determine chemical uptake by plants.	
Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	Single exposure concentration.	
Metric 12:	Testing at or Below Solubility Limit	Low	Reporting omissions prevented determination of whether exposure concentrations exceeded the water solubility limit in original application. Application concentration was presented in terms of ug/cm^2. A solvent was used so this likely enhanced the solubility of the test material.	
Domain 4: Test Organism				
Metric 13:	Test Organism Characteristics	Low	The test species were thin-cuticle species (Sinapis alba L.), a thick cuticle species (Brasica napus), and a common, wild-growing species (Achillea millefolium L.). No source was provided for the seeds.	
Metric 14:	Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions and all pretreatment conditions were the same for control and exposed organisms, such that the only difference was exposure to test substance.	
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Study Citation:	LäKke, H., Rasmussen, L. (1983). Phytotoxicological effects of Di-(2-ethyl hexyl)-phthalate and Di-n-butyl-phthalate on higher plants in laboratory and field experiments. Environmental Pollution Series A: Ecological and Biological 32(3):179-199.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Dermal (topical application)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Achilla millefolium</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	9430481			
Domain	Metric	Rating	Comments	
	Metric 15:	Number of Organisms and Replicates per Group	Low	12 plants per species utilized for the laboratory experiment per species; unclear whether this was a single replicate or multiple replicate with smaller numbers of plants.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Organism housing, environmental conditions, food, water, and nutrients were conducive to maintenance of health and biomass loading was appropriate as shown in Table 1. Biomass loading not described.
	Metric 17:	Outcome Assessment Methodology	High	Sampling, extraction, and measurement of chemicals from plant material and from carbon air filters in the chamber described adequately. The units expressed by the authors are in ug chemical per plant. The authors did not characterize metabolic products or translocation of the material throughout the plant.
	Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure (e.g., infection) that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Authors performed a time course but did not report significant differences in chemical concentration per plant. Approximate data points provided in Figures 1-5.
	Metric 22:	Reporting of Data	Medium	Data for treated plants as ug chemical per plant shown in Figures 1-5 but no data for controls was represented.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes, or unexpected outcomes were satisfactorily explained.
Additional Comments:	This form applies to all three plant species and both test chemicals. Note that results for Brassica treated with DEHP in this laboratory experiment were not found in the publication. Study reported the absorption of the test materials through leaf tissue following spray application to leaves. The authors conducted the test as a timecourse of a single dose (therefore no information about whether the exposure concentration affects the uptake). Evaporation from plants also characterized for some species and data from some species specified amount absorbed vs amount on surface of the plant. The authors tested the components of the test chamber (carbon air filter) as well as the plant.			

Overall Quality Determination**Medium**

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Study Citation:	LäKke, H., Rasmussen, L. (1983). Phytotoxicological effects of Di-(2-ethyl hexyl)-phthalate and Di-n-butyl-phthalate on higher plants in laboratory and field experiments. Environmental Pollution Series A: Ecological and Biological 32(3):179-199.		
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days		
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Dermal (topical application)		
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Achilla millefolium</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	ADME (biotransformation)		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	9430481		
Domain	Metric	Rating	Comments

Study Citation:	LãKke, H., Rasmussen, L. (1983). Phytotoxicological effects of Di-(2-ethyl hexyl)-phthalate and Di-n-butyl-phthalate on higher plants in laboratory and field experiments. Environmental Pollution Series A: Ecological and Biological 32(3):179-199.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Dermal (topical application)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Achilla millefolium</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Photosynthesis			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	9430481			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemicals were identified as di-(2-ethyl hexyl)-phthalate (DEHP) and di-(n-butyl)-phthalate (DBP). No CASRN provided.
	Metric 2:	Test Substance Source	High	DBP was sourced from Fluka, Switzerland, and DEHP was sourced from Scandiflex, Ltd., Denmark. Analytical verification was conducted.
	Metric 3:	Test Substance Purity	High	DPB Purity >99.5%; DEHP Purity >99%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Negative controls contained untreated and solvent-treated (acetone (5%) + water (95%) + Tween 20 (0.1%)).
	Metric 5:	Negative Control Response	Low	The biological response of the negative control groups was not reported.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The test material was prepared consistently.
	Metric 8:	Consistency of Exposure Administration	Low	Spraying procedure not well-defined.
	Metric 9:	Measurement of Test Substance Concentration	High	The test concentrations were measured in the leaves immediately following treatment (day 0) and 1, 3, 7, 15 days after treatment.
	Metric 10:	Exposure Duration and Frequency	High	One-time application followed by 15 day monitoring seems adequate to determine chemical uptake by plants.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	Single exposure concentration.
	Metric 12:	Testing at or Below Solubility Limit	Low	Reporting omissions prevented determination of whether exposure concentrations exceeded the water solubility limit in original application. Application concentration was presented in terms of ug/cm^2. A solvent was used so this likely enhanced the solubility of the test material.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The test species were thin-cuticle species (Sinapis alba L.), a thick cuticle species (Brasica napus), and a common, wild-growing species (Achillea millefolium L.). No source was provided for the seeds.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions and all pretreatment conditions were the same for control and exposed organisms, such that the only difference was exposure to test substance.
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Study Citation:	LãKke, H., Rasmussen, L. (1983). Phytotoxicological effects of Di-(2-ethyl hexyl)-phthalate and Di-n-butyl-phthalate on higher plants in laboratory and field experiments. Environmental Pollution Series A: Ecological and Biological 32(3):179-199.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Dermal (topical application)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Achilla millefolium</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Photosynthesis			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	9430481			
Domain	Metric	Rating	Comments	
	Metric 15: Number of Organisms and Replicates per Group	Low	12 plants per species utilized for the laboratory experiment per species; unclear whether this was a single replicate or multiple replicate with smaller numbers of plants.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Medium	Organism housing, environmental conditions, food, water, and nutrients were conducive to maintenance of health and biomass loading was appropriate as shown in Table 1. Biomass loading not described.	
	Metric 17: Outcome Assessment Methodology	High	Chlorophyll analysis was described in the Analytical techniques section.	
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure (e.g., infection) that could influence the outcome assessment.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	Low	Statistical analysis for chlorophyll assessment was not adequately described in the methods.	
	Metric 22: Reporting of Data	Low	No data was shown for chlorophyll analysis. Brief results were described in the text on page 13 of the pdf.	
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes, or unexpected outcomes were satisfactorily explained.	
Additional Comments:	This form applies to all three plant species and both chemicals. Note that results for Brassica treated with DEHP in this laboratory experiment were not found in the publication. Study reported the absorption of the test materials through leaf tissue following spray application to leaves. The authors conducted the test as a timecourse of a single dose (therefore no information about whether the exposure concentration affects the uptake). Evaporation from plants also characterized for some species and data from some species specified amount absorbed vs amount on surface of the plant. The authors tested the components of the test chamber (carbon air filter) as well as the plant.			

Overall Quality Determination**Medium**

Study Citation:	Isogai, Y., Komoda, Y., Okamoto, T. (1972). Biological activities of n-butyl phthalate and its analogous compounds on various bioassays of plant growth regulators. Scientific papers of the College of General Education, University of Tokyo 22(2):129-135.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Avena sativa</i> ; cv Victory; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5551990			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical name and structure were used to identify the test chemical.
	Metric 2:	Test Substance Source	Low	The source was not reported.
	Metric 3:	Test Substance Purity	Low	Purity or grade of the test substance was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using a concurrent negative control group.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations and test concentrations were not measured. Vessel material was not reported.
	Metric 8:	Consistency of Exposure Administration	Low	The study provided few details on exposure administration.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type.
	Metric 11:	Number of Exposure Groups/Spacing of Exposure Levels	Medium	Only three concentrations were used, but a wide range was tested.
	Metric 12:	Testing at or Below Solubility Limit	Low	Exposure was via an aqueous test substance solution, but the carrier and quantities of material were not reported.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test seeds was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups.
	Metric 15:	Number of Organisms and Replicates per Group	Low	There were fifteen coleoptile sections per treatment, but the number of replicates was not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.
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Study Citation:	Isogai, Y., Komoda, Y., Okamoto, T. (1972). Biological activities of n-butyl phthalate and its analogous compounds on various bioassays of plant growth regulators. Scientific papers of the College of General Education, University of Tokyo 22(2):129-135.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Avena sativa</i> ; cv Victory; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5551990			
Domain	Metric	Rating	Comments	
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was not conducted.
	Metric 22:	Reporting of Data	High	Results were reported in Tables 5 and 6 for the control response and the exposure responses. The data was appropriate for the outcomes of interest.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.
Additional Comments:	This portion of the study was on the plant growth promoting activity of DBP on <i>Avena sativa</i> -cv Victory. Development/Growth was selected as the outcome of interest.			
Overall Quality Determination		Low		

Study Citation:	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. Frontiers of Environmental Science & Engineering 9(2):259-268.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Avena sativa</i> ; Embryo			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Photosynthesis			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2915866			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The chemical was identified by name only.
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity was reported as 99.6%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was adequate.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were justified for a dose response.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Seeds were exposed via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test seeds were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed seeds.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
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Study Citation:	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. Frontiers of Environmental Science & Engineering 9(2):259-268.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Avena sativa</i> ; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Photosynthesis
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	2915866

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	Nothing was reported to indicate there were differences among groups that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Section 2.7 (page 3/10) does not present the statistical tests used to perform analysis other than the program used and p-value for significance.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group and were adequate to determine values for the endpoint.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments:	Pigment content”The results indicate that longer periods of cultivation of the test plants may make it easier to interpret the changes in pigment contents.”		

Overall Quality Determination**High**

Study Citation:	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. Frontiers of Environmental Science & Engineering 9(2):259-268.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Avena sativa</i> ; Embryo			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2915866			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	Chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity reported as 99.6%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was adequate.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.	
	Metric 8: Consistency of Exposure	High	exposures were administered consistently across study groups.	
	Metric 9: Administration Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were justified for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Seeds exposed via soil.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test seeds were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed seeds.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.	
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Study Citation:	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. <i>Frontiers of Environmental Science & Engineering</i> 9(2):259-268.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Avena sativa</i> ; Embryo
Health Outcome:	Reproductive/Teratogenic
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	2915866

Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	Nothing reported to indicate there were differences among groups that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Section 2.7 (page 3/10) does not present the statistical tests used to perform analysis other than the program used and p-value for significance.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group and were adequate to determine values for the endpoint.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments: This form represents the germination rate results presented in Table 1 for <i>Avena sativa</i> with DBP exposure.			

Overall Quality Determination**High**

Study Citation:	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. Frontiers of Environmental Science & Engineering 9(2):259-268.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Avena sativa</i> ; Embryo			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2915866			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	Chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity reported as 99.6%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was adequate.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.	
	Metric 8: Consistency of Exposure	High	exposures were administered consistently across study groups.	
	Metric 9: Administration Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were justified for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Seeds exposed via soil.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test seeds were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed seeds.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.	
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Study Citation:	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. <i>Frontiers of Environmental Science & Engineering</i> 9(2):259-268.		
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)		
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Avena sativa</i> ; Embryo		
Health Outcome:	Development/Growth		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	2915866		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
Metric 20:	Outcomes Unrelated to Exposure	Medium	Nothing reported to indicate there were differences among groups that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	High	Section 2.7 (page 3/10) does not present the statistical tests used to perform analysis other than the program used and p-value for significance.
Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group and were adequate to determine values for the endpoint.
Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments:	"DnBP and DEHP at a range of concentrations in the experimental soil showed no discernible effect on the germination rate of the seven test plant species. However, they did exert effects on root elongation, seedling growth and biomass to different extents, indicating the potential applicability of seedling growth in the evaluation of the phytotoxicity of PAE compounds."		

Overall Quality Determination**High**

Study Citation:	Dueck, T. A., Dijk, Van, C. J., David, F., Scholz, N., Vanwalleghem, F. (2003). Chronic effects of vapour phase di-n-butyl phthalate (DBP) on six plant species. Chemosphere 53(8):911-920.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica campestris</i> ; chinensis; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1302103			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Study authors reported using an appropriate concurrent negative control group, note that all controls had some DBP detected. Control group DBP concentrations are reported in Table 1.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail. The chambers were constructed of "hardened glass" and aluminum.
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups based on timed samplings
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies. Samples were taken frequently: "Actual DBP concentrations were measured by taking air samples from each chamber using an automated gas sampling device (GS-1 gas sampler, Gerstel, M €ulheim, Germany). Sampling was performed with Tenax TA tubes (2,6-diphenylphenylene oxide polymer) at 250ml min ¹ for 20 min. During the fumigation experiment, each chamber was sampled in duplicate on a weekly basis during the complete exposure period (76 days)."
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	The number of exposure groups and spacing of exposure levels were suitable. Concentration gradient is reported as both actual and nominal within table 1.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via fumigation in a continually renewed chamber.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The crop and herbaceous species originated from germinated seeds, but source was not explicitly reported.

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Study Citation:	Dueck, T. A., Dijk, Van, C. J., David, F., Scholz, N., Vanwalleghem, F. (2003). Chronic effects of vapour phase di-n-butyl phthalate (DBP) on six plant species. Chemosphere 53(8):911-920.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica campestris</i> ; chinensis; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1302103			
Domain	Metric	Rating	Comments	
	Metric 14: Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions	
	Metric 15: Number of Organisms and Replicates per Group	Low	Four plants per treatment and no replicates reported, "Three to four weeks after sowing, when the first mature leaves appeared, four uniform individuals per species including the tree species were placed in each fumigation chamber (the maximum number physically possible) for exposure to DBP.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	
	Metric 18: Consistency of Outcome Assessment	Medium	Outcomes were assessed consistently across study groups but few details were reported.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.	
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.	
	Metric 23: Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.	
Additional Comments:	This form represents growth outcomes associated with Dry Weight reported for Brassica shoot and roots within Figure 3 on page 6/10.			
Overall Quality Determination		High		

Study Citation:	Dueck, T. A., Dijk, Van, C. J., David, F., Scholz, N., Vanwalleghem, F. (2003). Chronic effects of vapour phase di-n-butyl phthalate (DBP) on six plant species. Chemosphere 53(8):911-920.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica campestris</i> ; chinensis; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1302103			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The chemical was identified by name and CAS#.
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity or grade of the test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Study authors reported using an appropriate concurrent negative control group, note that all controls had some DBP detected. Control group DBP concentrations are reported in Table 1.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail. The chambers were constructed of "hardened glass" and aluminum.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups based on timed samplings.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies. Samples were taken frequently: "Actual DBP concentrations were measured by taking air samples from each chamber using an automated gas sampling device (GS-1 gas sampler, Gerstel, M €ulheim, Germany). Sampling was performed with Tenax TA tubes (2,6-diphenylphenylene oxide polymer) at 250ml min ¹ for 20 min. During the fumigation experiment, each chamber was sampled in duplicate on a weekly basis during the complete exposure period (76 days)."
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	The number of exposure groups and spacing of exposure levels were suitable. Concentration gradient is reported as both actual and nominal within table 1.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via fumigation in a continually renewed chamber.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The crop and herbaceous species originated from germinated seeds, but the source was not explicitly reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions.
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Study Citation:	Dueck, T. A., Dijk, Van, C. J., David, F., Scholz, N., Vanwalleghem, F. (2003). Chronic effects of vapour phase di-n-butyl phthalate (DBP) on six plant species. Chemosphere 53(8):911-920.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica campestris</i> ; chinensis; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1302103			
Domain	Metric	Rating	Comments	
	Metric 15: Number of Organisms and Replicates per Group	Low	There were four plants per treatment and no replicates reported. "Three to four weeks after sowing, when the first mature leaves appeared, four uniform individuals per species including the tree species were placed in each fumigation chamber (the maximum number physically possible) for exposure to DBP."	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	
	Metric 18: Consistency of Outcome Assessment	Medium	Outcomes were assessed consistently across study groups but few details were reported.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.	
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group in Table 2.	
	Metric 23: Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.	
Additional Comments:	This form is for ADME (biotransformation) assessment of DBP concentration in leaf tissue.			
Overall Quality Determination		High		

Study Citation:	Kong, X., Jin, D., Jin, S., Wang, Z., Yin, H., Xu, M., Deng, Y. (2018). Responses of bacterial community to dibutyl phthalate pollution in a soil-vegetable ecosystem. Journal of Hazardous Materials 353(Elsevier):142-150.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica napus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Photosynthesis-Nutritional and Metabolic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	4829418			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	High	The test substance was identified by HPLC.
	Metric 3:	Test Substance Purity	Low	Purity or grade of test substance was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using a concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentration. Exposure was conducted in a greenhouse with seedlings contained in ceramic pots.
	Metric 8:	Consistency of Exposure Administration	Low	Only general methods of exposure administration were reported so assessment was difficult to determine
	Metric 9:	Measurement of Test Substance Concentration	Low	Supplemental data needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels was suitable
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	Source was suitable, age of seedlings was not reported
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups
	Metric 15:	Number of Organisms and Replicates per Group	Medium	10 seedlings with six replicates was reasonable
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate

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Study Citation:	Kong, X., Jin, D., Jin, S., Wang, Z., Yin, H., Xu, M., Deng, Y. (2018). Responses of bacterial community to dibutyl phthalate pollution in a soil-vegetable ecosystem. Journal of Hazardous Materials 353(Elsevier):142-150.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica napus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Photosynthesis-Nutritional and Metabolic
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	4829418

Domain	Metric	Rating	Comments
	Metric 17: Outcome Assessment Methodology	Low	Few details regarding the execution of the study protocol for outcome assessment were reported.
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23: Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained

Additional Comments: None

Overall Quality Determination**Medium**

Study Citation:	Kong, X., Jin, D., Jin, S., Wang, Z., Yin, H., Xu, M., Deng, Y. (2018). Responses of bacterial community to dibutyl phthalate pollution in a soil-vegetable ecosystem. Journal of Hazardous Materials 353(Elsevier):142-150.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica napus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	4829418			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	Chemical was identified by name only	
	Metric 2: Test Substance Source	High	The test substance was identified by HPLC.	
	Metric 3: Test Substance Purity	Low	Purity or grade of test substance was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentration. Exposure was conducted in a greenhouse with seedlings contained in ceramic pots.	
	Metric 8: Consistency of Exposure Administration	Low	Only general methods of exposure administration were reported so assessment was difficult to determine	
	Metric 9: Measurement of Test Substance Concentration	Low	Supplemental data needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels was suitable	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via soil	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Medium	Source was suitable, age of seedlings was not reported	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups	
	Metric 15: Number of Organisms and Replicates per Group	Medium	10 seedlings with six replicates was reasonable	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate	
	Metric 17: Outcome Assessment Methodology	Low	Few details regarding the execution of the study protocol for outcome assessment were reported.	
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Study Citation:	Kong, X., Jin, D., Jin, S., Wang, Z., Yin, H., Xu, M., Deng, Y. (2018). Responses of bacterial community to dibutyl phthalate pollution in a soil-vegetable ecosystem. Journal of Hazardous Materials 353(Elsevier):142-150.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica napus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	ADME (biotransformation)
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	4829418

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described
	Metric 22: Reporting of Data	Low	Supplemental data needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
	Metric 23: Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained

Additional Comments: None

Overall Quality Determination**Medium**

Study Citation:	Kong, X., Jin, D., Jin, S., Wang, Z., Yin, H., Xu, M., Deng, Y. (2018). Responses of bacterial community to dibutyl phthalate pollution in a soil-vegetable ecosystem. Journal of Hazardous Materials 353(Elsevier):142-150.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica napus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	4829418			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	High	The test substance was identified by HPLC.	
	Metric 3: Test Substance Purity	Low	Purity or grade of the test substance were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations. The exposure was conducted in a greenhouse with seedlings contained in ceramic pots.	
	Metric 8: Consistency of Exposure Administration	Low	Only general methods of exposure administration were reported so assessment was difficult to determine.	
	Metric 9: Measurement of Test Substance Concentration	Low	Supplemental data needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels was suitable.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via soil.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Medium	The organism source was adequate. The age of seedlings was not reported.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The 10 seedlings with six replicates was reasonable.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.	
	Metric 17: Outcome Assessment Methodology	Low	Few details regarding the execution of the study protocol for outcome assessment were reported.	
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Study Citation:	Kong, X., Jin, D., Jin, S., Wang, Z., Yin, H., Xu, M., Deng, Y. (2018). Responses of bacterial community to dibutyl phthalate pollution in a soil-vegetable ecosystem. Journal of Hazardous Materials 353(Elsevier):142-150.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica napus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	4829418

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.

Additional Comments: None

Overall Quality Determination**Medium**

Study Citation:	Hardwick, R. C., Cole, R. A., Fyfield, T. P. (1984). Injury to and death of cabbage (brassica-oleracea) seedlings caused by vapors of di butyl phthalate emitted from certain plastics. Annals of Applied Biology 105(1):97-105.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica oleracea</i> ; Derby Day cultivar; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5678863			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
Metric 1:	Test Substance Identity	Low	The DBP originated from glazing strips in a greenhouse. DBP was measured in the greenhouse or in the cuvettes during the experiment. However, it is possible there was contamination from substances other than DBP is the glazing strips. DBP was reported to be used as a plasticizer for the strips, but other chemical exposure cannot be ruled out.	
Metric 2:	Test Substance Source	Low	The source of the glazing strip was not reported.	
Metric 3:	Test Substance Purity	Low	The purity of the DBP was not reported. The	
Domain 2: Test Design				
Metric 4:	Negative Controls	High	It was reported there was negative control that contained zero plastic.	
Metric 5:	Negative Control Response	Low	The biological response of the negative control was not reported. Several different plas- tics were reported to be testing, many of which had ND or a trace of DBP, but these did not appear to be the true negative control response.	
Metric 6:	Randomized Allocation	Low	It was not reported how the plants were allocated into study groups.	
Domain 3: Exposure Characterization				
Metric 7:	Experimental System/Test Media Preparation	Low	The test system was reported to be conducted in glass cuvettes containing 115mm di- ameter dwarf pots with 4 plants each. The plastic was suspended on a wire at the top of the cuvette that was ventilated. There was a non-plastic control for each type of plastic used. Measurements were taken of the DBP in the air. It is uncertain if the exposure was to DBP alone since other chemicals are involved in making plastic.	
Metric 8:	Consistency of Exposure Administration	Medium	The exposure was reported to be for 4 weeks for all plants. All the plants were visually assessed for 1 month post sowing. All seedling were raised in 115mm diameter dwarf pots with 4 plants per pot.	
Metric 9:	Measurement of Test Substance Concentration	High	Samples of air from the test environment were measured using gas chromatography to test for various phthalates.	
Metric 10:	Exposure Duration and Frequency	High	The exposure duration was reported to be 4 weeks. This appeared adequate to observe the outcome of interest.	
Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	The purpose of the study was not to observe a dose response, to compare a glazing using DBP to a glazing using DIDP and to other plastics potentially plasticized with DBP.	
Metric 12:	Testing at or Below Solubility Limit	High	The test concentrations measured using gas chromatography were below the water solu- bility limit.	
Domain 4: Test Organism				

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Study Citation:	Hardwick, R. C., Cole, R. A., Fyfield, T. P. (1984). Injury to and death of cabbage (brassica-oleracea) seedlings caused by vapors of di butyl phthalate emitted from certain plastics. Annals of Applied Biology 105(1):97-105.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica oleracea</i> ; Derby Day cultivar; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5678863			
Domain	Metric	Rating	Comments	
	Metric 13: Test Organism Characteristics	Low	The cabbage was reported to be the Derby Day cultivar, but the source was not reported.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if any acclimation occurred.	
	Metric 15: Number of Organisms and Replicates per Group	Low	For each plastic tested, there were three replicates. In each cuvette, there were 4 plants per pot.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	The plants were kept at 20C during the day and 15C at night, but little other details were provided on environmental conditions.	
	Metric 17: Outcome Assessment Methodology	Low	It was reported the plants were visually assessed for signs of toxicity 4 weeks after sowing, but details regarding the assessment were not provided.	
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported. Plants were assessed for signs of toxicity, but this protocol was not described.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	Uninformative	Statistical analysis for this portion of the study was not conducted. Results were not presented in a manner that allowed for statistical analysis.	
	Metric 22: Reporting of Data	Medium	Results for the exposures were provided in Table 2, but negative control results were not reported.	
	Metric 23: Explanation of Unexpected Outcomes	Low	Insufficient information was provided to determine if excessive variability or unexpected outcomes occurred.	
Additional Comments:	This portion of the evaluation was conducted in glass cuvettes. The test plastic was suspended from the top of the cuvette and chemicals vaporized into the cuvette. Several plastics were tested. It was reported that a control was run for each type of plastic, but control results were not reported. This study received an unacceptable ranking due to the lack of statistical analysis. There is also concern that other chemicals in the plastic could be vaporizing and causing toxicity as well. The study authors only tested for phthalates. Authors reported visually assessing the plants 1 month after sowing. Details regarding this protocol were not provided. The outcomes assessed for were mortality, growth, and chlorosis.			

Overall Quality Determination

Uninformative

Study Citation:	Hardwick, R. C., Cole, R. A., Fyfield, T. P. (1984). Injury to and death of cabbage (brassica-oleracea) seedlings caused by vapors of di butyl phthalate emitted from certain plastics. Annals of Applied Biology 105(1):97-105.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica oleracea</i> ; Derby Day cultivar; Juvenile			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5678863			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The DBP originated from glazing strips in a greenhouse. DBP was measured in the greenhouse or in the cuvettes during the experiment. However, it is possible there was contamination from substances other than DBP is the glazing strips. DBP was reported to be used as a plasticizer for the strips, but other chemical exposure cannot be ruled out.	
	Metric 2: Test Substance Source	Low	The source of the glazing strip was not reported.	
	Metric 3: Test Substance Purity	Low	The purity of the DBP was not reported. The	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	It was reported there was negative control that contained zero plastic.	
	Metric 5: Negative Control Response	Low	The biological response of the negative control was not reported. Several different plas- tics were reported to be testing, many of which had ND or a trace of DBP, but these did not appear to be the true negative control response.	
	Metric 6: Randomized Allocation	Low	It was not reported how the plants were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The test system was reported to be conducted in glass cuvettes containing 115mm di- ameter dwarf pots with 4 plants each. The plastic was suspended on a wire at the top of the cuvette that was ventilated. There was a non-plastic control for each type of plastic used. Measurements were taken of the DBP in the air. It is uncertain if the exposure was to DBP alone since other chemicals are involved in making plastic.	
	Metric 8: Consistency of Exposure Administration	Medium	The exposure was reported to be for 4 weeks for all plants. All the plants were visually assessed for 1 month post sowing. All seedling were raised in 115mm diameter dwarf pots with 4 plants per pot.	
	Metric 9: Measurement of Test Substance Concentration	High	Samples of air from the test environment were measured using gas chromatography to test for various phthalates.	
	Metric 10: Exposure Duration and Frequency	High	The exposure duration was reported to be 4 weeks. This appeared adequate to observe the outcome of interest.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	N/A	The purpose of the study was not to observe a dose response, to compare a glazing using DBP to a glazing using DIDP and to other plastics potentially plasticized with DBP.	
	Metric 12: Testing at or Below Solubility Limit	High	The test concentrations measured using gas chromatography were below the water solu- bility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The cabbage was reported to be the Derby Day cultivar, but the source was not reported.	
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Study Citation:	Hardwick, R. C., Cole, R. A., Fyfield, T. P. (1984). Injury to and death of cabbage (brassica-oleracea) seedlings caused by vapors of di butyl phthalate emitted from certain plastics. Annals of Applied Biology 105(1):97-105.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica oleracea</i> ; Derby Day cultivar; Juvenile			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5678863			
Domain	Metric	Rating	Comments	
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if any acclimation occurred.
	Metric 15:	Number of Organisms and Replicates per Group	Low	For each plastic tested, there were three replicates. In each cuvette, there were 4 plants per pot.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	The plants were kept at 20C during the day and 15C at night, but little other details were provided on environmental conditions.
	Metric 17:	Outcome Assessment Methodology	Low	It was reported the plants were visually assessed for signs of toxicity 4 weeks after sowing, but details regarding the assessment were not provided.
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported. Plants were assessed for signs of toxicity, but this protocol was not described.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Uninformative	Statistical analysis for this portion of the study was not conducted. Results were not presented in a manner that allowed for statistical analysis.
	Metric 22:	Reporting of Data	Medium	Results for the exposures were provided in Table 2, but negative control results were not reported.
	Metric 23:	Explanation of Unexpected Outcomes	Low	Insufficient information was provided to determine if excessive variability or unexpected outcomes occurred.
Additional Comments:	This portion of the evaluation was conducted in glass cuvettes. The test plastic was suspended from the top of the cuvette and chemicals vaporized into the cuvette. Several plastics were tested. It was reported that a control was run for each type of plastic, but control results were not reported. This study received an unacceptable ranking due to the lack of statistical analysis. There is also concern that other chemicals in the plastic could be vaporizing and causing toxicity as well. The study authors only tested for phthalates. Authors reported visually assessing the plants 1 month after sowing. Details regarding this protocol were not provided. The outcomes assessed for were mortality, growth, and chlorosis.			

Overall Quality Determination**Uninformative**

Study Citation:	Hardwick, R. C., Cole, R. A., Fyfield, T. P. (1984). Injury to and death of cabbage (brassica-oleracea) seedlings caused by vapors of di butyl phthalate emitted from certain plastics. Annals of Applied Biology 105(1):97-105.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica oleracea</i> ; Derby Day cultivar; Juvenile			
Health Outcome:	Mechanistic-Photosynthesis			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5678863			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
Metric 1:	Test Substance Identity	Low	The DBP originated from glazing strips in a greenhouse. DBP was measured in the greenhouse or in the cuvettes during the experiment. However, it is possible there was contamination from substances other than DBP is the glazing strips. DBP was reported to be used as a plasticizer for the strips, but other chemical exposure cannot be ruled out.	
Metric 2:	Test Substance Source	Low	The source of the glazing strip was not reported.	
Metric 3:	Test Substance Purity	Low	The purity of the DBP was not reported.	
Domain 2: Test Design				
Metric 4:	Negative Controls	High	It was reported there was a negative control that contained zero plastic.	
Metric 5:	Negative Control Response	Low	The biological response of the negative control was not reported. Several different plastics were reported to be testing, many of which had ND or a trace of DBP, but these did not appear to be the true negative control response.	
Metric 6:	Randomized Allocation	Low	It was not reported how the plants were allocated into study groups.	
Domain 3: Exposure Characterization				
Metric 7:	Experimental System/Test Media Preparation	Low	The test system was reported to be conducted in glass cuvettes containing 115mm diameter dwarf pots with four plants each. The plastic was suspended on a wire at the top of the cuvette that was ventilated. There was a non-plastic control for each type of plastic used. Measurements were taken of the DBP in the air. It is uncertain if the exposure was to DBP alone since other chemicals are involved in making plastic.	
Metric 8:	Consistency of Exposure Administration	Medium	The exposure was reported to be for four weeks for all plants. All the plants were visually assessed for one month post sowing. All seedlings were raised in 115mm diameter dwarf pots with four plants per pot.	
Metric 9:	Measurement of Test Substance Concentration	High	Samples of air from the test environment were measured using gas chromatography to test for various phthalates.	
Metric 10:	Exposure Duration and Frequency	High	The exposure duration was reported to be four weeks. This appeared adequate to observe the outcome of interest.	
Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	The purpose of the study was not to observe a dose response, but to compare a glazing using DBP to a glazing using DIDP and to other plastics potentially plasticized with DBP.	
Metric 12:	Testing at or Below Solubility Limit	High	The test concentrations measured using gas chromatography were below the water solubility limit.	
Domain 4: Test Organism				
Metric 13:	Test Organism Characteristics	Low	The cabbage was reported to be the Derby Day cultivar, but the source was not reported.	

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Study Citation:	Hardwick, R. C., Cole, R. A., Fyfield, T. P. (1984). Injury to and death of cabbage (brassica-oleracea) seedlings caused by vapors of di butyl phthalate emitted from certain plastics. Annals of Applied Biology 105(1):97-105.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica oleracea</i> ; Derby Day cultivar; Juvenile			
Health Outcome:	Mechanistic-Photosynthesis			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5678863			
Domain	Metric	Rating	Comments	
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if any acclimation occurred.
	Metric 15:	Number of Organisms and Replicates per Group	Low	For each plastic tested, there were three replicates. In each cuvette, there were four plants per pot.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	The plants were kept at 20C during the day and 15C at night, but little other details were provided on environmental conditions.
	Metric 17:	Outcome Assessment Methodology	Low	It was reported the plants were visually assessed for signs of toxicity four weeks after sowing, but details regarding the assessment were not provided.
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported. Plants were assessed for signs of toxicity, but this protocol was not described.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Uninformative	Statistical analysis for this portion of the study was not conducted. Results were not presented in a manner that allowed for statistical analysis.
	Metric 22:	Reporting of Data	Medium	Results for the exposures were provided in Table 2, but negative control results were not reported.
	Metric 23:	Explanation of Unexpected Outcomes	Low	Insufficient information was provided to determine if excessive variability or unexpected outcomes occurred.
Additional Comments:	This portion of the evaluation was conducted in glass cuvettes. The test plastic was suspended from the top of the cuvette and chemicals vaporized into the cuvette. Several plastics were tested. It was reported that a control was run for each type of plastic, but control results were not reported. This study received an unacceptable ranking due to the lack of statistical analysis. There is also concern that other chemicals in the plastic could be vaporizing and causing toxicity as well. The study authors only tested for phthalates. Authors reported visually assessing the plants one month after sowing. Details regarding this protocol were not provided. The outcomes assessed for were mortality, growth, and chlorosis.			

Overall Quality Determination**Uninformative**

Study Citation:	Hardwick, R. C., Cole, R. A., Fyfield, T. P. (1984). Injury to and death of cabbage (brassica-oleracea) seedlings caused by vapors of di butyl phthalate emitted from certain plastics. Annals of Applied Biology 105(1):97-105.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica oleracea</i> ; Derby Day cultivar; Juvenile			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5678863			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The DBP originated from glazing strips in a greenhouse. DBP was measured in the greenhouse or in the cuvettes during the experiment. However, it is possible there was contamination from substances other than DBP is the glazing strips. DBP was reported to be used as a plasticizer for the strips, but other chemical exposure cannot be ruled out.
	Metric 2:	Test Substance Source	Low	The source of the glazing strip was not reported.
	Metric 3:	Test Substance Purity	Low	The purity of the DBP was not reported. The
Domain 2: Test Design				
	Metric 4:	Negative Controls	Uninformative	For the exposure in the greenhouse only, it did not appear as though an appropriate negative control was run concurrently with the test. Study authors reported growing plants in the original greenhouse with the DBP glazed strips and growing plants in the reglazed greenhouse, which reported the use of DIDP as the plasticizing agent. This is not a true chemical-free negative control.
	Metric 5:	Negative Control Response	Uninformative	There was not a true negative control used in this study, so an appropriate negative control response was not reported.
	Metric 6:	Randomized Allocation	Low	It was not reported how the plants were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The test system was reported to be a glasshouse. One pot with 14 seedlings was placed in a compartment with the original glazing that contained DBP and another pot of 14 seedlings was placed in a compartment reglazed with DIDP. Little other details were provided about the test system.
	Metric 8:	Consistency of Exposure Administration	Low	The exposure was reported to be for 4 weeks for all plants. However, according to Table 1, DBP levels were measured and appeared to be quite variable over time in the glasshouse. It is possible that exposure levels could have had a great deal of variation over the 4 week exposure period.
	Metric 9:	Measurement of Test Substance Concentration	High	Samples of air from the test environment were measured using gas chromatography to test for various phthalates.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration was reported to be 16 weeks. This appeared adequate to observe the outcome of interest.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	The purpose of the study was not to observe a dose response, to compare a glazing using DBP to a glazing using DIDP.
	Metric 12:	Testing at or Below Solubility Limit	High	The test concentrations measured using gas chromatography were below the water solubility limit.
Domain 4: Test Organism				
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Study Citation:	Hardwick, R. C., Cole, R. A., Fyfield, T. P. (1984). Injury to and death of cabbage (brassica-oleracea) seedlings caused by vapors of di butyl phthalate emitted from certain plastics. Annals of Applied Biology 105(1):97-105.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica oleracea</i> ; Derby Day cultivar; Juvenile			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5678863			
Domain	Metric	Rating	Comments	
	Metric 13:	Test Organism Characteristics	Low	The cabbage was reported to be the Derby Day cultivar, but the source was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if any acclimation occurred.
	Metric 15:	Number of Organisms and Replicates per Group	Low	There was one pot with 14 plants for each treatment, which is lower than is typical.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	The plants were kept at 20C during the day and 15C at night, but little other details were provided on environmental conditions.
	Metric 17:	Outcome Assessment Methodology	Low	Plants were assessed for signs of toxicity at the end of the test, but this process was not described. No signs of toxicity were observed.
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported. Plats were assessed for signs of toxicity, but this protocol was not described.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Uninformative	Statistical analysis for this portion of the study was not conducted.
	Metric 22:	Reporting of Data	Low	Results were reported in the text only for this portion of the study.
	Metric 23:	Explanation of Unexpected Outcomes	Low	Insufficient information was provided to determine if excessive variability or unexpected outcomes occurred.
Additional Comments:	This portion of the evaluation was conducted in compartments in a glasshouse. One pot was grown in a section that contained glazed strips containing DBP. The other pot was grown in section that contained strips glazed with DIDP. This study received an unacceptable ranking due to the lack of a true negative control and due to lack of statistics. The study authors reported assessing the plants for signs of toxicity. This included mortality, growth, and chlorosis.			

Overall Quality Determination**Uninformative**

Study Citation:	Hardwick, R. C., Cole, R. A., Fyfield, T. P. (1984). Injury to and death of cabbage (brassica-oleracea) seedlings caused by vapors of di butyl phthalate emitted from certain plastics. Annals of Applied Biology 105(1):97-105.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica oleracea</i> ; Derby Day cultivar; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5678863			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The DBP originated from glazing strips in a greenhouse. DBP was measured in the greenhouse or in the cuvettes during the experiment. However, it is possible there was contamination from substances other than DBP is the glazing strips. DBP was reported to be used as a plasticizer for the strips, but other chemical exposure cannot be ruled out.
	Metric 2:	Test Substance Source	Low	The source of the glazing strip was not reported.
	Metric 3:	Test Substance Purity	Low	The purity of the DBP was not reported. The
Domain 2: Test Design				
	Metric 4:	Negative Controls	Uninformative	For the exposure in the greenhouse only, it did not appear as though an appropriate negative control was run concurrently with the test. Study authors reported growing plants in the original greenhouse with the DBP glazed strips and growing plants in the reglazed greenhouse, which reported the use of DIDP as the plasticizing agent. This is not a true chemical-free negative control.
	Metric 5:	Negative Control Response	Uninformative	There was not a true negative control used in this study, so an appropriate negative control response was not reported.
	Metric 6:	Randomized Allocation	Low	It was not reported how the plants were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The test system was reported to be a glasshouse. One pot with 14 seedlings was placed in a compartment with the original glazing that contained DBP and another pot of 14 seedlings was placed in a compartment reglazed with DIDP. Little other details were provided about the test system.
	Metric 8:	Consistency of Exposure Administration	Low	The exposure was reported to be for 4 weeks for all plants. However, according to Table 1, DBP levels were measured and appeared to be quite variable over time in the glasshouse. It is possible that exposure levels could have had a great deal of variation over the 4 week exposure period.
	Metric 9:	Measurement of Test Substance Concentration	High	Samples of air from the test environment were measured using gas chromatography to test for various phthalates.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration was reported to be 16 weeks. This appeared adequate to observe the outcome of interest.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	The purpose of the study was not to observe a dose response, to compare a glazing using DBP to a glazing using DIDP.
	Metric 12:	Testing at or Below Solubility Limit	High	The test concentrations measured using gas chromatography were below the water solubility limit.
Domain 4: Test Organism				
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Study Citation:	Hardwick, R. C., Cole, R. A., Fyfield, T. P. (1984). Injury to and death of cabbage (brassica-oleracea) seedlings caused by vapors of di butyl phthalate emitted from certain plastics. Annals of Applied Biology 105(1):97-105.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica oleracea</i> ; Derby Day cultivar; Juvenile			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5678863			
Domain		Metric	Rating	Comments
	Metric 13:	Test Organism Characteristics	Low	The cabbage was reported to be the Derby Day cultivar, but the source was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if any acclimation occurred.
	Metric 15:	Number of Organisms and Replicates per Group	Low	There was one pot with 14 plants for each treatment, which is lower than is typical.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	The plants were kept at 20C during the day and 15C at night, but little other details were provided on environmental conditions.
	Metric 17:	Outcome Assessment Methodology	Low	Plants were assessed for signs of toxicity at the end of the test, but this process was not described. No signs of toxicity were observed.
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported. Plats were assessed for signs of toxicity, but this protocol was not described.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Uninformative	Statistical analysis for this portion of the study was not conducted.
	Metric 22:	Reporting of Data	Low	Results were reported in the text only for this portion of the study.
	Metric 23:	Explanation of Unexpected Outcomes	Low	Insufficient information was provided to determine if excessive variability or unexpected outcomes occurred.
Additional Comments:	This portion of the evaluation was conducted in compartments in a glasshouse. One pot was grown in a section that contained glazed strips containing DBP. The other pot was grown in section that contained strips glazed with DIDP. This study received an unacceptable ranking due to the lack of a true negative control and due to lack of statistics. The study authors reported assessing the plants for signs of toxicity. This included mortality, growth, and chlorosis.			

Overall Quality Determination**Uninformative**

Study Citation:	Hardwick, R. C., Cole, R. A., Fyfield, T. P. (1984). Injury to and death of cabbage (brassica-oleracea) seedlings caused by vapors of di butyl phthalate emitted from certain plastics. Annals of Applied Biology 105(1):97-105.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica oleracea</i> ; Derby Day cultivar; Juvenile			
Health Outcome:	Mechanistic-Photosynthesis			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5678863			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The DBP originated from glazing strips in a greenhouse. DBP was measured in the greenhouse or in the cuvettes during the experiment. However, it is possible there was contamination from substances other than DBP in the glazing strips. DBP was reported to be used as a plasticizer for the strips, but other chemical exposure cannot be ruled out.
	Metric 2:	Test Substance Source	Low	The source of the glazing strip was not reported.
	Metric 3:	Test Substance Purity	Low	The purity of the DBP was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Uninformative	For the exposure in the greenhouse only, it did not appear as though an appropriate negative control was run concurrently with the test. Study authors reported growing plants in the original greenhouse with the DBP glazed strips and growing plants in the reglazed greenhouse, which reported the use of DIDP as the plasticizing agent. This is not a true chemical-free negative control.
	Metric 5:	Negative Control Response	Uninformative	There was not a true negative control used in this study, so an appropriate negative control response was not reported.
	Metric 6:	Randomized Allocation	Low	It was not reported how the plants were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The test system was reported to be a glasshouse. One pot with 14 seedlings was placed in a compartment with the original glazing that contained DBP and another pot of 14 seedlings was placed in a compartment reglazed with DIDP. Little other details were provided about the test system.
	Metric 8:	Consistency of Exposure Administration	Low	The exposure was reported to be for four weeks for all plants. However, according to Table 1, DBP levels were measured and appeared to be quite variable over time in the glasshouse. It is possible that exposure levels could have had a great deal of variation over the 4-week exposure period.
	Metric 9:	Measurement of Test Substance Concentration	High	Samples of air from the test environment were measured using gas chromatography to test for various phthalates.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration was reported to be 16 weeks. This appeared adequate to observe the outcome of interest.
	Metric 11:	Number of Exposure Groups/Spacing of Exposure Levels	N/A	The purpose of the study was not to observe a dose response, to compare a glazing using DBP to a glazing using DIDP.
	Metric 12:	Testing at or Below Solubility Limit	High	The test concentrations measured using gas chromatography were below the water solubility limit.
Domain 4: Test Organism				
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Study Citation:	Hardwick, R. C., Cole, R. A., Fyfield, T. P. (1984). Injury to and death of cabbage (brassica-oleracea) seedlings caused by vapors of di butyl phthalate emitted from certain plastics. Annals of Applied Biology 105(1):97-105.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica oleracea</i> ; Derby Day cultivar; Juvenile			
Health Outcome:	Mechanistic-Photosynthesis			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5678863			
Domain		Metric	Rating	Comments
	Metric 13:	Test Organism Characteristics	Low	The cabbage was reported to be the Derby Day cultivar, but the source was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if any acclimation occurred.
	Metric 15:	Number of Organisms and Replicates per Group	Low	There was one pot with 14 plants for each treatment, which is lower than is typical.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	The plants were kept at 20C during the day and 15C at night, but little other details were provided on environmental conditions.
	Metric 17:	Outcome Assessment Methodology	Low	Plants were assessed for signs of toxicity at the end of the test, but this process was not described. No signs of toxicity were observed.
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported. Plants were assessed for signs of toxicity, but this protocol was not described.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Uninformative	Statistical analysis for this portion of the study was not conducted.
	Metric 22:	Reporting of Data	Low	Results were reported in the text only for this portion of the study.
	Metric 23:	Explanation of Unexpected Outcomes	Low	Insufficient information was provided to determine if excessive variability or unexpected outcomes occurred.
Additional Comments:	This portion of the evaluation was conducted in compartments in a glasshouse. One pot was grown in a section that contained glazed strips containing DBP. The other pot was grown in section that contained strips glazed with DIDP. This study received an unacceptable ranking due to the lack of a true negative control and due to lack of statistics. The study authors reported assessing the plants for signs of toxicity. This included mortality, growth, and chlorosis.			

Overall Quality Determination**Uninformative**

Study Citation:	Zhao, H. M., Huang, H. B., Luo, Y. M., Huang, C. Q., Du, H., Xiang, L., Cai, Q. Y., Li, Y. W., Li, H., Mo, C. H., He, Z. (2018). Differences in root physiological and proteomic responses to dibutyl phthalate (DBP) exposure between low- and high-DBP accumulation cultivars of Brassica parachinensis. Journal of Agricultural and Food Chemistry 66(51):13541-13551.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Water; Root uptake			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica parachinensis</i> ; Huaguan; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Cytotoxicity-Oxidative stress (including redox biology)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5043543			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	Chemical was identified by name only	
	Metric 2: Test Substance Source	Low	The source was not reported.	
	Metric 3: Test Substance Purity	Low	Purity or grade of test substance was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations	
	Metric 8: Consistency of Exposure Administration	Low	Only general methods of exposure administration were reported so assessment was difficult to determine	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and exposure frequency were reported and appropriate for the study type	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	A suitable number of treatments over an adequate range of concentrations was reported	
	Metric 12: Testing at or Below Solubility Limit	Uninformative	All test concentrations exceeded the water solubility value of 13 mg/L	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the test plants was not reported	
	Metric 14: Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms	
	Metric 15: Number of Organisms and Replicates per Group	Low	Initial number not reported. 5 roots were pooled per sampling in triplicate	
Domain 5: Outcome Assessment				
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Study Citation:	Zhao, H. M., Huang, H. B., Luo, Y. M., Huang, C. Q., Du, H., Xiang, L., Cai, Q. Y., Li, Y. W., Li, H., Mo, C. H., He, Z. (2018). Differences in root physiological and proteomic responses to dibutyl phthalate (DBP) exposure between low- and high-DBP accumulation cultivars of Brassica parachinensis. Journal of Agricultural and Food Chemistry 66(51):13541-13551.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Water; Root uptake			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica parachinensis</i> ; Huaguan; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Cytotoxicity-Oxidative stress (including redox biology)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5043543			
Domain	Metric	Rating	Comments	
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest	
	Metric 18: Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	Low	Statistical analysis was performed but not described adequately.	
	Metric 22: Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group for every measurement, but results were described in the text, figures and charts	
	Metric 23: Explanation of Unexpected Outcomes	Low	insufficient information was provided to determine if unexpected outcomes occurred	
Additional Comments:	None			
Overall Quality Determination		Uninformative		

Study Citation:	Zhao, H. M., Huang, H. B., Luo, Y. M., Huang, C. Q., Du, H., Xiang, L., Cai, Q. Y., Li, Y. W., Li, H., Mo, C. H., He, Z. (2018). Differences in root physiological and proteomic responses to dibutyl phthalate (DBP) exposure between low- and high-DBP accumulation cultivars of <i>Brassica parachinensis</i> . Journal of Agricultural and Food Chemistry 66(51):13541-13551.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Water; Root uptake			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica parachinensis</i> ; Lvbao; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5043543			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The chemical was identified by name only.
	Metric 2:	Test Substance Source	Low	The source was not reported.
	Metric 3:	Test Substance Purity	Low	Purity or grade of the test substance was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	Low	The biological response of the negative control group was not reported.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.
	Metric 8:	Consistency of Exposure Administration	Low	Only general methods of exposure administration were reported so assessment was difficult to determine.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	A suitable number of treatments over an adequate range of concentrations was reported.
	Metric 12:	Testing at or Below Solubility Limit	Uninformative	All test concentrations exceeded the water solubility value of 13 mg/L.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test plants was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Low	The initial number of organisms was not reported. Five roots were pooled per sampling in triplicate.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported.

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Study Citation:	Zhao, H. M., Huang, H. B., Luo, Y. M., Huang, C. Q., Du, H., Xiang, L., Cai, Q. Y., Li, Y. W., Li, H., Mo, C. H., He, Z. (2018). Differences in root physiological and proteomic responses to dibutyl phthalate (DBP) exposure between low- and high-DBP accumulation cultivars of Brassica parachinensis. Journal of Agricultural and Food Chemistry 66(51):13541-13551.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Water; Root uptake			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica parachinensis</i> ; Lvbao; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5043543			
Domain	Metric	Rating	Comments	
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22:	Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group for every measurement, but results were described in the text.
	Metric 23:	Explanation of Unexpected Outcomes	Low	There was insufficient information provided to determine if unexpected outcomes occurred.
Additional Comments: None				
Overall Quality Determination		Uninformative		

Study Citation:	Zhao, H. M., Huang, H. B., Luo, Y. M., Huang, C. Q., Du, H., Xiang, L., Cai, Q. Y., Li, Y. W., Li, H., Mo, C. H., He, Z. (2018). Differences in root physiological and proteomic responses to dibutyl phthalate (DBP) exposure between low- and high-DBP accumulation cultivars of <i>Brassica parachinensis</i> . Journal of Agricultural and Food Chemistry 66(51):13541-13551.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Water; Root uptake			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica parachinensis</i> ; Lvbao; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Cytotoxicity-Oxidative stress (including redox biology)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5043543			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	Low	The source was not reported.
	Metric 3:	Test Substance Purity	Low	Purity or grade of test substance was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Low	Only general methods of exposure administration were reported so assessment was difficult to determine
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	A suitable number of treatments over an adequate range of concentrations was reported
	Metric 12:	Testing at or Below Solubility Limit	Uninformative	All test concentrations exceeded the water solubility value of 13 mg/L
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test plants was not reported
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Low	Initial number not reported. 5 roots were pooled per sampling in triplicate
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest

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Study Citation:	Zhao, H. M., Huang, H. B., Luo, Y. M., Huang, C. Q., Du, H., Xiang, L., Cai, Q. Y., Li, Y. W., Li, H., Mo, C. H., He, Z. (2018). Differences in root physiological and proteomic responses to dibutyl phthalate (DBP) exposure between low- and high-DBP accumulation cultivars of Brassica parachinensis. Journal of Agricultural and Food Chemistry 66(51):13541-13551.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Water; Root uptake			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica parachinensis</i> ; Lvbao; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Cytotoxicity-Oxidative stress (including redox biology)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5043543			
Domain	Metric	Rating	Comments	
	Metric 18: Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	Low	Statistical analysis was performed but not described adequately.	
	Metric 22: Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group for every measurement, but results were described in the text, figures and charts	
	Metric 23: Explanation of Unexpected Outcomes	Low	insufficient information was provided to determine if unexpected outcomes occurred	
Additional Comments:	None			

Overall Quality Determination**Uninformative**

Study Citation:	Zhao, H. M., Huang, H. B., Luo, Y. M., Huang, C. Q., Du, H., Xiang, L., Cai, Q. Y., Li, Y. W., Li, H., Mo, C. H., He, Z. (2018). Differences in root physiological and proteomic responses to dibutyl phthalate (DBP) exposure between low- and high-DBP accumulation cultivars of <i>Brassica parachinensis</i> . Journal of Agricultural and Food Chemistry 66(51):13541-13551.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Water; Root uptake			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica parachinensis</i> ; Huaguan; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5043543			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	Low	The source was not reported.
	Metric 3:	Test Substance Purity	Low	Purity or grade of test substance was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	Low	The biological response of the negative control group was not reported
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Low	Only general methods of exposure administration were reported so assessment was difficult to determine
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	A suitable number of treatments over an adequate range of concentrations was reported
	Metric 12:	Testing at or Below Solubility Limit	Uninformative	All test concentrations exceeded the water solubility value of 13 mg/L
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test plants was not reported
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Low	Initial number not reported. 5 roots were pooled per sampling in triplicate
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported

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Study Citation:	Zhao, H. M., Huang, H. B., Luo, Y. M., Huang, C. Q., Du, H., Xiang, L., Cai, Q. Y., Li, Y. W., Li, H., Mo, C. H., He, Z. (2018). Differences in root physiological and proteomic responses to dibutyl phthalate (DBP) exposure between low- and high-DBP accumulation cultivars of Brassica parachinensis. Journal of Agricultural and Food Chemistry 66(51):13541-13551.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Water; Root uptake			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica parachinensis</i> ; Huaguan; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5043543			
Domain	Metric	Rating	Comments	
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	Low	Statistical analysis was performed but not described adequately.	
	Metric 22: Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group for every measurement, but results were described in the text	
	Metric 23: Explanation of Unexpected Outcomes	Low	insufficient information was provided to determine if unexpected outcomes occurred	
Additional Comments:	This paper was judged unacceptable because all exposures were above the solubility of DBP in water, 13 mg/L.			

Overall Quality Determination**Uninformative**

Study Citation:	Zhao, H. M., Du, H., Xiang, L., Li, Y. W., Li, H., Cai, Q. Y., Mo, C. H., Cao, G., Wong, M. H. (2016). Physiological differences in response to di-n-butyl phthalate (DBP) exposure between low- and high-DBP accumulating cultivars of Chinese flowering cabbage (<i>Brassica parachinensis</i> L.). Environmental Pollution 208(Pt B):840-849.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica parachinensis</i> L.; Lvbao70 cultivar; Embryo			
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)-Photosynthesis			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3070947			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The DBP was identified by name only.	
	Metric 2: Test Substance Source	High	The source of the DBP was reported to be Aladdin Chemistry Co. Ltd. in China, and DBP was verified by GC/MS.	
	Metric 3: Test Substance Purity	High	The purity was reported to be 98.7%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using soil that was not spiked with DBP as a negative control.	
	Metric 5: Negative Control Response	High	The negative control response was reported in Table 3 and in Figures 2-4. The control response was adequate for the outcomes of interest.	
	Metric 6: Randomized Allocation	Low	It was not reported how the seeds were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	Soil was spiked with the appropriate amount of DBP dissolved in acetone. The acetone was allowed to evaporate and the soil was aged for 2 weeks before the start of the study. Exposure was conducted in ceramic pots.	
	Metric 8: Consistency of Exposure Administration	High	All exposures were for 45d in ceramic pots that were 20cm in diameter and 14cm in height in a greenhouse with natural lighting conditions. seedlings were thinned to 5 seedlings per pot at 15d and watered daily.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if the test concentrations were measured at any point in the study.	
	Metric 10: Exposure Duration and Frequency	High	The study duration was reported to be for 45 days. This was adequate for the outcomes of interest.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Low	There were only 2 exposure groups, which is quite a bit lower than normal. This was still adequate to see a response.	
	Metric 12: Testing at or Below Solubility Limit	N/A	The exposure was via soil.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the B. parachinensis L. was not reported.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the organisms were acclimated in any way.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 5 seedlings per test chamber. Each exposure was performed in triplicate.	

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Study Citation:	Zhao, H. M., Du, H., Xiang, L., Li, Y. W., Li, H., Cai, Q. Y., Mo, C. H., Cao, G., Wong, M. H. (2016). Physiological differences in response to di-n-butyl phthalate (DBP) exposure between low- and high-DBP accumulating cultivars of Chinese flowering cabbage (<i>Brassica parachinensis</i> L.). Environmental Pollution 208(Pt B):840-849.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica parachinensis</i> L.; Lvbao70 cultivar; Embryo			
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)-Photosynthesis			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3070947			
Domain	Metric	Rating	Comments	
Domain 5: Outcome Assessment				
Metric 16:	Adequacy of Test Conditions	Low	It was reported that natural light was used in the greenhouse, but it was not reported what this was. Temperature was reported to be 25-32C. It was unclear if plants were fertilized more than once. Relative humidity was not reported.	
Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—changes in chlorophyll, and changes in SOD, CAT, GST, and MDA.	
Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups.	
Domain 6: Confounding / Variable Control				
Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.	
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
Metric 21:	Statistical Methods	High	Statistical methods were reported in the "Statistical Analysis" section and were appropriate for the study.	
Metric 22:	Reporting of Data	High	Data for the exposure response and the control response was reported in Table 3 and in Figures 2-4.	
Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in the figures and tables.	
Additional Comments:	This portion of the evaluation was on the effect of DBP on chlorophyll a, SOD, CAT, GST, and MDA the Lvbao70 cultivar of B parachinensis L. The mechanistic outcomes of photosynthesis and oxidative stress were chosen as the outcomes of interest.			

Overall Quality Determination**Medium**

Study Citation:	Zhao, H. M., Du, H., Xiang, L., Li, Y. W., Li, H., Cai, Q. Y., Mo, C. H., Cao, G., Wong, M. H. (2016). Physiological differences in response to di-n-butyl phthalate (DBP) exposure between low- and high-DBP accumulating cultivars of Chinese flowering cabbage (<i>Brassica parachinensis</i> L.). <i>Environmental Pollution</i> 208(Pt B):840-849.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica parachinensis</i> L.; Huaguan cultivar; Embryo			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3070947			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The DBP was identified by name only.	
	Metric 2: Test Substance Source	High	The source of the DBP was reported to be Aladdin Chemistry Co. Ltd. in China, and DBP was verified by GC/MS.	
	Metric 3: Test Substance Purity	High	The purity was reported to be 98.7%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using soil that was not spiked with DBP as a negative control.	
	Metric 5: Negative Control Response	High	The negative control response was reported in Figure S1a. It was appropriate for the outcomes of interest.	
	Metric 6: Randomized Allocation	Low	It was not reported how the seeds were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	Soil was spiked with the appropriate amount of DBP dissolved in acetone. The acetone was allowed to evaporate and the soil was aged for 2 weeks before the start of the study. Exposure was conducted in ceramic pots.	
	Metric 8: Consistency of Exposure Administration	High	All exposures were for 45d in ceramic pots that were 20cm in diameter and 14cm in height in a greenhouse with natural lighting conditions. seedlings were thinned to 5 seedlings per pot at 15d and watered daily.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if the test concentrations were measured at any point in the study.	
	Metric 10: Exposure Duration and Frequency	High	The study duration was reported to be for 45 days. This was adequate for the outcomes of interest.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Low	There were only 2 exposure groups, which is quite a bit lower than normal. This was still adequate to see a response.	
	Metric 12: Testing at or Below Solubility Limit	N/A	The exposure was via soil.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the <i>B. parachinensis</i> L. was not reported.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the organisms were acclimated in any way.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 5 seedlings per test chamber. Each exposure was performed in triplicate.	
Domain 5: Outcome Assessment				
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Study Citation:	Zhao, H. M., Du, H., Xiang, L., Li, Y. W., Li, H., Cai, Q. Y., Mo, C. H., Cao, G., Wong, M. H. (2016). Physiological differences in response to di-n-butyl phthalate (DBP) exposure between low- and high-DBP accumulating cultivars of Chinese flowering cabbage (<i>Brassica parachinensis</i> L.). <i>Environmental Pollution</i> 208(Pt B):840-849.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica parachinensis</i> L.; Huaguan cultivar; Embryo			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3070947			
Domain	Metric	Rating	Comments	
	Metric 16:	Adequacy of Test Conditions	Low	It was reported that natural light was used in the greenhouse, but it was not reported what this was. Temperature was reported to be 25-32C. It was unclear if plants were fertilized more than once. Relative humidity was not reported.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest–DBP accumulation in the roots and shoots.
	Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups–DBP accumulation was determined by GC-MS.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were reported in the “Statistical Analysis” section and were appropriate for the study.
	Metric 22:	Reporting of Data	High	Data for the exposure response and the control response was reported in the supplemental figure 1a and was appropriate for the outcomes of interest.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in the figure.
Additional Comments:	This potion of the evaluation was on the accumulation of DBP in the shoots and roots of <i>B. parachinensis</i> cultivar Huaguan. ADME was selected as the outcome of interest.			
Overall Quality Determination		Medium		

Study Citation:	Zhao, H. M., Du, H., Xiang, L., Li, Y. W., Li, H., Cai, Q. Y., Mo, C. H., Cao, G., Wong, M. H. (2016). Physiological differences in response to di-n-butyl phthalate (DBP) exposure between low- and high-DBP accumulating cultivars of Chinese flowering cabbage (<i>Brassica parachinensis</i> L.). <i>Environmental Pollution</i> 208(Pt B):840-849.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica parachinensis</i> L.; Lvbao70 cultivar; Embryo			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3070947			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The DBP was identified by name only.	
	Metric 2: Test Substance Source	High	The source of the DBP was reported to be Aladdin Chemistry Co. Ltd. in China, and DBP was verified by GC/MS.	
	Metric 3: Test Substance Purity	High	The purity was reported to be 98.7%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using soil that was not spiked with DBP as a negative control.	
	Metric 5: Negative Control Response	High	The negative control response was reported in Figure S1b and in Figure 1. It was appropriate for the outcomes of interest.	
	Metric 6: Randomized Allocation	Low	It was not reported how the seeds were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	Soil was spiked with the appropriate amount of DBP dissolved in acetone. The acetone was allowed to evaporate and the soil was aged for 2 weeks before the start of the study. Exposure was conducted in ceramic pots.	
	Metric 8: Consistency of Exposure Administration	High	All exposures were for 45d in ceramic pots that were 20cm in diameter and 14cm in height in a greenhouse with natural lighting conditions. seedlings were thinned to 5 seedlings per pot at 15d and watered daily.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if the test concentrations were measured at any point in the study.	
	Metric 10: Exposure Duration and Frequency	High	The study duration was reported to be for 45 days. This was adequate for the outcomes of interest.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Low	There were only 2 exposure groups, which is quite a bit lower than normal. This was still adequate to see a response.	
	Metric 12: Testing at or Below Solubility Limit	N/A	The exposure was via soil.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the <i>B. parachinensis</i> L. was not reported.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the organisms were acclimated in any way.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 5 seedlings per test chamber. Each exposure was performed in triplicate.	
Domain 5: Outcome Assessment				
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Study Citation:	Zhao, H. M., Du, H., Xiang, L., Li, Y. W., Li, H., Cai, Q. Y., Mo, C. H., Cao, G., Wong, M. H. (2016). Physiological differences in response to di-n-butyl phthalate (DBP) exposure between low- and high-DBP accumulating cultivars of Chinese flowering cabbage (<i>Brassica parachinensis</i> L.). Environmental Pollution 208(Pt B):840-849.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica parachinensis</i> L.; Lvbao70 cultivar; Embryo			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3070947			
Domain	Metric	Rating	Comments	
	Metric 16:	Adequacy of Test Conditions	Low	It was reported that natural light was used in the greenhouse, but it was not reported what this was. Temperature was reported to be 25-32C. It was unclear if plants were fertilized more than once. Relative humidity was not reported.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—plant biomass and changes in structure.
	Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups—at the end of the study, plants were divided into roots and shoots, and fresh weights were recorded. Plant tissues were also examined via TEM to assess leaf ultrastructure.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were reported in the "Statistical Analysis" section and were appropriate for the study.
	Metric 22:	Reporting of Data	High	Data for the exposure response and the control response was reported in the supplemental figure 1b and was appropriate for the outcomes of interest. Leaf structure TEM was provided in Figure 1 for both treatments and controls.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in the figure.
Additional Comments:	This portion of the evaluation was on the effect of DBP on plant biomass and leaf structure of <i>B. parachinensis</i> L cultivar Lvbao70. Development and growth was selected as the outcome of interest.			

Overall Quality Determination**Medium**

Study Citation:	Zhao, H. M., Du, H., Xiang, L., Li, Y. W., Li, H., Cai, Q. Y., Mo, C. H., Cao, G., Wong, M. H. (2016). Physiological differences in response to di-n-butyl phthalate (DBP) exposure between low- and high-DBP accumulating cultivars of Chinese flowering cabbage (<i>Brassica parachinensis</i> L.). Environmental Pollution 208(Pt B):840-849.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica parachinensis</i> L.; Huaguan cultivar; Embryo			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3070947			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The DBP was identified by name only.	
	Metric 2: Test Substance Source	High	The source of the DBP was reported to be Aladdin Chemistry Co. Ltd. in China, and DBP was verified by GC/MS.	
	Metric 3: Test Substance Purity	High	The purity was reported to be 98.7%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using soil that was not spiked with DBP as a negative control.	
	Metric 5: Negative Control Response	High	The negative control response was reported in Figure S1b and in Figure 1. It was appropriate for the outcomes of interest.	
	Metric 6: Randomized Allocation	Low	It was not reported how the seeds were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	Soil was spiked with the appropriate amount of DBP dissolved in acetone. The acetone was allowed to evaporate and the soil was aged for 2 weeks before the start of the study. Exposure was conducted in ceramic pots.	
	Metric 8: Consistency of Exposure Administration	High	All exposures were for 45d in ceramic pots that were 20cm in diameter and 14cm in height in a greenhouse with natural lighting conditions. seedlings were thinned to 5 seedlings per pot at 15d and watered daily.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if the test concentrations were measured at any point in the study.	
	Metric 10: Exposure Duration and Frequency	High	The study duration was reported to be for 45 days. This was adequate for the outcomes of interest.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Low	There were only 2 exposure groups, which is quite a bit lower than normal. This was still adequate to see a response.	
	Metric 12: Testing at or Below Solubility Limit	N/A	The exposure was via soil.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the <i>B. parachinensis</i> L. was not reported.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the organisms were acclimated in any way.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 5 seedlings per test chamber. Each exposure was performed in triplicate.	
Domain 5: Outcome Assessment				
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Study Citation:	Zhao, H. M., Du, H., Xiang, L., Li, Y. W., Li, H., Cai, Q. Y., Mo, C. H., Cao, G., Wong, M. H. (2016). Physiological differences in response to di-n-butyl phthalate (DBP) exposure between low- and high-DBP accumulating cultivars of Chinese flowering cabbage (<i>Brassica parachinensis</i> L.). Environmental Pollution 208(Pt B):840-849.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica parachinensis</i> L.; Huaguan cultivar; Embryo			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3070947			
Domain	Metric	Rating	Comments	
	Metric 16: Adequacy of Test Conditions	Low	It was reported that natural light was used in the greenhouse, but it was not reported what this was. Temperature was reported to be 25-32C. It was unclear if plants were fertilized more than once. Relative humidity was not reported.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—plant biomass and changes in structure.	
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups—at the end of the study, plants were divided into roots and shoots, and fresh weights were recorded. Plant tissues were also examined via TEM to assess leaf ultrastructure.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical methods were reported in the "Statistical Analysis" section and were appropriate for the study.	
	Metric 22: Reporting of Data	High	Data for the exposure response and the control response was reported in the supplemental figure 1b and was appropriate for the outcomes of interest. Leaf structure TEM was provided in Figure 1 for both treatments and controls.	
	Metric 23: Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in the figure.	
Additional Comments:	This portion of the evaluation was on the effect of DBP on plant biomass and leaf structure of <i>B. parachinensis</i> L cultivar Huaguan. Development and growth was selected as the outcome of interest.			

Overall Quality Determination**Medium**

Study Citation:	Zhao, H. M., Du, H., Xiang, L., Li, Y. W., Li, H., Cai, Q. Y., Mo, C. H., Cao, G., Wong, M. H. (2016). Physiological differences in response to di-n-butyl phthalate (DBP) exposure between low- and high-DBP accumulating cultivars of Chinese flowering cabbage (<i>Brassica parachinensis</i> L.). Environmental Pollution 208(Pt B):840-849.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica parachinensis</i> L.; Lvbao70 cultivar; Embryo			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3070947			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The DBP was identified by name only.
	Metric 2:	Test Substance Source	High	The source of the DBP was reported to be Aladdin Chemistry Co. Ltd. in China, and DBP was verified by GC/MS.
	Metric 3:	Test Substance Purity	High	The purity was reported to be 98.7%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using soil that was not spiked with DBP as a negative control.
	Metric 5:	Negative Control Response	High	The negative control response was reported in Figure S1a. It was appropriate for the outcomes of interest.
	Metric 6:	Randomized Allocation	Low	It was not reported how the seeds were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	Soil was spiked with the appropriate amount of DBP dissolved in acetone. The acetone was allowed to evaporate and the soil was aged for 2 weeks before the start of the study. Exposure was conducted in ceramic pots.
	Metric 8:	Consistency of Exposure Administration	High	All exposures were for 45d in ceramic pots that were 20cm in diameter and 14cm in height in a greenhouse with natural lighting conditions. seedlings were thinned to 5 seedlings per pot at 15d and watered daily.
	Metric 9:	Measurement of Test Substance Concentration	Low	It was not reported if the test concentrations were measured at any point in the study.
	Metric 10:	Exposure Duration and Frequency	High	The study duration was reported to be for 45 days. This was adequate for the outcomes of interest.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	There were only 2 exposure groups, which is lower than normal. This was still adequate to see a response.
	Metric 12:	Testing at or Below Solubility Limit	N/A	The exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the B. parachinensis L. was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the organisms were acclimated in any way.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	There were 5 seedlings per test chamber. Each exposure was performed in triplicate.
Domain 5: Outcome Assessment				

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Study Citation:	Zhao, H. M., Du, H., Xiang, L., Li, Y. W., Li, H., Cai, Q. Y., Mo, C. H., Cao, G., Wong, M. H. (2016). Physiological differences in response to di-n-butyl phthalate (DBP) exposure between low- and high-DBP accumulating cultivars of Chinese flowering cabbage (<i>Brassica parachinensis</i> L.). <i>Environmental Pollution</i> 208(Pt B):840-849.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica parachinensis</i> L.; Lvbao70 cultivar; Embryo			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3070947			
Domain	Metric	Rating	Comments	
	Metric 16:	Adequacy of Test Conditions	Low	It was reported that natural light was used in the greenhouse, but it was not reported what this was. Temperature was reported to be 25-32C. It was unclear if plants were fertilized more than once. Relative humidity was not reported.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest–DBP accumulation in the roots and shoots.
	Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups–DBP accumulation was determined by GC-MS.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were reported in the “Statistical Analysis” section and were appropriate for the study.
	Metric 22:	Reporting of Data	High	Data for the exposure response and the control response was reported in the supplemental figure 1a and was appropriate for the outcomes of interest.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in the figure.
Additional Comments:	This potion of the evaluation was on the accumulation of DBP in the shoots and roots of <i>B. parachinensis</i> cultivar Lvbao70. ADME was selected as the outcome of interest.			
Overall Quality Determination		Medium		

Study Citation:	Zhao, H. M., Du, H., Xiang, L., Li, Y. W., Li, H., Cai, Q. Y., Mo, C. H., Cao, G., Wong, M. H. (2016). Physiological differences in response to di-n-butyl phthalate (DBP) exposure between low- and high-DBP accumulating cultivars of Chinese flowering cabbage (<i>Brassica parachinensis</i> L.). <i>Environmental Pollution</i> 208(Pt B):840-849.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica parachinensis</i> L.; Huaguan cultivar; Embryo			
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)-Photosynthesis			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3070947			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The DBP was identified by name only.
	Metric 2:	Test Substance Source	High	The source of the DBP was reported to be Aladdin Chemistry Co. Ltd. in China, and DBP was verified by GC/MS.
	Metric 3:	Test Substance Purity	High	The purity was reported to be 98.7%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using soil that was not spiked with DBP as a negative control.
	Metric 5:	Negative Control Response	High	The negative control response was reported in Table 3 and in Figures 2-4. The control response was adequate for the outcomes of interest.
	Metric 6:	Randomized Allocation	Low	It was not reported how the seeds were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	Soil was spiked with the appropriate amount of DBP dissolved in acetone. The acetone was allowed to evaporate, and the soil was aged for two weeks before the start of the study. Exposure was conducted in ceramic pots.
	Metric 8:	Consistency of Exposure Administration	High	All exposures were for 45d in ceramic pots that were 20cm in diameter and 14cm in height in a greenhouse with natural lighting conditions. Seedlings were thinned to five per pot at 15d and watered daily.
	Metric 9:	Measurement of Test Substance Concentration	Low	It was not reported if the test concentrations were measured at any point in the study.
	Metric 10:	Exposure Duration and Frequency	High	The study duration was reported to be for 45 days. This was adequate for the outcomes of interest.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	There were only two exposure groups, which is quite a bit lower than normal. This was still adequate to see a response.
	Metric 12:	Testing at or Below Solubility Limit	N/A	The exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the <i>B. parachinensis</i> L. was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the organisms were acclimated in any way.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	There were five seedlings per test chamber. Each exposure was performed in triplicate.
Domain 5: Outcome Assessment				
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Study Citation:	Zhao, H. M., Du, H., Xiang, L., Li, Y. W., Li, H., Cai, Q. Y., Mo, C. H., Cao, G., Wong, M. H. (2016). Physiological differences in response to di-n-butyl phthalate (DBP) exposure between low- and high-DBP accumulating cultivars of Chinese flowering cabbage (<i>Brassica parachinensis</i> L.). <i>Environmental Pollution</i> 208(Pt B):840-849.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica parachinensis</i> L.; Huaguan cultivar; Embryo			
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)-Photosynthesis			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3070947			
Domain		Metric	Rating	Comments
	Metric 16:	Adequacy of Test Conditions	Low	It was reported that natural light was used in the greenhouse, but it was not reported what this was. Temperature was reported to be 25-32C. It was unclear if plants were fertilized more than once. Relative humidity was not reported.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—changes in chlorophyll, and changes in SOD, CAT, GST, and MDA.
	Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were reported in the "Statistical Analysis" section and were appropriate for the study.
	Metric 22:	Reporting of Data	High	Data for the exposure response and the control response was reported in Table 3 and in Figures 2-4.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in the figures and tables.
Additional Comments:	This portion of the evaluation was on the effect of DBP on chlorophyll a, SOD, CAT, GST, and MDA the Huaguan cultivar of B parachinensis L. The mechanistic outcomes of photosynthesis and oxidative stress were chosen as the outcomes of interest.			

Overall Quality Determination**Medium**

Study Citation:	Liao, C. S., Yen, J. H., Wang, Y. S. (2009). Growth inhibition in Chinese cabbage (<i>Brassica rapa</i> var. <i>chinensis</i>) growth exposed to di-n-butyl phthalate. Journal of Hazardous Materials 163(2-3):625-631.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica rapa</i> ; var. <i>chinensis</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1296241			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	DBP was identified by CAS number.	
	Metric 2: Test Substance Source	Low	The source of the DBP was reported to be Riedel-deHaen Co. in Germany, but it was not reported if it was analytically verified.	
	Metric 3: Test Substance Purity	High	The purity was reported to be 98.7%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate solvent control.	
	Metric 5: Negative Control Response	High	The negative control response was adequate and was reported in Tables 1 and 3.	
	Metric 6: Randomized Allocation	Low	It was not reported how the cabbage was allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	DBP was added to acetone at a concentration of 100g/L to create the stock solution. It was not reported how the test concentrations were prepared. It was also not reported how often new test solution was added to the system. The type of experimental vessel (plastic or glass) used was not reported.	
	Metric 8: Consistency of Exposure Administration	Medium	All test exposures were conducted in 3L pots with 2.5L of solution at the same temperature and relative humidity. Photoperiod was 16L:8D with 25C during the day and 20C at night. It was not reported how often the test solution was replaced.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if the DBP was measured at any point in the study.	
	Metric 10: Exposure Duration and Frequency	High	The duration of the study was reported to be 42d. This was adequate to observe a response.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	Study authors reported four study groups plus a solvent control. This was adequate for the outcomes of interest.	
	Metric 12: Testing at or Below Solubility Limit	High	Study authors reported the use of a vehicle solvent. The solvent control response was appropriate.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the cabbage seeds was not reported.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the plants were acclimated in any way to test conditions.	
	Metric 15: Number of Organisms and Replicates per Group	Low	The number of test organisms in each replicate was not reported. It was reported that tests were carried out in triplicate.	

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Study Citation:	Liao, C. S., Yen, J. H., Wang, Y. S. (2009). Growth inhibition in Chinese cabbage (<i>Brassica rapa</i> var. <i>chinensis</i>) growth exposed to di-n-butyl phthalate. <i>Journal of Hazardous Materials</i> 163(2-3):625-631.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica rapa</i> ; var. <i>chinensis</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	ADME (biotransformation)
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	1296241

Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
Metric 16:	Adequacy of Test Conditions	High	Authors reported growing the organisms in a solution adapted from Hoagland's solution. There was 2.5L of media in each pot. Organisms were grown at 25C during the day and 20C at night with a 16L:8D photoperiod. Relative humidity was reported to be 70-95%.
Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—accumulation of DBP in plant tissue.
Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. The assessment process was discussed in detail in section 2.4.
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. It was not reported if the plants were acclimated.
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	High	T-tests were used to analyze data.
Metric 22:	Reporting of Data	High	All accumulation data was reported in Table 1 along with the control response. Data was appropriate for the outcome of interest. Table 3 also had a summary of the data.
Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in Table 1.
Additional Comments:	This portion of the evaluation was on the accumulation of DBP in Chinese cabbage after a 42-day exposure at various concentrations. ADME was selected as the outcome of interest.		

Overall Quality Determination**Medium**

Study Citation:	Liao, C. S., Yen, J. H., Wang, Y. S. (2009). Growth inhibition in Chinese cabbage (<i>Brassica rapa</i> var. <i>chinensis</i>) growth exposed to di-n-butyl phthalate. Journal of Hazardous Materials 163(2-3):625-631.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica rapa</i> ; var. <i>chinensis</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Photosynthesis			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1296241			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	DBP was identified by CAS number.	
	Metric 2: Test Substance Source	Low	The source of the DBP was reported to be Riedel-deHaen Co. in Germany, but it was not reported if it was analytically verified.	
	Metric 3: Test Substance Purity	High	The purity was reported to be 98.7%	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate solvent control.	
	Metric 5: Negative Control Response	High	The negative control response was adequate and was reported in Table 2 for chlorophyll and in Figures 3 and 4 for the proteomic portion of the study.	
	Metric 6: Randomized Allocation	Low	It was not reported how the cabbage was allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	DBP was added to acetone at a concentration of 100g/L to create the stock solution. It was not reported how the test concentrations were prepared. It was also not reported how often new test solution was added to the system. The type of experimental vessel (glass or plastic) used was not reported.	
	Metric 8: Consistency of Exposure Administration	Medium	All test exposures were conducted in 3L pots with 2.5L of solution at the same temperature and relative humidity. Photoperiod was 16L:8D with 25C during the day and 20C at night. It was not reported how often the test solution was replaced.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if the DBP was measured at any point in the study.	
	Metric 10: Exposure Duration and Frequency	High	The duration of the study was reported to be 42d. This was adequate to observe a response.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	Study authors reported 4 study groups plus a solvent control. This was adequate for the outcomes of interest.	
	Metric 12: Testing at or Below Solubility Limit	High	Study authors reported the use of a vehicle solvent. The solvent control response was appropriate.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the cabbage seeds was not reported.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the plants were acclimated in any way to test conditions.	
	Metric 15: Number of Organisms and Replicates per Group	Low	The number of test organisms in each replicate was not reported. It was reported that tests were carried out in triplicate.	
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Study Citation:	Liao, C. S., Yen, J. H., Wang, Y. S. (2009). Growth inhibition in Chinese cabbage (<i>Brassica rapa</i> var. <i>chinensis</i>) growth exposed to di-n-butyl phthalate. Journal of Hazardous Materials 163(2-3):625-631.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica rapa</i> ; var. <i>chinensis</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Photosynthesis			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1296241			
Domain	Metric	Rating	Comments	
Domain 5: Outcome Assessment				
Metric 16:	Adequacy of Test Conditions	High	Authors reported growing the organisms in a solution adapted from Hoagland’s solution. There was 2.5L of media in each pot. Organisms were grown at 25C during the day and 20C at night with a 16L:8D photoperiod. Relative humidity was reported to be 70-95%.	
Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest–chlorophyll a and b concentration and protein expression.	
Metric 18:	Consistency of Outcome Assessment	High	Details concerning the proteomics assessment portion of the study were reported in detail in section 2.5.	
Domain 6: Confounding / Variable Control				
Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. It was not reported if the plants were acclimated.	
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
Metric 21:	Statistical Methods	High	T-tests were used to analyze data.	
Metric 22:	Reporting of Data	High	Data was reported in Table 2 for chlorophyll concentrations and in Figures 3 and 4 and Table 4 for protein expression. Control data was reported for both mechanistic outcomes.	
Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in Table 2.	
Additional Comments:	This portion of the evaluation was on the effect of DBP on chlorophyll a and b concentration and on protein expression in Chinese cabbage. The photosynthesis and biomarker mechanistic outcomes were chosen as the outcomes of interest.			

Overall Quality Determination**Medium**

Study Citation:	Liao, C. S., Yen, J. H., Wang, Y. S. (2009). Growth inhibition in Chinese cabbage (<i>Brassica rapa</i> var. <i>chinensis</i>) growth exposed to di-n-butyl phthalate. Journal of Hazardous Materials 163(2-3):625-631.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica rapa</i> ; var. <i>chinensis</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1296241			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	DBP was identified by CAS number.	
	Metric 2: Test Substance Source	Low	The source of the DBP was reported to be Riedel-deHaen Co. in Germany, but it was not reported if it was analytically verified.	
	Metric 3: Test Substance Purity	High	The purity was reported to be 98.7%	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate solvent control.	
	Metric 5: Negative Control Response	High	The negative control response was adequate and was reported in Table 3 in terms of biomass.	
	Metric 6: Randomized Allocation	Low	It was not reported how the cabbage was allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	DBP was added to acetone at a concentration of 100g/L to create the stock solution. It was not reported how the test concentrations were prepared. It was also not reported how often new test solution was added to the system. The type of experimental vessel (plastic or glass) used is not reported.	
	Metric 8: Consistency of Exposure Administration	Medium	All test exposures were conducted in 3L pots with 2.5L of solution at the same temperature and relative humidity. Photoperiod was 16L:8D with 25C during the day and 20C at night. It was not reported how often the test solution was replaced.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if the DBP was measured at any point in the study.	
	Metric 10: Exposure Duration and Frequency	High	The duration of the study was reported to be 42d. This was adequate to observe a response.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	Study authors reported 4 study groups plus a solvent control. This was adequate for the outcomes of interest.	
	Metric 12: Testing at or Below Solubility Limit	High	Study authors reported the use of a vehicle solvent. The solvent control response was appropriate.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the cabbage seeds was not reported.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the plants were acclimated in any way to test conditions.	
	Metric 15: Number of Organisms and Replicates per Group	Low	The number of test organisms in each replicate was not reported. It was reported that tests were carried out in triplicate.	
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Study Citation:	Liao, C. S., Yen, J. H., Wang, Y. S. (2009). Growth inhibition in Chinese cabbage (<i>Brassica rapa</i> var. <i>chinensis</i>) growth exposed to di-n-butyl phthalate. <i>Journal of Hazardous Materials</i> 163(2-3):625-631.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica rapa</i> ; var. <i>chinensis</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1296241			
Domain	Metric	Rating	Comments	
Domain 5: Outcome Assessment				
Metric 16:	Adequacy of Test Conditions	High	Authors reported growing the organisms in a solution adapted from Hoagland’s solution. There was 2.5L of media in each pot. Organisms were grown at 25C during the day and 20C at night with a 16L:8D photoperiod. Relative humidity was reported to be 70-95%.	
Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—development and growth of the plants in terms of biomass and in terms of physical changes in leaf color and etiolation.	
Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the evaluation of the plant development, morphology, and biomass were limited.	
Domain 6: Confounding / Variable Control				
Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. It was not reported if the plants were acclimated.	
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
Metric 21:	Statistical Methods	High	T-tests were used to analyze data.	
Metric 22:	Reporting of Data	High	Physiological differences were reported in Figure 2 for both treatment and control plants. Biomass for all exposure levels and for the control was reported in Table 3.	
Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in Table 3.	
Additional Comments:	This portion of the evaluation was on the effect of DBP on plant development and growth over 42 days. Leaf size and coloring were monitored, and biomass was taken. Development and growth was chosen as the outcome of interest.			

Overall Quality Determination**Medium**

Study Citation:	Liao, C. S., Yen, J. H., Wang, Y. S. (2006). Effects of endocrine disruptor di-n-butyl phthalate on the growth of Bok choy (<i>Brassica rapa</i> subsp. <i>chinensis</i>). Chemosphere 65(10):1715-1722.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Water; Root uptake			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica rapa</i> ; subsp. <i>chinensis</i> ; Juvenile			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Oxidative stress (including redox biology)-Photosynthesis			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1298079			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The DBP was identified by CAS number.
	Metric 2:	Test Substance Source	High	The source of the DBP was reported to be Riedel-deHaen Co. in Germany. The DBP was analytically verified during the residual DBP analysis part of the study, by GC/MS.
	Metric 3:	Test Substance Purity	High	
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Study authors reported the use of a concurrent negative control. However, they did not specify if a solvent control was used. Acetone was used as a vehicle solvent, so a solvent control would be necessary.
	Metric 5:	Negative Control Response	High	The biological response of the negative control was adequate and was reported in Tables 2 and 3 for photosynthesis and in Figure 4 for the proteomics portion of the study.
	Metric 6:	Randomized Allocation	Low	It was not reported how the Bok choy was allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	Study authors reported dissolving DBP in acetone at a concentration of 100g/L, but they did not elaborate on how the test concentrations were prepared. It was also not reported how often new test solution was added to the system.
	Metric 8:	Consistency of Exposure Administration	Medium	All exposures were conducted in 2.5L of test solution at the same temperature and relative humidity. It was not reported how often the test solution was renewed, which creates doubt regarding the consistency.
	Metric 9:	Measurement of Test Substance Concentration	Low	Study authors did not report if the DBP was measured at any point in the study.
	Metric 10:	Exposure Duration and Frequency	High	The study duration was reported to be 42 days and was appropriate for the outcomes of interest.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	There were five concentrations plus a control. Spacing was adequate to see a response.
	Metric 12:	Testing at or Below Solubility Limit	Medium	Study authors reported using a vehicle solvent, but they did not report if the negative control was a solvent control, so it cannot be determined if the solvent concentration was appropriate.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the Bok choy seeds was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the organisms were acclimated to test conditions.

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Study Citation:	Liao, C. S., Yen, J. H., Wang, Y. S. (2006). Effects of endocrine disruptor di-n-butyl phthalate on the growth of Bok choy (<i>Brassica rapa</i> subsp. <i>chinensis</i>). <i>Chemosphere</i> 65(10):1715-1722.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Water; Root uptake			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica rapa</i> ; subsp. <i>chinensis</i> ; Juvenile			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Oxidative stress (including redox biology)-Photosynthesis			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1298079			
Domain	Metric	Rating	Comments	
	Metric 15: Number of Organisms and Replicates per Group	Low	It was reported that the tests were completed in triplicate, but the number of plants per treatment was low: four plants each.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Organisms were kept at 25C during the day and 20C at night with a 16L:8D photoperiod. Relative humidity was at 70-95%. A modification of Hoagland’s medium was used in the hydroponic growth chambers.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest–chlorophyll concentration and chloroplast morphology and protein expression.	
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment were reported for chlorophyll concentration and chloroplast morphology in sections 3.2 and 3.1.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. It was not reported if the plants were acclimated to test conditions.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	Low	Study authors did not report conducting statistical analysis, but independent analysis may be conducted from the data provided.	
	Metric 22: Reporting of Data	High	Data for control response and exposure related findings was adequate and can be found in Figure 2 and Tables 2 and 3 for photosynthesis and in Figure 4 for the protein expression.	
	Metric 23: Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in the tables.	
Additional Comments:	This portion of the evaluation was on the effect of DBP on Bok choy chlorophyll concentration and chloroplast morphology as well protein expression.			
Overall Quality Determination		Medium		

Study Citation:	Liao, C. S., Yen, J. H., Wang, Y. S. (2006). Effects of endocrine disruptor di-n-butyl phthalate on the growth of Bok choy (<i>Brassica rapa</i> subsp. <i>chinensis</i>). <i>Chemosphere</i> 65(10):1715-1722.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Water; Root uptake			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica rapa</i> ; subsp. <i>chinensis</i> ; Juvenile			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1298079			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The DBP was identified by CAS number.	
	Metric 2: Test Substance Source	High	The source of the DBP was reported to be Riedel-deHaen Co. in Germany. The DBP was analytically verified during the residual DBP analysis part of the study, by GC/MS.	
	Metric 3: Test Substance Purity	High	The purity was reported to be 98.7%	
Domain 2: Test Design				
	Metric 4: Negative Controls	Low	Study authors reported the use of a concurrent negative control. However, they did not specify if a solvent control was used. Acetone was used as a vehicle solvent, so a solvent control would be necessary.	
	Metric 5: Negative Control Response	High	The biological response of the negative control was adequate and was reported in Table 3 and Figure 1.	
	Metric 6: Randomized Allocation	Low	It was not reported how the Bok choy was allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	Study authors reported dissolving DBP in acetone at a concentration of 100g/L, but they did not elaborate on how the test concentrations were prepared. It was also not reported how often new test solution was added to the system.	
	Metric 8: Consistency of Exposure Administration	Medium	All exposures were conducted in 2.5L of test solution at the same temperature and relative humidity. It was not reported how often the test solution was renewed, which creates doubt regarding the consistency.	
	Metric 9: Measurement of Test Substance Concentration	Low	Study authors did not report if the DBP was measured at any point in the study.	
	Metric 10: Exposure Duration and Frequency	High	The study duration was reported to be 42 days and was appropriate for the outcomes of interest.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	There were 5 concentrations plus a control. Spacing was adequate to see a response.	
	Metric 12: Testing at or Below Solubility Limit	Medium	Study authors reported using a vehicle solvent, but they did not report if the negative control was a solvent control, so it cannot be determined if the solvent concentration was appropriate.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the Bok choy seeds was not reported.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the organisms were acclimated to test conditions.	
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Study Citation:	Liao, C. S., Yen, J. H., Wang, Y. S. (2006). Effects of endocrine disruptor di-n-butyl phthalate on the growth of Bok choy (<i>Brassica rapa</i> subsp. <i>chinensis</i>). <i>Chemosphere</i> 65(10):1715-1722.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Water; Root uptake			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica rapa</i> ; subsp. <i>chinensis</i> ; Juvenile			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1298079			
Domain	Metric	Rating	Comments	
	Metric 15: Number of Organisms and Replicates per Group	Low	It was reported that the tests were completed in triplicate, but the number of plants per treatment was low: four plants each.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Organisms were kept at 25C during the day and 20C at night with a 16L:8D photoperiod. Relative humidity was at 70-95%. A modification of Hoagland’s medium as used in the hydroponic growth chambers.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest–development and growth of the Bok choy.	
	Metric 18: Consistency of Outcome Assessment	Low	Details of the outcome assessment were limited for biomass determination and for plant morphology.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. It was not reported if the plants were acclimated to test conditions.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	Low	Study authors did not report conducting statistical analysis, but independent analysis may be conducted from the data provided.	
	Metric 22: Reporting of Data	High	Data for control response and exposure related findings was adequate and can be found in Figure 1 and Table 3.	
	Metric 23: Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in the table.	
Additional Comments:	This portion of the evaluation was on the effect of DBP on Bok choy biomass and plant morphology/coloring. Development/growth was selected as the outcome of interest.			
Overall Quality Determination		Medium		

Study Citation:	Liao, C. S., Yen, J. H., Wang, Y. S. (2006). Effects of endocrine disruptor di-n-butyl phthalate on the growth of Bok choy (<i>Brassica rapa</i> subsp. <i>chinensis</i>). <i>Chemosphere</i> 65(10):1715-1722.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Water; Root uptake			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica rapa</i> ; subsp. <i>chinensis</i> ; Juvenile			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1298079			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The DBP was identified by CAS number.	
	Metric 2: Test Substance Source	High	The source of the DBP was reported to be Riedel-deHaen Co. in Germany. The DBP was analytically verified during the residual DBP analysis part of the study, by GC/MS.	
	Metric 3: Test Substance Purity	High	The purity was reported to be 98.7%	
Domain 2: Test Design				
	Metric 4: Negative Controls	Low	Study authors reported the use of a concurrent negative control. However, they did not specify if a solvent control was used. Acetone was used as a vehicle solvent, so a solvent control would be necessary.	
	Metric 5: Negative Control Response	High	The biological response of the negative control was adequate and was reported in Tables 1 and 3.	
	Metric 6: Randomized Allocation	Low	It was not reported how the Bok choy was allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	Study authors reported dissolving DBP in acetone at a concentration of 100g/L, but they did not elaborate on how the test concentrations were prepared. It was also not reported how often new test solution was added to the system.	
	Metric 8: Consistency of Exposure Administration	Medium	All exposures were conducted in 2.5L of test solution at the same temperature and relative humidity. It was not reported how often the test solution was renewed, which creates doubt regarding the consistency.	
	Metric 9: Measurement of Test Substance Concentration	Low	Study authors did not report if the DBP was measured at any point in the study.	
	Metric 10: Exposure Duration and Frequency	High	The study duration was reported to be 42 days and was appropriate for the outcomes of interest.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	There were 5 concentrations plus a control. Spacing was adequate to see a response.	
	Metric 12: Testing at or Below Solubility Limit	Medium	Study authors reported using a vehicle solvent, but they did not report if the negative control was a solvent control, so it cannot be determined if the solvent concentration was appropriate.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the Bok choy was not reported.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the organisms were acclimated to test conditions.	
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Study Citation:	Liao, C. S., Yen, J. H., Wang, Y. S. (2006). Effects of endocrine disruptor di-n-butyl phthalate on the growth of Bok choy (<i>Brassica rapa</i> subsp. <i>chinensis</i>). <i>Chemosphere</i> 65(10):1715-1722.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Water; Root uptake			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica rapa</i> ; subsp. <i>chinensis</i> ; Juvenile			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1298079			
Domain	Metric	Rating	Comments	
	Metric 15:	Number of Organisms and Replicates per Group	Low	It was reported that the tests were completed in triplicate, but the number of plants per treatment was low: four plants each.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Organisms were kept at 25C during the day and 20C at night with a 16L:8D photoperiod. Relative humidity was at 70-95%. A modification of Hoagland’s medium as used in the hydroponic growth chambers.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest–DBP accumulation in Bok choy tissues.
	Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. Assessment was described in detail in section 2.4.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. It was not reported if the plants were acclimated to test conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Study authors did not report conducting statistical analysis, but independent analysis may be conducted from the data provided.
	Metric 22:	Reporting of Data	High	Data for control response and exposure related findings was adequate and can be found in Figure 3 and in Tables 1 and 3.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in the tables.
Additional Comments:	This portion of the evaluation was on the absorption of DBP in Bok Choy leaves at various exposure levels. ADME was selected as the outcome of interest.			

Overall Quality Determination**Medium**

Study Citation:	Zhu, F., Zhu, C., Chen, N., Zhou, D., Gao, J. (2018). Will spent mushroom substrate application affect the dissipation and plant uptake of phthalate esters?. Journal of Soils and Sediments 18(4):1579-1589.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica rapa</i> ; Subspecies: Chinensis; Embryo			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5605728			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	CAS number, nomenclature and structure were reported by the authors.
	Metric 2:	Test Substance Source	Low	Source was reported (Sigma) but not verified.
	Metric 3:	Test Substance Purity	High	The purity from the manufacturer was reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Control soil was reported but authors did not detail if it was a solvent or abiotic control.
	Metric 5:	Negative Control Response	Uninformative	The control soil without DPB added at 50mg/kg has several phthalates in it already indicating the soil is contaminated. As a result, the ability to determine ADME between control and treated samples is not possible.
	Metric 6:	Randomized Allocation	Medium	Allocation was reported as random.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The addition of DBP in the soil was well described and appeared to be appropriate.
	Metric 8:	Consistency of Exposure Administration	High	There was only one exposure group at 50mg/kg in the soil.
	Metric 9:	Measurement of Test Substance Concentration	Low	Authors did not verify treatment soil concentrations.
	Metric 10:	Exposure Duration and Frequency	High	The authors allowed for 30 days of seedling growth with a previous 7-day germination period.
	Metric 11:	Number of Exposure Groups/Spacing of Exposure Levels	N/A	The authors only had one concentration of DBP (50 mg/kg).
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The source of seeds was listed, but the storage conditions of seeds prior to the experiment were not described.
	Metric 14:	Acclimatization and Pretreatment Conditions	Medium	Conditions before germination were not described.
	Metric 15:	Number of Organisms and Replicates per Group	Low	Authors reported six seeds per pot, but no record of the number of pots per treatment group or replication. Samples were replicated for analytical analysis.
Domain 5: Outcome Assessment				
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Study Citation:	Zhu, F., Zhu, C., Chen, N., Zhou, D., Gao, J. (2018). Will spent mushroom substrate application affect the dissipation and plant uptake of phthalate esters?. Journal of Soils and Sediments 18(4):1579-1589.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Brassica rapa</i> ; Subspecies: Chinensis; Embryo			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5605728			
Domain	Metric	Rating	Comments	
	Metric 16:	Adequacy of Test Conditions	Medium	Soil conditions were described and environmental conditions were controlled.
	Metric 17:	Outcome Assessment Methodology	Low	Table 5 reports the DBP concentrations, however, all plants have other phthalates that indicate controlled conditions for the uptake of just DBP were not possible.
	Metric 18:	Consistency of Outcome Assessment	High	
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	Authors do not report any differences among groups that would influence results.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	Nothing provided indicates that differences were due to plant health or attrition.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Authors used ANOVA with Tukey post-hoc to determine differences among treatment groups.
	Metric 22:	Reporting of Data	Low	Values are presented for DBP concentrations as Means +/- SD but some parts of the plant are only presented as single values.
	Metric 23:	Explanation of Unexpected Outcomes	Medium	It is not clear why other phthalate concentrations are present in Table 5, if control soil was not initially contaminated.
Additional Comments:	There is phthalate contamination in the control soil. Exposure concentrations were not verified.			

Overall Quality Determination**Uninformative**

Study Citation:	Virgin, H. I., A-M, Holst, Morner, J. (1981). Effect of di-n-butylphthalate on the carotenoid synthesis in green plants. Physiologia Plantarum 53(2):158-163.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Browallia speciosa</i> ; Major; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Photosynthesis			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1333234			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	High	The test substance identity was analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Medium	Pro analysis quality was reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	Low	Untreated plants were used as controls but few details were provided.	
	Metric 5: Negative Control Response	Low	The biological response of the negative control groups was not reported.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	Several methods of obtaining vapors were reported but effects were not attributed to a particular method.	
	Metric 8: Consistency of Exposure Administration	Low	It was difficult to determine as durations and concentrations were not clearly reported.	
	Metric 9: Measurement of Test Substance Concentration	Medium	Exposure concentrations were measured but not clearly reported.	
	Metric 10: Exposure Duration and Frequency	Low	Exposures seemed to be around 4-7 days, but study length was not clearly reported.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Uninformative	No information is provided on the number of exposure groups, the range seems to be 10-150 ng/L, but it's unclear if this is one treatment that varied over time or if there were multiple treatment levels.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via vapors.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the test plants was not reported, and age or stage at test initiation was not clear.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.	
	Metric 15: Number of Organisms and Replicates per Group	Low	Numbers and replicates were not reported.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.	
	Metric 17: Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported.	

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Study Citation:	Virgin, H. I., A-M, Holst, Morner, J. (1981). Effect of di-n-butylphthalate on the carotenoid synthesis in green plants. Physiologia Plantarum 53(2):158-163.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Browallia speciosa</i> ; Major; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Photosynthesis			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1333234			
Domain		Metric	Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were confusing.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	N/A	Statistical analysis was not possible, and information was not quantified.
	Metric 22:	Reporting of Data	Low	Data were only reported for some outcomes.
	Metric 23:	Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability.
Additional Comments:	This was a closed system and light was a variable.			

Overall Quality Determination**Uninformative**

Study Citation:	Virgin, H. I., A-M, Holst, Morner, J. (1981). Effect of di-n-butylphthalate on the carotenoid synthesis in green plants. Physiologia Plantarum 53(2):158-163.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Browallia speciosa</i> ; Major; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1333234			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	Chemical was identified by name only	
	Metric 2: Test Substance Source	High	The test substance identity was analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Medium	pro analysis quality was reported	
Domain 2: Test Design				
	Metric 4: Negative Controls	Low	Untreated plants were used as controls but few details were provided	
	Metric 5: Negative Control Response	Low	The biological response of the negative control groups was not reported.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	Several methods of obtaining vapors was reported but effects were not attributed to a particular method	
	Metric 8: Consistency of Exposure Administration	Low	Difficult to determine as durations and concentrations were not clearly reported	
	Metric 9: Measurement of Test Substance Concentration	Medium	Exposure concentrations were measured but not clearly reported	
	Metric 10: Exposure Duration and Frequency	Low	Exposures seemed to be around 4-7 days but study length was not clearly reported	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Uninformative	No information is provided on the number of exposure groups, the range seems to be 10-150 ng/L but unclear if this is one treatment that varied over time or if there were multiple treatment levels	
	Metric 12: Testing at or Below Solubility Limit	N/A	exposure was via vapors	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the test plants was not reported, age or stage at test initiation was not clear.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized	
	Metric 15: Number of Organisms and Replicates per Group	Low	Numbers and replicates were not reported	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate	
	Metric 17: Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported	
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were confusing	

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Study Citation:	Virgin, H. I., A-M, Holst, Morner, J. (1981). Effect of di-n-butylphthalate on the carotenoid synthesis in green plants. <i>Physiologia Plantarum</i> 53(2):158-163.		
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported		
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Browallia speciosa</i> ; Major; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Mortality		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	1333234		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	N/A	Statistical analysis was not possible, information was not quantified
Metric 22:	Reporting of Data	Low	There was brief mention of plant mortality in the results section text. No data was shown.
Metric 23:	Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability
Additional Comments: closed system, light was a variable			

Overall Quality Determination**Uninformative**

Study Citation:	Virgin, H. I., A-M, Holst, Morner, J. (1981). Effect of di-n-butylphthalate on the carotenoid synthesis in green plants. Physiologia Plantarum 53(2):158-163.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Browallia speciosa</i> ; Major; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Cytotoxicity-Photosynthesis			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1333234			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	Chemical was identified by name only	
	Metric 2: Test Substance Source	High	The test substance identity was analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Medium	pro analysis quality was reported	
Domain 2: Test Design				
	Metric 4: Negative Controls	Low	Untreated plants were used as controls but few details were provided	
	Metric 5: Negative Control Response	Low	The biological response of the negative control groups was not reported.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	Several methods of obtaining vapors was reported but effects were not attributed to a particular method	
	Metric 8: Consistency of Exposure Administration	Low	Difficult to determine as durations and concentrations were not clearly reported	
	Metric 9: Measurement of Test Substance Concentration	Medium	Exposure concentrations were measured but not clearly reported	
	Metric 10: Exposure Duration and Frequency	Low	Exposures seemed to be around 4-7 days but study length was not clearly reported	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Uninformative	No information is provided on the number of exposure groups, the range seems to be 10-150 ng/L but unclear if this is one treatment that varied over time or if there were multiple treatment levels	
	Metric 12: Testing at or Below Solubility Limit	N/A	exposure was via vapors	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the test plants was not reported, age or stage at test initiation was not clear.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized	
	Metric 15: Number of Organisms and Replicates per Group	Low	Numbers and replicates were not reported	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate	
	Metric 17: Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported	
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were confusing	

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Study Citation:	Virgin, H. I., A-M, Holst, Morner, J. (1981). Effect of di-n-butylphthalate on the carotenoid synthesis in green plants. <i>Physiologia Plantarum</i> 53(2):158-163.		
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported		
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Browallia speciosa</i> ; Major; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Mechanistic-Cytotoxicity-Photosynthesis		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	1333234		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	N/A	Statistical analysis was not possible, information was not quantified
Metric 22:	Reporting of Data	Low	Data were only reported for some outcomes
Metric 23:	Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability
Additional Comments: open system, light was a variable			

Overall Quality Determination**Uninformative**

Study Citation:	Kannan, S., Ramani, S. (1987). Mechanisms of fe-deficiency tolerance in crop cultivars - effects of dibutyl phthalate and caffeic acid on fe-chlorosis recovery. Journal of Plant Nutrition 10(9-16):1051-1058.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Terrestrial; Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Carica papaya</i> ; Co-1; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Cell signaling/function-Nutritional and Metabolic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5433168			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	Low	The source was not reported.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Author reported a treatment without DBP or caffeic acid
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcome
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided no details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Low	No details of exposure administration were reported
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	Uninformative	duration was not reported
	Metric 11:	Number of Exposure Groups/Spacing of Exposure Levels	N/A	Only one concentration tested
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test plants was not reported
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of test organisms and/or replicates was not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported

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Study Citation:	Kannan, S., Ramani, S. (1987). Mechanisms of fe-deficiency tolerance in crop cultivars - effects of dibutyl phthalate and caffeic acid on fe-chlorosis recovery. Journal of Plant Nutrition 10(9-16):1051-1058.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Terrestrial; Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Carica papaya</i> ; Co-1; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Cell signaling/function-Nutritional and Metabolic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5433168			
Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Uninformative	Statistical analysis was not conducted.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability
Additional Comments: None				

Overall Quality Determination**Uninformative**

Study Citation:	Kannan, S., Ramani, S. (1987). Mechanisms of fe-deficiency tolerance in crop cultivars - effects of dibutyl phthalate and caffeic acid on fe-chlorosis recovery. Journal of Plant Nutrition 10(9-16):1051-1058.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Terrestrial; Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Carica papaya</i> ; Co-3; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Cell signaling/function-Nutritional and Metabolic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5433168			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	Low	The source was not reported.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Author reported a treatment without DBP or caffeic acid
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcome
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided no details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Low	No details of exposure administration were reported
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	Uninformative	duration was not reported
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	Only one concentration tested
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test plants was not reported
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of test organisms and/or replicates was not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported

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Study Citation:	Kannan, S., Ramani, S. (1987). Mechanisms of fe-deficiency tolerance in crop cultivars - effects of dibutyl phthalate and caffeic acid on fe-chlorosis recovery. Journal of Plant Nutrition 10(9-16):1051-1058.		
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported		
Exposure Route, Media, Path:	Terrestrial; Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Carica papaya</i> ; Co-3; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Mechanistic-Cell signaling/function-Nutritional and Metabolic		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	5433168		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	Uninformative	Statistical analysis was not conducted.
Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
Metric 23:	Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability
Additional Comments: None			
Overall Quality Determination		Uninformative	

Study Citation:	Kannan, S., Ramani, S. (1987). Mechanisms of fe-deficiency tolerance in crop cultivars - effects of dibutyl phthalate and caffeic acid on fe-chlorosis recovery. Journal of Plant Nutrition 10(9-16):1051-1058.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Terrestrial; Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Carica papaya</i> ; Co-4; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Cell signaling/function-Nutritional and Metabolic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5433168			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The chemical was identified by name only.
	Metric 2:	Test Substance Source	Low	The source was not reported.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of the test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Author reported a treatment without DBP or caffeic acid.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for the assessed outcome.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided no details on the measures taken to appropriately prepare test concentrations.
	Metric 8:	Consistency of Exposure Administration	Low	No details of exposure administration were reported.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	Uninformative	Exposure duration was not reported.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	Only one concentration was tested.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test plants was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of test organisms and/or replicates were not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported.
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported.

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Study Citation:	Kannan, S., Ramani, S. (1987). Mechanisms of fe-deficiency tolerance in crop cultivars - effects of dibutyl phthalate and caffeic acid on fe-chlorosis recovery. Journal of Plant Nutrition 10(9-16):1051-1058.		
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported		
Exposure Route, Media, Path:	Terrestrial; Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Carica papaya</i> ; Co-4; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Mechanistic-Cell signaling/function-Nutritional and Metabolic		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	5433168		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	Uninformative	Statistical analysis was not conducted.
Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
Metric 23:	Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability.
Additional Comments: None			
Overall Quality Determination		Uninformative	

Study Citation:	Isogai, Y., Komoda, Y., Okamoto, T. (1972). Biological activities of n-butyl phthalate and its analogous compounds on various bioassays of plant growth regulators. Scientific papers of the College of General Education, University of Tokyo 22(2):129-135.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Dermal (topical application)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Cucumis sativus</i> ; cv. National Pickling stock; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5551990			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical name and structure were used to identify the test chemical.
	Metric 2:	Test Substance Source	Low	The source was not reported.
	Metric 3:	Test Substance Purity	Low	Purity or grade of the test substance was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using a concurrent solvent control group.
	Metric 5:	Negative Control Response	Low	The biological response of the solvent control group was not reported.
	Metric 6:	Randomized Allocation	Low	A selection criteria was indicated (selected seedlings with 3 cm of hypocotyl), but details regarding assignment to groups were not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations and test concentrations were not measured. Vessel material was not reported.
	Metric 8:	Consistency of Exposure Administration	Low	The study provided few details on exposure administration.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type.
	Metric 11:	Number of Exposure Groups/Spacing of Exposure Levels	Medium	Only three concentrations were used, but a wide range was tested.
	Metric 12:	Testing at or Below Solubility Limit	Low	Both treatment solutions and the control solution were prepared with 90% ethanol, which is miscible with DBP. The biological response of the solvent control was not reported.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Cucumber are an appropriate species for this assay and the source of the seeds was reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups.
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of test organisms and replicates was not reported.

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Study Citation:	Isogai, Y., Komoda, Y., Okamoto, T. (1972). Biological activities of n-butyl phthalate and its analogous compounds on various bioassays of plant growth regulators. Scientific papers of the College of General Education, University of Tokyo 22(2):129-135.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Dermal (topical application)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Cucumis sativus</i> ; cv. National Pickling stock; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5551990			
Domain	Metric	Rating	Comments	
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed the intended outcome of interest.
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Uninformative	Statistical analysis was not conducted and no data was provided.
	Metric 22:	Reporting of Data	Low	Data were not shown for treatment or control groups, but results were described in the text.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.
Additional Comments:	This portion of the study was on the plant growth promoting activity of DBP on <i>Cucumis sativus</i> cv National Pickling stock. Development/Growth was selected as the outcome of interest. This portion of the study is rated unacceptable as statistical analysis was not conducted and no numerical data was provided.			

Overall Quality Determination**Uninformative**

Study Citation:	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. Frontiers of Environmental Science & Engineering 9(2):259-268.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Cucumis sativus</i> ; Embryo			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Photosynthesis			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2915866			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.6%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was adequate
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were justified for a dose response
	Metric 12:	Testing at or Below Solubility Limit	N/A	seeds exposed via soil
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test seeds were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed seeds
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
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Study Citation:	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. Frontiers of Environmental Science & Engineering 9(2):259-268.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Cucumis sativus</i> ; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Photosynthesis
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	2915866

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
	Metric 20: Outcomes Unrelated to Exposure	Medium	Nothing reported to indicate there were differences among groups that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Section 2.7 (page 3/10) does not present the statistical tests used to perform analysis other than the program used and p-value for significance.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group and were adequate to determine values for the endpoint
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes
Additional Comments:	Pigment content”The results indicate that longer periods of cultivation of the test plants may make it easier to interpret the changes in pigment contents.”		

Overall Quality Determination**High**

Study Citation:	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. Frontiers of Environmental Science & Engineering 9(2):259-268.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Cucumis sativus</i> ; Embryo			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2915866			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	Chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity reported as 99.6%	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was adequate.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were justified for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Seeds exposed via soil.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test seeds were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed seeds.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.	
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Study Citation:	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. <i>Frontiers of Environmental Science & Engineering</i> 9(2):259-268.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Cucumis sativus</i> ; Embryo
Health Outcome:	Reproductive/Teratogenic
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	2915866

Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	Nothing reported to indicate there were differences among groups that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Section 2.7 (page 3/10) does not present the statistical tests used to perform analysis other than the program used and p-value for significance.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group and were adequate to determine values for the endpoint.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments: This form represents the germination rate results presented in Table 1 for <i>Cucumis sativus</i> with DBP exposure.			

Overall Quality Determination**High**

Study Citation:	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. Frontiers of Environmental Science & Engineering 9(2):259-268.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Cucumis sativus</i> ; Embryo			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2915866			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity was reported as 99.6%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was adequate.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were justified for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Seeds were exposed via soil.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test seeds were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed seeds.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.	
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Study Citation:	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. Frontiers of Environmental Science & Engineering 9(2):259-268.		
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)		
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Cucumis sativus</i> ; Embryo		
Health Outcome:	Development/Growth		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	2915866		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
Metric 20:	Outcomes Unrelated to Exposure	Medium	Nothing was reported to indicate there were differences among groups that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	Low	Section 2.7 (page 3/10) does not present the statistical tests used to perform analysis other than the program used and p-value for significance.
Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group and were adequate to determine values for the endpoint.
Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments:	"DnBP and DEHP at a range of concentrations in the experimental soil showed no discernible effect on the germination rate of the seven test plant species. However, they did exert effects on root elongation, seedling growth and biomass to different extents, indicating the potential applicability of seedling growth in the evaluation of the phytotoxicity of PAE compounds."		

Overall Quality Determination**High**

Study Citation:	Wang, L., Sun, X., Chang, Q., Tao, Y., Wang, L., Dong, J., Lin, Y., Zhang, Y. (2016). Effect of di-n-butyl phthalate (DBP) on the fruit quality of cucumber and the health risk. Environmental Science and Pollution Research 23(23):24298-24304.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Cucumis sativus</i> ; Juvenile			
Health Outcome:	Mechanistic-Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3502464			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The DBP was identified by name only.	
	Metric 2: Test Substance Source	Low	The source of the DBP was not reported.	
	Metric 3: Test Substance Purity	Low	The purity was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of a negative control with black soil only.	
	Metric 5: Negative Control Response	High	The response of the negative control was reported in Table 1 and was adequate for the outcome of interest.	
	Metric 6: Randomized Allocation	Medium	It was reported that a random block design was used for this study.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The test media was prepared by making a stock solution of 100mg/mL DBP in ethanol and then adding that in the appropriate amount to black soil. The ethanol was allowed to evaporate before the soil was used for the study. The tests took place in 40 x 80 x 40 class boxes.	
	Metric 8: Consistency of Exposure Administration	Medium	All tests took place in class boxes of the same size with one seedling each until the harvest of the fruit.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if the DBP concentrations were measured at any point in the study.	
	Metric 10: Exposure Duration and Frequency	Low	The study was reported to be from the time of a seedling with three leaves until fruit harvest. This was given a low because the fruit was the central point of interest in the study, so harvest was an appropriate stopping point, but study authors did not report the number of days from the start of the study to the harvest.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	There were four exposure levels, and the spacing was appropriate for a response.	
	Metric 12: Testing at or Below Solubility Limit	N/A	The exposure was via soil.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the cucumber seeds was not reported.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if any acclimation occurred.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There was one organism per test chamber and five replicates per test concentration.	

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Study Citation:	Wang, L., Sun, X., Chang, Q., Tao, Y., Wang, L., Dong, J., Lin, Y., Zhang, Y. (2016). Effect of di-n-butyl phthalate (DBP) on the fruit quality of cucumber and the health risk. Environmental Science and Pollution Research 23(23):24298-24304.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Cucumis sativus</i> ; Juvenile			
Health Outcome:	Mechanistic-Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3502464			
Domain	Metric	Rating	Comments	
Domain 5: Outcome Assessment				
Metric 16:	Adequacy of Test Conditions	Medium	It was reported that the cucumbers were kept at 27C during the day and 20C at night with a relative humidity of 70%. Watering was not reported nor was photoperiod.	
Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—organic acid content, soluble protein content, soluble sugar content, and vitamin C content in cucumber fruits.	
Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups.	
Domain 6: Confounding / Variable Control				
Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.	
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
Metric 21:	Statistical Methods	High	Statistical methods were described in the "statistical analyses" section of the paper.	
Metric 22:	Reporting of Data	High	The exposure response and the control response are presented in Table 1 and are adequate for the outcomes of interest.	
Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in Table 1.	
Additional Comments:	This portion of the evaluation was on the effect of cucumber fruit quality after exposure to various levels of DBP. Since fruit is the reproductive organ of the plant and the study authors were observing effects on organic acid, soluble protein, soluble sugar, and vitamin C content of the fruit, the mechanistic reproduction outcome was chosen.			

Overall Quality Determination**Medium**

Study Citation:	Wang, L., Sun, X., Chang, Q., Tao, Y., Wang, L., Dong, J., Lin, Y., Zhang, Y. (2016). Effect of di-n-butyl phthalate (DBP) on the fruit quality of cucumber and the health risk. Environmental Science and Pollution Research 23(23):24298-24304.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Cucumis sativus</i> ; Juvenile			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3502464			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The DBP was identified by name only.	
	Metric 2: Test Substance Source	Low	The source of the DBP was not reported.	
	Metric 3: Test Substance Purity	Low	The purity was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of a negative control with black soil only.	
	Metric 5: Negative Control Response	High	The response of the negative control was reported in Table 2 and was adequate for the outcome of interest.	
	Metric 6: Randomized Allocation	Medium	It was reported that a random block design was used for this study.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	The test media was prepared by making a stock solution of 100mg/mL DBP in ethanol and then adding that in the appropriate amount to black soil. The ethanol was allowed to evaporate before the soil was used for the study. The tests took place in 40 x 80 x 40 class boxes.	
	Metric 8: Consistency of Exposure Administration	Medium	All tests took place in class boxes of the same size with one seedling each until the harvest of the fruit.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if the DBP concentrations were measured at any point in the study.	
	Metric 10: Exposure Duration and Frequency	Low	The study was reported to be from the time of a seedling with 3 leaves until fruit harvest. This was given a low because the fruit was the central point of interest in the study, so harvest was an appropriate stopping point, but study authors did not report the number of days from the start of the study to the harvest.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	There were 4 exposure levels, and the spacing was appropriate for a response.	
	Metric 12: Testing at or Below Solubility Limit	N/A	The exposure was via soil.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the cucumber seeds was not reported.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if any acclimation occurred.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There was one organism per test chamber and 5 replicates per test concentration.	
Domain 5: Outcome Assessment				
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Study Citation:	Wang, L., Sun, X., Chang, Q., Tao, Y., Wang, L., Dong, J., Lin, Y., Zhang, Y. (2016). Effect of di-n-butyl phthalate (DBP) on the fruit quality of cucumber and the health risk. Environmental Science and Pollution Research 23(23):24298-24304.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Cucumis sativus</i> ; Juvenile			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3502464			
Domain	Metric	Rating	Comments	
	Metric 16:	Adequacy of Test Conditions	Medium	It was reported that the cucumbers were kept at 27C during the day and 20C at night with a relative humidity of 70%. Watering was not reported nor was photoperiod.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—accumulation of DBP in cucumber fruit tissue.
	Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups—GC-MS was used to determine DBP levels in cucumber fruits.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were described in the "statistical analyses" section of the paper.
	Metric 22:	Reporting of Data	High	The exposure response and the control response are presented in Table 2 and are adequate for the outcomes of interest.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in Table 2.
Additional Comments:	This portion of the evaluation was on the accumulation of DBP in cucumber fruit tissue. ADME was selected as the outcome of interest.			

Overall Quality Determination**Medium**

Study Citation:	Wu, Y., Yuan, S. L. (2012). Dibutyl phthalate pollution on cotton growth and physiological characteristics of cotton. Advanced Materials Research 518:523:5436-5441.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Gossypium</i> ; Species not reported in text.; Embryo			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1639289			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The test substance was identified by name only.	
	Metric 2: Test Substance Source	Low	The source of the DBP was not reported.	
	Metric 3: Test Substance Purity	Low	The purity and grade of the test substance were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	The study authors reported the use of a concurrent negative control and a solvent control with acetone.	
	Metric 5: Negative Control Response	High	The negative control response and the solvent control response were reported in Figure 1 and in Table 1.	
	Metric 6: Randomized Allocation	Low	It was not reported how the cotton seeds were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	Limited details were provided on the test system and on the preparation of the test concentrations. Exposure was conducted in glass Petri dishes.	
	Metric 8: Consistency of Exposure Administration	Low	Little details were provided on the exposure administration.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if the test concentrations were measured at any point in the study.	
	Metric 10: Exposure Duration and Frequency	High	The exposure duration for the germination and root length portion of the study was reported to be five days. This was adequate to observe a response.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	There were 10 exposure levels plus a negative control and a solvent control. Spacing appeared adequate to observe a response.	
	Metric 12: Testing at or Below Solubility Limit	High	Study authors reported the use of acetone as a vehicle solvent. The solvent control response was adequate.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the cotton seeds was not reported. The scientific name was also not reported.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the organisms were acclimated.	
	Metric 15: Number of Organisms and Replicates per Group	Low	The number of cotton seeds used and the number of replicates was not reported.	

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Study Citation:	Wu, Y., Yuan, S. L. (2012). Dibutyl phthalate pollution on cotton growth and physiological characteristics of cotton. Advanced Materials Research 518-523:5436-5441.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Gossypium</i> ; Species not reported in text.; Embryo
Health Outcome:	Reproductive/Teratogenic
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	1639289

Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	Low	The environmental conditions were not sufficiently reported to evaluate if they were adequate for organism health.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—seed germination/reproduction.
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol were not reported.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Statistical analysis was performed but the methods used were not described.
	Metric 22: Reporting of Data	High	The control response and the exposure response for germination were reported in Figure 1 and in Table 1.
	Metric 23: Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in Table 1.

Additional Comments: This portion of the evaluation was on the effect of DBP on the germination of cotton seeds. Reproduction was selected as the outcome of interest.

Overall Quality Determination

Low

Study Citation:	Wu, Y., Yuan, S. L. (2012). Dibutyl phthalate pollution on cotton growth and physiological characteristics of cotton. Advanced Materials Research 518-523:5436-5441.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Gossypium</i> ; Species not reported in text.; Embryo			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1639289			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The test substance was identified by name only.	
	Metric 2: Test Substance Source	Low	The source of the DBP was not reported.	
	Metric 3: Test Substance Purity	Low	The purity and grade of the test substance were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	The study authors reported the use of a concurrent negative control and a solvent control with acetone.	
	Metric 5: Negative Control Response	Medium	The negative control response was reported in Table 1. It is uncertain if the control response reported was the solvent control or not.	
	Metric 6: Randomized Allocation	Low	It was not reported how the cotton seeds were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	Limited details were provided on the test system and on the preparation of the test concentrations. Exposure was conducted in glass Petri dishes.	
	Metric 8: Consistency of Exposure Administration	Low	Little details were provided on the exposure administration.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if the test concentrations were measured at any point in the study.	
	Metric 10: Exposure Duration and Frequency	High	The exposure duration for the germination and root length portion of the study was reported to be 5 days. This was adequate to observe a response.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	There were 10 exposure levels plus a negative control and a solvent control. Spacing appeared adequate to observe a response.	
	Metric 12: Testing at or Below Solubility Limit	High	Study authors reported the use of acetone a vehicle solvent. The solvent control response was adequate.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the cotton seeds was not reported. The scientific name was also not reported.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the organisms were acclimated.	
	Metric 15: Number of Organisms and Replicates per Group	Low	The number of cotton seeds used and the number of replicates was not reported.	
Domain 5: Outcome Assessment				
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Study Citation:	Wu, Y., Yuan, S. L. (2012). Dibutyl phthalate pollution on cotton growth and physiological characteristics of cotton. Advanced Materials Research 518-523:5436-5441.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Gossypium</i> ; Species not reported in text.; Embryo
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	1639289

Domain	Metric	Rating	Comments
	Metric 16: Adequacy of Test Conditions	Low	The environmental conditions were not sufficiently reported to evaluate if they were adequate for organism health.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—root length.
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol were not reported.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Statistical analysis was performed but the methods used were not described.
	Metric 22: Reporting of Data	High	The control response and the exposure response for germination were reported in Table 1.
	Metric 23: Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in Table 1.

Additional Comments: This portion of the evaluation was on the effect of DBP on the root length of cotton seeds. Development/growth was selected as the outcome of interest.

Overall Quality Determination

Low

Study Citation:	Dueck, T. A., Dijk, Van, C. J., David, F., Scholz, N., Vanwallegthem, F. (2003). Chronic effects of vapour phase di-n-butyl phthalate (DBP) on six plant species. Chemosphere 53(8):911-920.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Holcus lanatus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1302103			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#.
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Study authors reported using an appropriate concurrent negative control group, note that all controls had some DBP detected. Control group DBP concentrations are reported in Table 1.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail. The chambers were constructed of "hardened glass" and aluminum.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups based on timed samplings.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies. Samples were taken frequently: "Actual DBP concentrations were measured by taking air samples from each chamber using an automated gas sampling device (GS-1 gas sampler, Gerstel, M €ulheim, Germany). Sampling was performed with Tenax TA tubes (2,6-diphenylphenylene oxide polymer) at 250ml min ⁻¹ for 20 min. During the fumigation experiment, each chamber was sampled in duplicate on a weekly basis during the complete exposure period (76 days)."
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	The number of exposure groups and spacing of exposure levels were suitable. Concentration gradient is reported as both actual and nominal within table 1.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via fumigation in a continually renewed chamber.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The crop and herbaceous species originated from germinated seeds, but source was not explicitly reported.

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Study Citation:	Dueck, T. A., Dijk, Van, C. J., David, F., Scholz, N., Vanwalleghe, F. (2003). Chronic effects of vapour phase di-n-butyl phthalate (DBP) on six plant species. Chemosphere 53(8):911-920.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Holcus lanatus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1302103			
Domain	Metric	Rating	Comments	
	Metric 14: Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions.	
	Metric 15: Number of Organisms and Replicates per Group	Low	Four plants per treatment and no replicates reported, "Three to four weeks after sowing, when the first mature leaves appeared, four uniform individuals per species including the tree species were placed in each fumigation chamber (the maximum number physically possible) for exposure to DBP.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	
	Metric 18: Consistency of Outcome Assessment	Medium	Outcomes were assessed consistently across study groups but few details were reported.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.	
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group in Table 2.	
	Metric 23: Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.	
Additional Comments:	This form is for ADME (biotransformation) assessment of DBP concentration in leaf tissue.			

Overall Quality Determination**High**

Study Citation:	Dueck, T. A., Dijk, Van, C. J., David, F., Scholz, N., Vanwalleghem, F. (2003). Chronic effects of vapour phase di-n-butyl phthalate (DBP) on six plant species. Chemosphere 53(8):911-920.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Holcus lanatus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1302103			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The chemical was identified by name and CAS#.
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity or grade of the test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Study authors reported using an appropriate concurrent negative control group, note that all controls had some DBP detected. Control group DBP concentrations are reported in Table 1.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail. The chambers were constructed of "hardened glass" and aluminum.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups based on timed samplings.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies. Samples were taken frequently: "Actual DBP concentrations were measured by taking air samples from each chamber using an automated gas sampling device (GS-1 gas sampler, Gerstel, M €ulheim, Germany). Sampling was performed with Tenax TA tubes (2,6-diphenylphenylene oxide polymer) at 250ml min ¹ for 20 min. During the fumigation experiment, each chamber was sampled in duplicate on a weekly basis during the complete exposure period (76 days)."
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	The number of exposure groups and spacing of exposure levels were suitable. Concentration gradient is reported as both actual and nominal within table 1.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via fumigation in a continually renewed chamber.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The crop and herbaceous species originated from germinated seeds, but source was not explicitly reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions.
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Study Citation:	Dueck, T. A., Dijk, Van, C. J., David, F., Scholz, N., Vanwalleghe, F. (2003). Chronic effects of vapour phase di-n-butyl phthalate (DBP) on six plant species. Chemosphere 53(8):911-920.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Holcus lanatus</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	1302103

Domain	Metric	Rating	Comments
	Metric 15: Number of Organisms and Replicates per Group	Low	Four plants per treatment and no replicates reported, "Three to four weeks after sowing, when the first mature leaves appeared, four uniform individuals per species including the tree species were placed in each fumigation chamber (the maximum number physically possible) for exposure to DBP."
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18: Consistency of Outcome Assessment	Medium	Outcomes were assessed consistently across study groups but few details were reported.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.
Additional Comments: This form represents growth outcomes associated with Dry Weight reported for <i>Holcus</i> shoot and roots within Figure 3 on page 6/10.			

Overall Quality Determination**High**

Study Citation:	Chuah, T. S., Oh, H. Y., Habsah, M., Norhafizah, M. Z., Ismail, B. S. (2014). Potential of crude extract and isolated compounds from golden beard grass (<i>Chrysopogon serrulatus</i>) for control of sprangletop (<i>Leptochloa chinensis</i>) in aerobic rice systems. Crop and Pasture Science 65(5):461-469.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Leptochloa chinensis</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5432995			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory. This may be an extracted plant extract for this test.	
	Metric 3: Test Substance Purity	Uninformative	No information was provided on purity of the chemical which was an extracted fraction of plant material.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a negative control group.	
	Metric 5: Negative Control Response	Low	The biological response of the negative control group was not reported.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations, although the test concentration was not reported.	
	Metric 8: Consistency of Exposure Administration	Medium	There was no mention of irregularities in exposure administration.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and exposure frequency were reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	N/A	Only one treatment was tested.	
	Metric 12: Testing at or Below Solubility Limit	N/A	No concentration was reported.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	Fifty seeds were used, but there were no replicates.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.	

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Study Citation:	Chuah, T. S., Oh, H. Y., Habsah, M., Norhafizah, M. Z., Ismail, B. S. (2014). Potential of crude extract and isolated compounds from golden beard grass (Chrysopogon serrulatus) for control of sprangletop (Leptochloa chinensis) in aerobic rice systems. Crop and Pasture Science 65(5):461-469.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Leptochloa chinensis</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5432995			
Domain	Metric	Rating	Comments	
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described.
	Metric 22:	Reporting of Data	Low	Control data was not presented.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments:	This is for table 4 germination. This outcome was judged unacceptable because the source of the exposure compound was unclear, and it appears to have been applied as a mixture of compounds derived from solvent extraction of a plant homogenate. The exposure concentration is unknown.			

Overall Quality Determination**Uninformative**

Study Citation:	Chuah, T. S., Oh, H. Y., Habsah, M., Norhafizah, M. Z., Ismail, B. S. (2014). Potential of crude extract and isolated compounds from golden beard grass (<i>Chrysopogon serrulatus</i>) for control of sprangletop (<i>Leptochloa chinensis</i>) in aerobic rice systems. <i>Crop and Pasture Science</i> 65(5):461-469.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Leptochloa chinensis</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5432995			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	PESTANAL analytic standard grade was used.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported.	
	Metric 6: Randomized Allocation	Medium	"Each experiment was arranged in a completely randomized design with five replications. Each experiment was repeated twice."	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.	
	Metric 8: Consistency of Exposure Administration	Medium	There was no mention of irregularities in exposure administration.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and exposure frequency were reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	Three treatments over an acceptable range were tested.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via soil.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	"Each experiment was arranged in a completely randomized design with five replications. Each experiment was repeated twice."	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.	
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Study Citation:	Chuah, T. S., Oh, H. Y., Habsah, M., Norhafizah, M. Z., Ismail, B. S. (2014). Potential of crude extract and isolated compounds from golden beard grass (<i>Chrysopogon serrulatus</i>) for control of sprangletop (<i>Leptochloa chinensis</i>) in aerobic rice systems. <i>Crop and Pasture Science</i> 65(5):461-469.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Leptochloa chinensis</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	5432995

Domain	Metric	Rating	Comments
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups, and data was pooled over several days of measurements.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Statistical differences were not determined. Standard deviations of the mean were provided visually in Fig. 4.
	Metric 22: Reporting of Data	Medium	Data was pooled for all stages of seedlings.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This form is for Fig. 4, germination and shoot weight.

Overall Quality Determination

Medium

Study Citation:	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. Frontiers of Environmental Science & Engineering 9(2):259-268.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Lolium perenne</i> ; Embryo			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2915866			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The source was reported, but the test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity was reported as 99.6%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was adequate.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were justified for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Seeds were exposed via soil.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test seeds were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed seeds.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	

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Study Citation:	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. Frontiers of Environmental Science & Engineering 9(2):259-268.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Lolium perenne</i> ; Embryo			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2915866			
Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	Nothing was reported to indicate there were differences among groups that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Section 2.7 (page 3/10) does not present the statistical tests used to perform analysis other than the program used and p-value for significance.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group and were adequate to determine values for the endpoint.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments:	"DnBP and DEHP at a range of concentrations in the experimental soil showed no discernible effect on the germination rate of the seven test plant species. However, they did exert effects on root elongation, seedling growth and biomass to different extents, indicating the potential applicability of seedling growth in the evaluation of the phytotoxicity of PAE compounds."			

Overall Quality Determination**High**

Study Citation:	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. Frontiers of Environmental Science & Engineering 9(2):259-268.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Lolium perenne</i> ; Embryo			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Photosynthesis			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2915866			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity was reported as 99.6%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was adequate.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were justified for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	N/A	seeds were exposed via soil.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test seeds were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed seeds.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.	
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Study Citation:	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. Frontiers of Environmental Science & Engineering 9(2):259-268.		
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)		
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Lolium perenne</i> ; Embryo		
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Photosynthesis		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	2915866		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was nothing reported to indicate there were differences among groups that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	Low	Section 2.7 (page 3/10) does not present the statistical tests used to perform analysis other than the program used and p-value for significance.
Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group and were adequate to determine values for the endpoint.
Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments:	This is for pigment content."The results indicate that longer periods of cultivation of the test plants may make it easier to interpret the changes in pigment contents."		
Overall Quality Determination		High	

Study Citation:	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. Frontiers of Environmental Science & Engineering 9(2):259-268.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Lolium perenne</i> ; Embryo			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2915866			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only.
	Metric 2:	Test Substance Source	Low	Source was reported but he test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.6%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was adequate.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were justified for a dose response.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Seeds exposed via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test seeds were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed seeds.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.

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Study Citation:	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. Frontiers of Environmental Science & Engineering 9(2):259-268.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Lolium perenne</i> ; Embryo
Health Outcome:	Reproductive/Teratogenic
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	2915866

Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	Nothing reported to indicate there were differences among groups that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Section 2.7 (page 3/10) does not present the statistical tests used to perform analysis other than the program used and p-value for significance.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group and were adequate to determine values for the endpoint.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments: This form represents the germination rate results presented in Table 1 for <i>Lolium perenne</i> with DBP exposure.			

Overall Quality Determination**High**

Study Citation:	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. Frontiers of Environmental Science & Engineering 9(2):259-268.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Medicago sativa</i> ; Embryo			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2915866			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only.
	Metric 2:	Test Substance Source	Low	Source was reported, the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was adequate.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were justified for a dose response.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Seeds exposed via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test seeds were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed seeds.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.

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Study Citation:	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. Frontiers of Environmental Science & Engineering 9(2):259-268.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Medicago sativa</i> ; Embryo			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2915866			
Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	Nothing reported to indicate there were differences among groups that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Section 2.7 (page 3/10) does not present the statistical tests used to perform analysis other than the program used and p-value for significance.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group and were adequate to determine values for the endpoint.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments:	"DnBP and DEHP at a range of concentrations in the experimental soil showed no discernible effect on the germination rate of the seven test plant species. However, they did exert effects on root elongation, seedling growth and biomass to different extents, indicating the potential applicability of seedling growth in the evaluation of the phytotoxicity of PAE compounds."			

Overall Quality Determination**High**

Study Citation:	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. Frontiers of Environmental Science & Engineering 9(2):259-268.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Medicago sativa</i> ; Embryo			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Photosynthesis			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2915866			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	Chemical was identified by name only	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity reported as 99.6%	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was adequate	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail	
	Metric 8: Consistency of Exposure Administration	High	exposures were administered consistently across study groups	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were justified for a dose response	
	Metric 12: Testing at or Below Solubility Limit	N/A	seeds exposed via soil	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test seeds were adequately described and were obtained from a reliable source	
	Metric 14: Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed seeds	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest	
	Metric 18: Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups	
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Study Citation:	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. Frontiers of Environmental Science & Engineering 9(2):259-268.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Medicago sativa</i> ; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Photosynthesis
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	2915866

Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
	Metric 20: Outcomes Unrelated to Exposure	Medium	Nothing reported to indicate there were differences among groups that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Section 2.7 (page 3/10) does not present the statistical tests used to perform analysis other than the program used and p-value for significance.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group and were adequate to determine values for the endpoint
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes
Additional Comments: Pigment Content”The results indicate that longer periods of cultivation of the test plants may make it easier to interpret the changes in pigment contents.”			

Overall Quality Determination**High**

Study Citation:	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. Frontiers of Environmental Science & Engineering 9(2):259-268.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Medicago sativa</i> ; Embryo			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2915866			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only.
	Metric 2:	Test Substance Source	Low	Source was reported, the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.6%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was adequate.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were justified for a dose response.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Seeds exposed via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test seeds were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed seeds.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.

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Study Citation:	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. Frontiers of Environmental Science & Engineering 9(2):259-268.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Medicago sativa</i> ; Embryo
Health Outcome:	Reproductive/Teratogenic
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	2915866

Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	Nothing reported to indicate there were differences among groups that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Section 2.7 (page 3/10) does not present the statistical tests used to perform analysis other than the program used and p-value for significance.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group and were adequate to determine values for the endpoint.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments: This form represents the germination rate results presented in Table 1 for <i>Triticum medicago sativa</i> with DBP exposure.			

Overall Quality Determination

High

Study Citation:	Deng, J., Zhang, Y., Hu, J., Jiao, J., Hu, F., Li, H., Zhang, S. (2017). Autotoxicity of phthalate esters in tobacco root exudates: Effects on seed germination and seedling growth. <i>Pedosphere</i> 27(6):1073-1082.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Nicotiana tabacum</i> ; cv K326; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5627041			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Reported as "guaranteed reagent grade"
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable
	Metric 6:	Randomized Allocation	Medium	The study reported that test containers were randomly distributed
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations. Authors reported using glassware: vials, funnels, bottles and beakers. No use of plastic vessels reported.
	Metric 8:	Consistency of Exposure Administration	Low	Only general methods of exposure administration were reported so assessment was difficult to determine
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The source of the seeds was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of test plants was not reported, three replicates used
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Environmental conditions of test system (controlled chamber) were conducive to maintenance of organism health
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Study Citation:	Deng, J., Zhang, Y., Hu, J., Jiao, J., Hu, F., Li, H., Zhang, S. (2017). Autotoxicity of phthalate esters in tobacco root exudates: Effects on seed germination and seedling growth. <i>Pedosphere</i> 27(6):1073-1082.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Nicotiana tabacum</i> ; cv K326; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Reproductive/Teratogenic
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	5627041

Domain	Metric	Rating	Comments
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23: Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained

Additional Comments: germination effects

Overall Quality Determination**High**

Study Citation:	Deng, J., Zhang, Y., Hu, J., Jiao, J., Hu, F., Li, H., Zhang, S. (2017). Autotoxicity of phthalate esters in tobacco root exudates: Effects on seed germination and seedling growth. <i>Pedosphere</i> 27(6):1073-1082.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Nicotiana tabacum</i> ; cv K326; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5627041			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	The test substance was reported as "guaranteed reagent grade."	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable.	
	Metric 6: Randomized Allocation	Medium	The study reported that test containers were randomly distributed.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations. Authors reported using glassware: vials, funnels, bottles and beakers. No use of plastic vessels was reported.	
	Metric 8: Consistency of Exposure Administration	Low	Only general methods of exposure administration were reported, so assessment was difficult to determine	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Medium	The source of the seeds was not reported.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.	
	Metric 15: Number of Organisms and Replicates per Group	Low	The number of test plants was not reported, but three replicates were used.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Medium	Environmental conditions of the test system (controlled chamber) were conducive to maintenance of organism health.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.	

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Study Citation:	Deng, J., Zhang, Y., Hu, J., Jiao, J., Hu, F., Li, H., Zhang, S. (2017). Autotoxicity of phthalate esters in tobacco root exudates: Effects on seed germination and seedling growth. <i>Pedosphere</i> 27(6):1073-1082.		
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days		
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Nicotiana tabacum</i> ; cv K326; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Development/Growth		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	5627041		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.
Additional Comments: This form is for vigor index and length.			
Overall Quality Determination		High	

Study Citation:	Jia, Z. H., Yi, J. H., Su, Y. R., Shen, H. (2011). Autotoxic substances in the root exudates from continuous tobacco cropping. Allelopathy Journal 27(1):87-96.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Terrestrial; Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Nicotinana tobacum</i> ; Hong da; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	792357			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The source was not reported.	
	Metric 3: Test Substance Purity	Low	Purity and/or grade of the test substance were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how seeds were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations. The composition of the containers that the filter papers were kept in during the experiment was not reported.	
	Metric 8: Consistency of Exposure Administration	Medium	Only general methods of exposure administration were reported, so assessment was difficult to determine.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	Low	Exposure concentrations were above the solubility limit (11.2 mg/L at 25C). The lowest concentration, 0.1 mM, corresponds to approximately 27.8 mg/L.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the test seeds was not clear.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups.	
	Metric 15: Number of Organisms and Replicates per Group	Low	The number of test organisms or replicates were not reported.	
Domain 5: Outcome Assessment				
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Study Citation:	Jia, Z. H., Yi, J. H., Su, Y. R., Shen, H. (2011). Autotoxic substances in the root exudates from continuous tobacco cropping. Allelopathy Journal 27(1):87-96.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Terrestrial; Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Nicotinana tabacum</i> ; Hong da; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	792357

Domain	Metric	Rating	Comments
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.
	Metric 17: Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported.
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments: Growth (Development-Slowed, Retarded, Delayed or Non-development, Response Site: Not reported)			

Overall Quality Determination**Low**

Study Citation:	Jia, Z. H., Yi, J. H., Su, Y. R., Shen, H. (2011). Autotoxic substances in the root exudates from continuous tobacco cropping. Allelopathy Journal 27(1):87-96.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Terrestrial; Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Nicotinana tabacum</i> ; G168; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	792357			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	Chemical was identified by name only	
	Metric 2: Test Substance Source	Low	The source was not reported.	
	Metric 3: Test Substance Purity	Low	Purity and/or grade of test substance were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes	
	Metric 6: Randomized Allocation	Low	Researchers did not report how seeds were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations. The composition of the containers the filter papers were kept in during the experiment was not reported.	
	Metric 8: Consistency of Exposure Administration	Medium	Only general methods of exposure administration were reported so assessment was difficult to determine	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type (7 days of exposure on filter paper).	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response	
	Metric 12: Testing at or Below Solubility Limit	Low	Exposure concentrations were above the solubility limit (11.2 mg/L at 25C). The lowest concentration, 0.1 mM, corresponds to approximately 27.8 mg/L.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the test seeds was not clear.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups	
	Metric 15: Number of Organisms and Replicates per Group	Low	The number of test organisms or replicates was not reported.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate	
	Metric 17: Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported	
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Study Citation:	Jia, Z. H., Yi, J. H., Su, Y. R., Shen, H. (2011). Autotoxic substances in the root exudates from continuous tobacco cropping. Allelopathy Journal 27(1):87-96.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Terrestrial; Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Nicotinana tobacum</i> ; G168; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	792357			
Domain	Metric	Rating	Comments	
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical methods were adequately described	
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group	
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes	
Additional Comments:	Growth (Development-Slowed, Retarded, Delayed or Non-development, Response Site: Not reported)			

Overall Quality Determination**Low**

Study Citation:	Jia, Z. H., Yi, J. H., Su, Y. R., Shen, H. (2011). Autotoxic substances in the root exudates from continuous tobacco cropping. Allelopathy Journal 27(1):87-96.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Terrestrial; Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Nicotinana tobacum</i> ; G168; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	792357			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance	Metric 1: Test Substance Identity	Low	Chemical was identified by name only	
	Metric 2: Test Substance Source	Low	The source was not reported.	
	Metric 3: Test Substance Purity	Low	Purity and/or grade of test substance were not reported.	
Domain 2: Test Design	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes	
	Metric 6: Randomized Allocation	Low	Researchers did not report how seeds were allocated to study groups.	
Domain 3: Exposure Characterization	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations. The composition of the containers the filter papers were kept in during the experiment was not reported.	
	Metric 8: Consistency of Exposure Administration	Medium	Only general methods of exposure administration were reported so assessment was difficult to determine	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type (7 days of exposure on filter paper).	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response	
	Metric 12: Testing at or Below Solubility Limit	Low	Exposure concentrations were above the solubility limit (11.2 mg/L at 25C). The lowest concentration, 0.1 mM, corresponds to approximately 27.8 mg/L.	
Domain 4: Test Organism	Metric 13: Test Organism Characteristics	Low	The source of the test seeds was not clear.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups	
	Metric 15: Number of Organisms and Replicates per Group	Low	The number of test organisms or replicates was not reported.	
Domain 5: Outcome Assessment	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate	
	Metric 17: Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported	
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Study Citation:	Jia, Z. H., Yi, J. H., Su, Y. R., Shen, H. (2011). Autotoxic substances in the root exudates from continuous tobacco cropping. <i>Allelopathy Journal</i> 27(1):87-96.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Terrestrial; Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Nicotinana tobacum</i> ; G168; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Reproductive/Teratogenic
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	792357

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: Germination

Overall Quality Determination	Low
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Study Citation:	Jia, Z. H., Yi, J. H., Su, Y. R., Shen, H. (2011). Autotoxic substances in the root exudates from continuous tobacco cropping. <i>Allelopathy Journal</i> 27(1):87-96.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Terrestrial; Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Nicotinana tobacum</i> ; Hong da; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	792357			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	Chemical was identified by name only	
	Metric 2: Test Substance Source	Low	The source was not reported.	
	Metric 3: Test Substance Purity	Low	Purity and/or grade of test substance were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes	
	Metric 6: Randomized Allocation	Low	Researchers did not report how seeds were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations. The composition of the containers the filter papers were kept in during the experiment was not reported.	
	Metric 8: Consistency of Exposure Administration	Medium	Only general methods of exposure administration were reported so assessment was difficult to determine	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response	
	Metric 12: Testing at or Below Solubility Limit	Low	Exposure concentrations were above the solubility limit (11.2 mg/L at 25C). The lowest concentration, 0.1 mM, corresponds to approximately 27.8 mg/L.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the test seeds was not clear.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups	
	Metric 15: Number of Organisms and Replicates per Group	Low	The number of test organisms or replicates was not reported.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate	
	Metric 17: Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported	
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Study Citation:	Jia, Z. H., Yi, J. H., Su, Y. R., Shen, H. (2011). Autotoxic substances in the root exudates from continuous tobacco cropping. <i>Allelopathy Journal</i> 27(1):87-96.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Terrestrial; Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Nicotinana tabacum</i> ; Hong da; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Reproductive/Teratogenic
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	792357

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: Germination

Overall Quality Determination	Low
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Study Citation:	Isogai, Y., Komoda, Y., Okamoto, T. (1972). Biological activities of n-butyl phthalate and its analogous compounds on various bioassays of plant growth regulators. Scientific papers of the College of General Education, University of Tokyo 22(2):129-135.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Oryza sativa</i> ; cv Kotaketamanishiki; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5551990			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	Chemical name and structure were used to identify test chemical.	
	Metric 2: Test Substance Source	Low	The source was not reported.	
	Metric 3: Test Substance Purity	Low	Purity or grade of test substance was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations and test concentrations were not measured. Glass vessels were used.	
	Metric 8: Consistency of Exposure Administration	Low	The study provided few details on exposure administration.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Medium	Only three concentrations but a wide range tested.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via culture medium.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the test seeds was not reported.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups.	
	Metric 15: Number of Organisms and Replicates per Group	Low	25 seeds were tested per treatment but number of replicates was not reported	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Number of plants per container and environmental conditions were not sufficiently reported to evaluate if adequate.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	
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Study Citation:	Isogai, Y., Komoda, Y., Okamoto, T. (1972). Biological activities of n-butyl phthalate and its analogous compounds on various bioassays of plant growth regulators. Scientific papers of the College of General Education, University of Tokyo 22(2):129-135.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Oryza sativa</i> ; cv Kotaketamanishiki; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	5551990

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Statistical analysis was not conducted.
	Metric 22: Reporting of Data	High	Results were reported in Table 2 for the control response and the exposure responses. The data was appropriate for the outcomes of interest.
	Metric 23: Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.
Additional Comments:	This portion of the study was on the plant growth promoting activity of DBP on <i>Oryza sativa</i> -cv Kotaketamanishiki. Development/Growth was selected as the outcome of interest.		

Overall Quality Determination**Medium**

Study Citation:	Isogai, Y., Komoda, Y., Okamoto, T. (1972). Biological activities of n-butyl phthalate and its analogous compounds on various bioassays of plant growth regulators. Scientific papers of the College of General Education, University of Tokyo 22(2):129-135.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Oryza sativa</i> ; cv Tanginbozu; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5551990			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	Chemical name and structure were used to identify test chemical.	
	Metric 2: Test Substance Source	Low	The source was not reported.	
	Metric 3: Test Substance Purity	Low	Purity or grade of test substance was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations and test concentrations were not measured. Glass vessels were used.	
	Metric 8: Consistency of Exposure Administration	Low	The study provided few details on exposure administration.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Medium	Only three concentrations but a wide range tested.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via culture medium.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the test seeds was not reported.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups.	
	Metric 15: Number of Organisms and Replicates per Group	Low	25 seeds per treatment but number of replicates was not reported.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Number of plants per container and environmental conditions were not sufficiently reported to evaluate if adequate.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.	

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Study Citation:	Isogai, Y., Komoda, Y., Okamoto, T. (1972). Biological activities of n-butyl phthalate and its analogous compounds on various bioassays of plant growth regulators. Scientific papers of the College of General Education, University of Tokyo 22(2):129-135.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Oryza sativa</i> ; cv Tanginbozu; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	5551990

Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Statistical analysis was not conducted.
	Metric 22: Reporting of Data	High	Results were reported in Table 2 for the control response and the exposure responses. The data was appropriate for the outcomes of interest.
	Metric 23: Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.
Additional Comments:	This portion of the study was on the plant growth promoting activity of DBP on <i>Oryza sativa</i> -cv Tanginbozu. Development/Growth was selected as the outcome of interest.		

Overall Quality Determination**Medium**

Study Citation:	Isogai, Y., Komoda, Y., Okamoto, T. (1972). Biological activities of n-butyl phthalate and its analogous compounds on various bioassays of plant growth regulators. Scientific papers of the College of General Education, University of Tokyo 22(2):129-135.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Oryza sativa</i> ; cv Norin #29; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5551990			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical name and structure were used to identify the test chemical.
	Metric 2:	Test Substance Source	Low	The source was not reported.
	Metric 3:	Test Substance Purity	Low	Purity or grade of the test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using a concurrent negative control group.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations and test concentrations were not measured. Glass vessels were used.
	Metric 8:	Consistency of Exposure Administration	Low	The study provided few details on exposure administration.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type.
	Metric 11:	Number of Exposure Groups/Spacing of Exposure Levels	Medium	Only three concentrations were used, but a wide range was tested.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via culture medium.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test seeds was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups.
	Metric 15:	Number of Organisms and Replicates per Group	Low	There were 25 seeds tested per treatment, but the number of replicates was not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Number of plants per container and environmental conditions were not sufficiently reported to evaluate if adequate.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.

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Study Citation:	Isogai, Y., Komoda, Y., Okamoto, T. (1972). Biological activities of n-butyl phthalate and its analogous compounds on various bioassays of plant growth regulators. Scientific papers of the College of General Education, University of Tokyo 22(2):129-135.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Oryza sativa</i> ; cv Norin #29; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	5551990

Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	Low	Statistical analysis was not conducted.
Metric 22:	Reporting of Data	High	Results were reported in Table 1 for the control response and the exposure responses. The data was appropriate for the outcomes of interest.
Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.
Additional Comments:	This portion of the study was on the plant growth promoting activity of DBP on <i>Oryza sativa</i> -cv Norin #29. Development/Growth was selected as the outcome of interest.		

Overall Quality Determination**Medium**

Study Citation:	Isogai, Y., Komoda, Y., Okamoto, T. (1972). Biological activities of n-butyl phthalate and its analogous compounds on various bioassays of plant growth regulators. Scientific papers of the College of General Education, University of Tokyo 22(2):129-135.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Oryza sativa</i> ; cv Norin #22; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5551990			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	Chemical name and structure were used to identify test chemical.	
	Metric 2: Test Substance Source	Low	The source was not reported.	
	Metric 3: Test Substance Purity	Low	Purity or grade of test substance was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations and test concentrations were not measured. Glass vessels were used.	
	Metric 8: Consistency of Exposure Administration	Low	The study provided few details on exposure administration.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Medium	Only three concentrations but a wide range tested.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via culture medium.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the test seeds was not reported.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups.	
	Metric 15: Number of Organisms and Replicates per Group	Low	25 seeds were tested per treatment but number of replicates was not reported.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Number of plants per container and environmental conditions were not sufficiently reported to evaluate if adequate.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.	

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Study Citation:	Isogai, Y., Komoda, Y., Okamoto, T. (1972). Biological activities of n-butyl phthalate and its analogous compounds on various bioassays of plant growth regulators. Scientific papers of the College of General Education, University of Tokyo 22(2):129-135.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Oryza sativa</i> ; cv Norin #22; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	5551990

Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Statistical analysis was not conducted.
	Metric 22: Reporting of Data	High	Results were reported in Table 1 for the control response and the exposure responses. The data was appropriate for the outcomes of interest.
	Metric 23: Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.
Additional Comments:	This portion of the study was on the plant growth promoting activity of DBP on <i>Oryza sativa</i> -cv Norin #22. Development/Growth was selected as the outcome of interest.		

Overall Quality Determination**Medium**

Study Citation:	Isogai, Y., Komoda, Y., Okamoto, T. (1972). Biological activities of n-butyl phthalate and its analogous compounds on various bioassays of plant growth regulators. Scientific papers of the College of General Education, University of Tokyo 22(2):129-135.			
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days			
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Oryza sativa</i> ; cv Kinmase; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5551990			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	Chemical name and structure were used to identify test chemical.	
	Metric 2: Test Substance Source	Low	The source was not reported.	
	Metric 3: Test Substance Purity	Low	Purity or grade of test substance was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations and test concentrations were not measured. Glass vessels were used.	
	Metric 8: Consistency of Exposure Administration	Low	The study provided few details on exposure administration.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was suitable for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Medium	Only three concentrations but a wide range tested.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via culture medium.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the test seeds was not reported.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups.	
	Metric 15: Number of Organisms and Replicates per Group	Low	25 seeds were tested per treatment but number of replicates was not reported.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Number of plants per container and environmental conditions were not sufficiently reported to evaluate if adequate.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited.	

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Study Citation:	Isogai, Y., Komoda, Y., Okamoto, T. (1972). Biological activities of n-butyl phthalate and its analogous compounds on various bioassays of plant growth regulators. Scientific papers of the College of General Education, University of Tokyo 22(2):129-135.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Oryza sativa</i> ; cv Kinmase; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	5551990

Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	Low	Statistical analysis was not conducted.
Metric 22:	Reporting of Data	High	Results were reported in Table 1 for the control response and the exposure responses. The data was appropriate for the outcomes of interest.
Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.
Additional Comments:	This portion of the study was on the plant growth promoting activity of DBP on <i>Oryza sativa</i> -cv Kinmase. Development/Growth was selected as the outcome of interest.		

Overall Quality Determination**Medium**

Study Citation:	Chuah, T. S., Oh, H. Y., Habsah, M., Norhafizah, M. Z., Ismail, B. S. (2014). Potential of crude extract and isolated compounds from golden beard grass (Chrysopogon serrulatus) for control of sprangletop (Leptochloa chinensis) in aerobic rice systems. Crop and Pasture Science 65(5):461-469.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Oryza sativa</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5432995			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	PESTANAL analytic standard grade was used.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using a negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations.	
	Metric 8: Consistency of Exposure Administration	Medium	There was no mention of irregularities in exposure administration.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and exposure frequency were reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	N/A	Three treatments over an acceptable range were tested.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via soil.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups.	
	Metric 15: Number of Organisms and Replicates per Group	Low	There was one rice seed per cup, with five replicates per exposure, and the experiment was repeated twice.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	
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Study Citation:	Chuah, T. S., Oh, H. Y., Habsah, M., Norhafizah, M. Z., Ismail, B. S. (2014). Potential of crude extract and isolated compounds from golden beard grass (<i>Chrysopogon serrulatus</i>) for control of sprangletop (<i>Leptochloa chinensis</i>) in aerobic rice systems. <i>Crop and Pasture Science</i> 65(5):461-469.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Oryza sativa</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	5432995

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	Medium	Data was pooled for application timing of seedlings.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This form is for Table 5.

Overall Quality Determination**Medium**

Study Citation:	Dueck, T. A., Dijk, Van, C. J., David, F., Scholz, N., Vanwalleghe, F. (2003). Chronic effects of vapour phase di-n-butyl phthalate (DBP) on six plant species. Chemosphere 53(8):911-920.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Phaseolus vulgaris</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1302103			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The chemical was identified by name and CAS#.
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity or grade of the test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Study authors reported using an appropriate concurrent negative control group, note that all controls had some DBP detected. Control group DBP concentrations are reported in Table 1.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail. The chambers were constructed of "hardened glass" and aluminum.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups based on timed samplings.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies. Samples were taken frequently: "Actual DBP concentrations were measured by taking air samples from each chamber using an automated gas sampling device (GS-1 gas sampler, Gerstel, M €ulheim, Germany). Sampling was performed with Tenax TA tubes (2,6-diphenylphenylene oxide polymer) at 250ml min ¹ for 20 min. During the fumigation experiment, each chamber was sampled in duplicate on a weekly basis during the complete exposure period (76 days)."
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	The number of exposure groups and spacing of exposure levels were suitable. Concentration gradient is reported as both actual and nominal within table 1.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via fumigation in a continually renewed chamber.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The crop and herbaceous species originated from germinated seeds, but source was not explicitly reported.

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Study Citation:	Dueck, T. A., Dijk, Van, C. J., David, F., Scholz, N., Vanwalleghem, F. (2003). Chronic effects of vapour phase di-n-butyl phthalate (DBP) on six plant species. Chemosphere 53(8):911-920.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Phaseolus vulgaris</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1302103			
Domain	Metric	Rating	Comments	
	Metric 14: Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions.	
	Metric 15: Number of Organisms and Replicates per Group	Low	There were four plants per treatment and no replicates reported. "Three to four weeks after sowing, when the first mature leaves appeared, four uniform individuals per species including the tree species were placed in each fumigation chamber (the maximum number physically possible) for exposure to DBP."	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	
	Metric 18: Consistency of Outcome Assessment	Medium	Outcomes were assessed consistently across study groups but few details were reported.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.	
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.	
	Metric 23: Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.	
Additional Comments:	This form represents growth outcomes associated with Dry Weight reported for Phaseolus shoot and roots within Figure 3 on page 6/10.			
Overall Quality Determination		High		

Study Citation:	Dueck, T. A., Dijk, Van, C. J., David, F., Scholz, N., Vanwalleghem, F. (2003). Chronic effects of vapour phase di-n-butyl phthalate (DBP) on six plant species. Chemosphere 53(8):911-920.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Picea abies</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1302103			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The chemical was identified by name and CAS#.
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity or grade of the test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Study authors reported using an appropriate concurrent negative control group, note that all controls had some DBP detected. Control group DBP concentrations are reported in Table 1.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail. The chambers were constructed of "hardened glass" and aluminum.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups based on timed samplings.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies. Samples were taken frequently: "Actual DBP concentrations were measured by taking air samples from each chamber using an automated gas sampling device (GS-1 gas sampler, Gerstel, M €ulheim, Germany). Sampling was performed with Tenax TA tubes (2,6-diphenylphenylene oxide polymer) at 250ml min ⁻¹ for 20 min. During the fumigation experiment, each chamber was sampled in duplicate on a weekly basis during the complete exposure period (76 days)."
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	The number of exposure groups and spacing of exposure levels were suitable. Concentration gradient is reported as both actual and nominal within table 1.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via fumigation in a continually renewed chamber.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The source of the test organisms (Picea abies) was from a local nursery. The crop and herbaceous species originated from germinated seeds.

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Study Citation:	Dueck, T. A., Dijk, Van, C. J., David, F., Scholz, N., Vanwalleghe, F. (2003). Chronic effects of vapour phase di-n-butyl phthalate (DBP) on six plant species. Chemosphere 53(8):911-920.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Picea abies</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	ADME (biotransformation)
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	1302103

Domain	Metric	Rating	Comments
	Metric 14: Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions.
	Metric 15: Number of Organisms and Replicates per Group	Low	There were four plants per treatment and no replicates reported. "Three to four weeks after sowing, when the first mature leaves appeared, four uniform individuals per species including the tree species were placed in each fumigation chamber (the maximum number physically possible) for exposure to DBP."
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18: Consistency of Outcome Assessment	Medium	Outcomes were assessed consistently across study groups but few details were reported.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group in Table 2.
	Metric 23: Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.
Additional Comments: This form is for ADME (biotransformation) assessment of DBP concentration in leaf tissue.			

Overall Quality Determination**High**

Study Citation:	Dueck, T. A., Dijk, Van, C. J., David, F., Scholz, N., Vanwalleghem, F. (2003). Chronic effects of vapour phase di-n-butyl phthalate (DBP) on six plant species. Chemosphere 53(8):911-920.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Picea abies</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1302103			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The chemical was identified by name and CAS#.
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity or grade of the test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Study authors reported using an appropriate concurrent negative control group, note that all controls had some DBP detected. Control group DBP concentrations are reported in Table 1.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail. The chambers were constructed of "hardened glass" and aluminum.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups based on timed samplings.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies. Samples were taken frequently: "Actual DBP concentrations were measured by taking air samples from each chamber using an automated gas sampling device (GS-1 gas sampler, Gerstel, M €ulheim, Germany). Sampling was performed with Tenax TA tubes (2,6-diphenylphenylene oxide polymer) at 250ml min ¹ for 20 min. During the fumigation experiment, each chamber was sampled in duplicate on a weekly basis during the complete exposure period (76 days)."
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	The number of exposure groups and spacing of exposure levels were suitable. Concentration gradient is reported as both actual and nominal within table 1.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via fumigation in a continually renewed chamber.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The source of the test organisms (Picea abies) was from a local nursery. The crop and herbaceous species originated from germinated seeds.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions.
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Study Citation:	Dueck, T. A., Dijk, Van, C. J., David, F., Scholz, N., Vanwalleggem, F. (2003). Chronic effects of vapour phase di-n-butyl phthalate (DBP) on six plant species. Chemosphere 53(8):911-920.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Picea abies</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	1302103

Domain	Metric	Rating	Comments
	Metric 15: Number of Organisms and Replicates per Group	Low	There were four plants per treatment and no replicates reported. "Three to four weeks after sowing, when the first mature leaves appeared, four uniform individuals per species including the tree species were placed in each fumigation chamber (the maximum number physically possible) for exposure to DBP."
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18: Consistency of Outcome Assessment	Medium	Outcomes were assessed consistently across study groups, but few details were reported.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23: Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.
Additional Comments: This form represents growth outcomes associated with Dry Weight reported for Picea within Figure 3 on page 6/10.			

Overall Quality Determination**High**

Study Citation:	Dueck, T. A., Dijk, Van, C. J., David, F., Scholz, N., Vanwalleghe, F. (2003). Chronic effects of vapour phase di-n-butyl phthalate (DBP) on six plant species. Chemosphere 53(8):911-920.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Plantago major</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1302103			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The chemical was identified by name and CAS#.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Low	Purity or grade of the test substance were not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	Low	Study authors reported using an appropriate concurrent negative control group, note that all controls had some DBP detected. Control group DBP concentrations are reported in Table 1.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail. The chambers were constructed of "hardened glass" and aluminum.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups based on timed samplings.	
	Metric 9: Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies. Samples were taken frequently: "Actual DBP concentrations were measured by taking air samples from each chamber using an automated gas sampling device (GS-1 gas sampler, Gerstel, M €ulheim, Germany). Sampling was performed with Tenax TA tubes (2,6-diphenylphenylene oxide polymer) at 250ml min1 for 20 min. During the fumigation experiment, each chamber was sampled in duplicate on a weekly basis during the complete exposure period (76 days)."	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Medium	The number of exposure groups and spacing of exposure levels were suitable. Concentration gradient is reported as both actual and nominal within table 1.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via fumigation in a continually renewed chamber.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The crop and herbaceous species originated from germinated seeds, but source was not explicitly reported.	
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Study Citation:	Dueck, T. A., Dijk, Van, C. J., David, F., Scholz, N., Vanwalleghem, F. (2003). Chronic effects of vapour phase di-n-butyl phthalate (DBP) on six plant species. Chemosphere 53(8):911-920.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Plantago major</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1302103			
Domain	Metric	Rating	Comments	
	Metric 14: Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions.	
	Metric 15: Number of Organisms and Replicates per Group	Low	There were four plants per treatment and no replicates reported. "Three to four weeks after sowing, when the first mature leaves appeared, four uniform individuals per species including the tree species were placed in each fumigation chamber (the maximum number physically possible) for exposure to DBP."	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	
	Metric 18: Consistency of Outcome Assessment	Medium	Outcomes were assessed consistently across study groups but few details were reported.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.	
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.	
	Metric 23: Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.	
Additional Comments:	This form represents growth outcomes associated with Dry Weight reported for Plantago shoot and roots within Figure 3 on page 6/10.			

Overall Quality Determination**High**

Study Citation:	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. Frontiers of Environmental Science & Engineering 9(2):259-268.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Raphanus sativus</i> ; Embryo			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2915866			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	Chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	Source was reported, the test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity reported as 99.6%	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was adequate.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were justified for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Seeds exposed via soil.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test seeds were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed seeds.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	

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Study Citation:	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. Frontiers of Environmental Science & Engineering 9(2):259-268.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Raphanus sativus</i> ; Embryo
Health Outcome:	Reproductive/Teratogenic
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	2915866

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	Nothing reported to indicate there were differences among groups that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Section 2.7 (page 3/10) does not present the statistical tests used to perform analysis other than the program used and p-value for significance.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group and were adequate to determine values for the endpoint.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This form represents the germination rate results presented in Table 1 for *Raphanus sativus* with DBP exposure.

Overall Quality Determination**High**

Study Citation:	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. Frontiers of Environmental Science & Engineering 9(2):259-268.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Raphanus sativus</i> ; Embryo			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Photosynthesis			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2915866			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity was reported as 99.6%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was adequate.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were justified for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Seeds were exposed via soil.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test seeds were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed seeds.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.	
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Study Citation:	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. Frontiers of Environmental Science & Engineering 9(2):259-268.		
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)		
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Raphanus sativus</i> ; Embryo		
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Photosynthesis		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	2915866		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
Metric 20:	Outcomes Unrelated to Exposure	Medium	Nothing was reported to indicate there were differences among groups that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	Low	Section 2.7 (page 3/10) does not present the statistical tests used to perform analysis other than the program used and p-value for significance.
Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group and were adequate to determine values for the endpoint.
Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments:	This is for pigment content."The results indicate that longer periods of cultivation of the test plants may make it easier to interpret the changes in pigment contents."		
Overall Quality Determination		High	

Study Citation:	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. Frontiers of Environmental Science & Engineering 9(2):259-268.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Raphanus sativus</i> ; Embryo			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2915866			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only.
	Metric 2:	Test Substance Source	Low	Source was reported, the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.6%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was adequate.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were justified for a dose response.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Seeds exposed via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test seeds were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed seeds.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.

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Study Citation:	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. Frontiers of Environmental Science & Engineering 9(2):259-268.		
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)		
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Raphanus sativus</i> ; Embryo		
Health Outcome:	Development/Growth		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	2915866		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	Nothing reported to indicate there were differences among groups that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Section 2.7 (page 3/10) does not present the statistical tests used to perform analysis other than the program used and p-value for significance.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group and were adequate to determine values for the endpoint.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments:	"DnBP and DEHP at a range of concentrations in the experimental soil showed no discernible effect on the germination rate of the seven test plant species. However, they did exert effects on root elongation, seedling growth and biomass to different extents, indicating the potential applicability of seedling growth in the evaluation of the phytotoxicity of PAE compounds."		
Overall Quality Determination		High	

Study Citation:	Virgin, H. I., A-M, Holst, Morner, J. (1981). Effect of di-n-butylphthalate on the carotenoid synthesis in green plants. Physiologia Plantarum 53(2):158-163.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Raphanus sativus</i> ; Cherry Belle; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1333234			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	High	The test substance identity was analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Medium	pro analysis quality was reported
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Untreated plants were used as controls but few details were provided
	Metric 5:	Negative Control Response	Low	The biological response of the negative control groups was not reported.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	Several methods of obtaining vapors was reported but effects were not attributed to a particular method
	Metric 8:	Consistency of Exposure Administration	Low	Difficult to determine as durations and concentrations were not clearly reported
	Metric 9:	Measurement of Test Substance Concentration	Medium	Exposure concentrations were measured but not clearly reported
	Metric 10:	Exposure Duration and Frequency	Low	Exposures seemed to be around 4-7 days but study length was not clearly reported
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Uninformative	No information is provided on the number of exposure groups, the range seems to be 10-150 ng/L but unclear if this is one treatment that varied over time or if there were multiple treatment levels
	Metric 12:	Testing at or Below Solubility Limit	N/A	exposure was via vapors
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test plants was not reported, age or stage at test initiation was not clear.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized
	Metric 15:	Number of Organisms and Replicates per Group	Low	Numbers and replicates were not reported
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported

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Study Citation:	Virgin, H. I., A-M, Holst, Morner, J. (1981). Effect of di-n-butylphthalate on the carotenoid synthesis in green plants. Physiologia Plantarum 53(2):158-163.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Raphanus sativus</i> ; Cherry Belle; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mortality			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1333234			
Domain		Metric	Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were confusing
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	N/A	Statistical analysis was not possible, information was not quantified
	Metric 22:	Reporting of Data	Low	There was brief mention of plant mortality in the results section text. No data was shown.
	Metric 23:	Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability
Additional Comments:	closed system, light was a variable			

Overall Quality Determination**Uninformative**

Study Citation:	Virgin, H. I., A-M, Holst, Morner, J. (1981). Effect of di-n-butylphthalate on the carotenoid synthesis in green plants. Physiologia Plantarum 53(2):158-163.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Raphanus sativus</i> ; Cherry Belle; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Photosynthesis			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1333234			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	High	The test substance identity was analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Medium	Pro analysis quality was reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	Low	Untreated plants were used as controls, but few details were provided.	
	Metric 5: Negative Control Response	Low	The biological response of the negative control group was not reported.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	Several methods of obtaining vapors were reported, but effects were not attributed to a particular method.	
	Metric 8: Consistency of Exposure Administration	Low	The consistency of exposure was difficult to determine as durations and concentrations were not clearly reported.	
	Metric 9: Measurement of Test Substance Concentration	Medium	Exposure concentrations were measured but not clearly reported.	
	Metric 10: Exposure Duration and Frequency	Low	Exposures seemed to be around 4-7 days but study length was not clearly reported.	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	Uninformative	No information is provided on the number of exposure groups, the range seems to be 10-150 ng/L, but it's unclear if this is one treatment that varied over time or if there were multiple treatment levels.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via vapors.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the test plants was not reported, and age or stage at test initiation was not clear.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.	
	Metric 15: Number of Organisms and Replicates per Group	Low	Numbers of organisms and replicates were not reported.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.	
	Metric 17: Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported.	
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were confusing.	

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Study Citation:	Virgin, H. I., A-M, Holst, Morner, J. (1981). Effect of di-n-butylphthalate on the carotenoid synthesis in green plants. <i>Physiologia Plantarum</i> 53(2):158-163.		
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported		
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Raphanus sativus</i> ; Cherry Belle; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Mechanistic-Photosynthesis		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	1333234		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	N/A	Statistical analysis was not possible, and information was not quantified.
Metric 22:	Reporting of Data	Low	Data were only reported for some outcomes.
Metric 23:	Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability.
Additional Comments: The test system was closed, and light was a variable.			

Overall Quality Determination**Uninformative**

Study Citation:	Virgin, H. I., A-M, Holst, Morner, J. (1981). Effect of di-n-butylphthalate on the carotenoid synthesis in green plants. Physiologia Plantarum 53(2):158-163.			
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Raphanus sativus</i> ; Cherry Belle; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Cytotoxicity-Photosynthesis			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1333234			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	Chemical was identified by name only	
	Metric 2: Test Substance Source	High	The test substance identity was analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	Medium	pro analysis quality was reported	
Domain 2: Test Design				
	Metric 4: Negative Controls	Low	Untreated plants were used as controls but few details were provided	
	Metric 5: Negative Control Response	Low	The biological response of the negative control groups was not reported.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Medium	Several methods of obtaining vapors was reported but effects were not attributed to a particular method	
	Metric 8: Consistency of Exposure Administration	Low	Difficult to determine as durations and concentrations were not clearly reported	
	Metric 9: Measurement of Test Substance Concentration	Medium	Exposure concentrations were measured but not clearly reported	
	Metric 10: Exposure Duration and Frequency	Low	Exposures seemed to be around 4-7 days but study length was not clearly reported	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Uninformative	No information is provided on the number of exposure groups, the range seems to be 10-150 ng/L but unclear if this is one treatment that varied over time or if there were multiple treatment levels	
	Metric 12: Testing at or Below Solubility Limit	N/A	exposure was via vapors	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	The source of the test plants was not reported, age or stage at test initiation was not clear.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized	
	Metric 15: Number of Organisms and Replicates per Group	Low	Numbers and replicates were not reported	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate	
	Metric 17: Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported	
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were confusing	

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Study Citation:	Virgin, H. I., A-M, Holst, Morner, J. (1981). Effect of di-n-butylphthalate on the carotenoid synthesis in green plants. <i>Physiologia Plantarum</i> 53(2):158-163.		
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported		
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Raphanus sativus</i> ; Cherry Belle; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Mechanistic-Cytotoxicity-Photosynthesis		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	1333234		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	N/A	Statistical analysis was not possible, information was not quantified
Metric 22:	Reporting of Data	Low	Data were only reported for some outcomes
Metric 23:	Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability
Additional Comments: open system, light was a variable			

Overall Quality Determination**Uninformative**

Study Citation:	Lã,Kke, H., Rasmussen, L. (1983). Phytotoxicological effects of Di-(2-ethyl hexyl)-phthalate and Di-n-butyl-phthalate on higher plants in laboratory and field experiments. Environmental Pollution Series A: Ecological and Biological 32(3):179-199.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Sinapis Alba</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Skin & Connective Tissue			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	9430481			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemicals were identified as di-(2-ethyl hexyl)-phthalate (DEHP) and di-(n-butyl)-phthalate (DBP). No CASRN was provided.
	Metric 2:	Test Substance Source	High	DBP was sourced from Fluka, Switzerland, and DEHP was sourced from Scandiflex, Ltd., Denmark. Analytical verification was conducted.
	Metric 3:	Test Substance Purity	High	DPB purity was >99.5% and DEHP purity was >99%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Negative controls included solvent-treated (acetone (5%) + water (95%) + Tween 20 (0.1%)) plots.
	Metric 5:	Negative Control Response	Medium	The biological response of the negative control groups is shown in Table 2 as no injury observed.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The plants were treated with a 2.5 m two-person operated pneumatic sprayer mounted with five flat spray nozzles with constant pressure applied to give a known amount of liquid per unit time at 35 mL per square meter. Chemicals were prepared in solvent as described in the laboratory experiments.
	Metric 8:	Consistency of Exposure	Low	It is unclear what plots were treated at which times.
	Metric 9:	Administration Measurement of Test Substance Concentration	Medium	The test concentrations were measured in the leaves in Sinapis at 1 and 3-hr post-application.
	Metric 10:	Exposure Duration and Frequency	Medium	It was applied one time with a solvent. It was reported that a thunderstorm 20 hours after treatment may have ended the exposure.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Exposure concentrations are shown in Table 2.
	Metric 12:	Testing at or Below Solubility Limit	Low	Reporting omissions prevented determination of whether exposure concentrations exceeded the water solubility limit. Application concentrations were presented in terms of mL per square meter. A solvent was used, so this likely enhanced the solubility of the test material.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The test species were Brassica and Sinapis. No source was provided for the seeds.

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Study Citation:	Lã Kke, H., Rasmussen, L. (1983). Phytotoxicological effects of Di-(2-ethyl hexyl)-phthalate and Di-n-butyl-phthalate on higher plants in laboratory and field experiments. Environmental Pollution Series A: Ecological and Biological 32(3):179-199.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Sinapis Alba</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Skin & Connective Tissue			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	9430481			
Domain	Metric	Rating	Comments	
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions and all pretreatment conditions were the same for control and exposed organisms, such that the only difference was exposure to test substance. The number of test organisms and/or replicates was not reported.
	Metric 15:	Number of Organisms and Replicates per Group	Low	
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reporting of housing and/or environmental conditions and/or food, water, and nutrients and/or biomass loading was not sufficiently reported to evaluate if adequate and whether differences occurred between control and exposed populations.
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported.
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment (e.g., timing of assessment across groups) were confusing, limited, or not reported.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure (e.g., infection) that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Uninformative	No statistical analysis was conducted.
	Metric 22:	Reporting of Data	Low	There was minimal reporting of data in the text of the study (data for most treatment groups was not described). Data in Table 2 was represented as +/-.
	Metric 23:	Explanation of Unexpected Outcomes	Low	Insufficient information was provided to determine if excessive variability or unexpected outcomes occurred. Authors reported thunderstorms approximately one day after spraying both species.
Additional Comments:	This form applies to both Sinapis and Brassica. DEHP, DBP, or a mixture was sprayed onto plants in plots at doses described in Table 2. Concentrations of chemical on plant measured at 1 and 3 hours post spraying. Injury described as chlorotic spots. Results in the text were not well described/quantified. Table 2 reports injury as a +/- (not quantified).			

Overall Quality Determination**Uninformative**

Study Citation:	Løkke, H., Bro-Rasmussen, F. (1981). Studies of mobility of di-iso-butyl phthalate (DiBP), di-N-butyl phthalate (DBP), and di-(2-ethyl hexyl) phthalate (DEHP) by plant foliage treatment in a closed terrestrial simulation chamber. Chemosphere 10(11-12):1223-1236.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Water; Dermal (topical application)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Sinapsis alba</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	680337			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
Metric 1:	Test Substance Identity	Low	Identified by nomenclature (Di-n-butyl phthalic acid ester). No other identifying information provided.	
Metric 2:	Test Substance Source	High	Fluke AG, CH-9470 Buchs, Switzerland	
Metric 3:	Test Substance Purity	High	>99.5% purity.	
Domain 2: Test Design				
Metric 4:	Negative Controls	High	Negative & positive (carrier) controls were present.	
Metric 5:	Negative Control Response	Low	Biological response of negative controls not reported.	
Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
Metric 7:	Experimental System/Test Media Preparation	High	Experimental system & test media were reported in detail.	
Metric 8:	Consistency of Exposure Administration	Low	Exposures were administered consistently, but spraying efficiency was "estimated to be 65-75% for <i>Sinapis alba</i> and <i>Lapsana communis</i> , and 40-50% for <i>Achillea millefolium</i> " with no further information given. Therefore, exposure was inconsistent among & within the study groups.	
Metric 9:	Measurement of Test Substance Concentration	Low	Given doses for the DBP experiment (1.5, 0.25, and 0.05 ug/ cm2 respectively of the whole leaf area) were not analytically verified & due to the spray efficiency concerns noted in Metric 8 cannot be expected to be similar to nominal concentrations.	
Metric 10:	Exposure Duration and Frequency	High	Single exposure was performed with monitoring for 15 days.	
Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Response was only found at the highest dose and there was a closely-spaced dose below that, indicating that lower doses are low enough to establish a NOEC/LOEC.	
Metric 12:	Testing at or Below Solubility Limit	High	No biological responses found in the solvent controls.	
Domain 4: Test Organism				
Metric 13:	Test Organism Characteristics	Low	Source of test species was not reported.	
Metric 14:	Acclimatization and Pretreatment Conditions	Low	Plants were not acclimatized to the elaborate climate chamber before the experiment began, and were instead reared outside it and placed inside immediately upon exposure. This chamber may have had an impact on the results.	
Metric 15:	Number of Organisms and Replicates per Group	Low	8 plants were used per group, with no replication.	

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Study Citation:	Løkke, H., Bro-Rasmussen, F. (1981). Studies of mobility of di-iso-butyl phthalate (DiBP), di-N-butyl phthalate (DBP), and di-(2-ethyl hexyl) phthalate (DEHP) by plant foliage treatment in a closed terrestrial simulation chamber. Chemosphere 10(11-12):1223-1236.		
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 0 - 4 days (0-96h)		
Exposure Route, Media, Path:	Terrestrial; Water; Dermal (topical application)		
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Sinapsis alba</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	ADME (biotransformation)		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	680337		
Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
Metric 16:	Adequacy of Test Conditions	High	Organism housing characteristics were described in detail & were appropriate for the rearing of plants.
Metric 17:	Outcome Assessment Methodology	Medium	The assessment methodology was not described. It was simply stated that "chlorosis developed in the new leaves".
Metric 18:	Consistency of Outcome Assessment	Low	Details regarding outcome assessment were not reported.
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	Uninformative	The DBP-treated plants were housed in the same sealed chamber as plants treated with DEHP and DiBP, virtually guaranteeing cross-contamination. The controls were separated from the other plants.
Metric 20:	Outcomes Unrelated to Exposure	High	No outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	N/A	Statistical analysis was not performed.
Metric 22:	Reporting of Data	Low	Data for control groups was not described in the text (was there any chlorosis in non-treated groups?)
Metric 23:	Explanation of Unexpected Outcomes	High	No unexplained outcomes.
Additional Comments:	The only study reported in this paper that was PECO-relevant is the exposure to DBP in <i>Sinapis alba</i> – the other experiments detailed are mixture studies where DBP, DiBP, and DEHP were applied. The DBP-alone exposure DBP residues in plants as a function of time, and observed chlorosis as the apical endpoint. This study had several methodological flaws, including housing DBP-treated plants in the same sealed chamber as plants treated with other phthalates, virtually guaranteeing cross-contamination. This form is for the ADME outcome.		
Overall Quality Determination		Uninformative	

Study Citation:	Løkke, H., Bro-Rasmussen, F. (1981). Studies of mobility of di-iso-butyl phthalate (DiBP), di-N-butyl phthalate (DBP), and di-(2-ethyl hexyl) phthalate (DEHP) by plant foliage treatment in a closed terrestrial simulation chamber. Chemosphere 10(11-12):1223-1236.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Water; Dermal (topical application)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Sinapsis alba</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	680337			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The test substance was identified by nomenclature (Di-n-butyl phthalic acid ester). No other identifying information was provided.
	Metric 2:	Test Substance Source	High	The source was Fluke AG, CH-9470 Buchs, Switzerland.
	Metric 3:	Test Substance Purity	High	The purity was reported as >99.5%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Negative & positive (carrier) controls were present.
	Metric 5:	Negative Control Response	Low	Biological response of negative controls was not reported.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system & test media were reported in detail.
	Metric 8:	Consistency of Exposure Administration	Low	Exposures were administered consistently, but spraying efficiency was ”estimated to be 65-75% for Sinapis alba and Lapsana communis, and 40-50% for Achillea millefol- lum” with no further information given. Therefore, exposure was inconsistent among & within the study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Given doses for the DBP experiment (1.5, 0.25, and 0.05 ug/ cm2 respectively of the whole leaf area) were not analytically verified & due to the spray efficiency concerns noted in Metric 8, they cannot be expected to be similar to nominal concentrations.
	Metric 10:	Exposure Duration and Frequency	High	Single exposure was performed with monitoring for 15 days.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Response was only found at the highest dose and there was a closely-spaced dose below that, indicating that lower doses are low enough to establish a NOEC/LOEC.
	Metric 12:	Testing at or Below Solubility Limit	High	No biological responses were found in the solvent controls.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	Source of test species was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	Plants were not acclimatized to the elaborate climate chamber before the experiment began, and were instead reared outside it and placed inside immediately upon exposure. This chamber may have had an impact on the results.
	Metric 15:	Number of Organisms and Replicates per Group	Low	Eight plants were used per group, with no replication.
Domain 5: Outcome Assessment				
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Study Citation:	Løkke, H., Bro-Rasmussen, F. (1981). Studies of mobility of di-iso-butyl phthalate (DiBP), di-N-butyl phthalate (DBP), and di-(2-ethyl hexyl) phthalate (DEHP) by plant foliage treatment in a closed terrestrial simulation chamber. Chemosphere 10(11-12):1223-1236.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Water; Dermal (topical application)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Sinapsis alba</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	680337			
Domain	Metric	Rating	Comments	
	Metric 16: Adequacy of Test Conditions	High	Organism housing characteristics were described in detail & were appropriate for the rearing of plants.	
	Metric 17: Outcome Assessment Methodology	Medium	The assessment methodology was not described. It was simply stated that "chlorosis developed in the new leaves."	
	Metric 18: Consistency of Outcome Assessment	Low	Details regarding outcome assessment were not reported.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Uninformative	The DBP-treated plants were housed in the same sealed chamber as plants treated with DEHP and DiBP, virtually guaranteeing cross-contamination. The controls were separated from the other plants.	
	Metric 20: Outcomes Unrelated to Exposure	High	There were no outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	N/A	Statistical analysis was not performed.	
	Metric 22: Reporting of Data	Low	Data for control groups was not described in the text (it is unknown if there was any chlorosis in the non-treated groups).	
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexplained outcomes.	
Additional Comments:	The only study reported in this paper that was PECO-relevant is the exposure to DBP in Sinapis alba – the other experiments detailed are mixture studies where DBP, DiBP, and DEHP were applied. The DBP-alone exposure used DBP residues in plants as a function of time, and observed chlorosis as the apical endpoint. This study had several methodological flaws, including housing DBP-treated plants in the same sealed chamber as plants treated with other phthalates, virtually guaranteeing cross-contamination. This form is for chlorosis.			
Overall Quality Determination		Uninformative		

Study Citation:	Kannan, S. (1986). Effects of dibutyl phthalate and phthalic-acid on chlorosis recovery in iron-deficiency stressed sorghum cultivars. Journal of Plant Nutrition 9(12):1543-1551.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Sorghum bicolor</i> ; cv. CSH-5; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5433174			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	Low	The source was not reported.
	Metric 3:	Test Substance Purity	Low	Purity or grade of test substance was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using a concurrent negative control group
	Metric 5:	Negative Control Response	Low	A picture of the control response was all that was reported
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided no details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Low	Only general methods of exposure administration were reported so assessment was difficult to determine
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	Medium	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/Spacing of Exposure Levels	N/A	Only one concentration tested
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test seeds was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of test organisms or replicates was not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	Low	Root growth was not quantitatively reported

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Study Citation:	Kannan, S. (1986). Effects of dibutyl phthalate and phthalic-acid on chlorosis recovery in iron-deficiency stressed sorghum cultivars. Journal of Plant Nutrition 9(12):1543-1551.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Sorghum bicolor</i> ; cv. CSH-5; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5433174			
Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Uninformative	Statistical analysis was not conducted.
	Metric 22:	Reporting of Data	Uninformative	Only figures were presented
	Metric 23:	Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability
Additional Comments: recovery from chlorosis was noted but not quantified				

Overall Quality Determination**Uninformative**

Study Citation:	Kannan, S. (1986). Effects of dibutyl phthalate and phthalic-acid on chlorosis recovery in iron-deficiency stressed sorghum cultivars. Journal of Plant Nutrition 9(12):1543-1551.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Sorghum bicolor</i> ; cv. CS-3541; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5433174			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The chemical was identified by name only.
	Metric 2:	Test Substance Source	Low	The source was not reported.
	Metric 3:	Test Substance Purity	Low	Purity or grade of the test substance was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using a concurrent negative control group.
	Metric 5:	Negative Control Response	Low	A picture of the control response was all that was reported.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided no details on the measures taken to appropriately prepare test concentrations.
	Metric 8:	Consistency of Exposure Administration	Low	Only general methods of exposure administration were reported, so assessment was difficult to determine.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	Medium	The duration of exposure was reported and suitable for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	Only one concentration was tested.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test seeds was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of test organisms or replicates was not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.
	Metric 17:	Outcome Assessment Methodology	Low	Root growth was not quantitatively reported.
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported.

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Study Citation:	Kannan, S. (1986). Effects of dibutyl phthalate and phthalic-acid on chlorosis recovery in iron-deficiency stressed sorghum cultivars. Journal of Plant Nutrition 9(12):1543-1551.		
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days		
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake		
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Sorghum bicolor</i> ; cv. CS-3541; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Development/Growth		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	5433174		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	Uninformative	Statistical analysis was not conducted.
Metric 22:	Reporting of Data	Uninformative	Only figures were presented.
Metric 23:	Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability.
Additional Comments: Recovery from chlorosis was noted but not quantified.			
Overall Quality Determination		Uninformative	

Study Citation:	Kannan, S. (1986). Effects of dibutyl phthalate and phthalic-acid on chlorosis recovery in iron-deficiency stressed sorghum cultivars. Journal of Plant Nutrition 9(12):1543-1551.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Sorghum bicolor</i> ; cv. CSH-5; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5433174			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	Low	The source was not reported.
	Metric 3:	Test Substance Purity	Low	Purity or grade of test substance was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using a concurrent negative control group
	Metric 5:	Negative Control Response	Low	A picture of the control response was all that was reported
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided no details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Low	Only general methods of exposure administration were reported so assessment was difficult to determine
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	Medium	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	Only one concentration tested
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test seeds was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of test organisms or replicates was not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	Low	Root growth was not quantitatively reported
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported

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Study Citation:	Kannan, S. (1986). Effects of dibutyl phthalate and phthalic-acid on chlorosis recovery in iron-deficiency stressed sorghum cultivars. Journal of Plant Nutrition 9(12):1543-1551.		
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days		
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake		
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Sorghum bicolor</i> ; cv. CSH-5; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Development/Growth		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	5433174		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	Uninformative	Statistical analysis was not conducted.
Metric 22:	Reporting of Data	Uninformative	Only figures were presented
Metric 23:	Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability
Additional Comments: recovery from chlorosis was noted but not quantified			
Overall Quality Determination		Uninformative	

Study Citation:	Kannan, S. (1986). Effects of dibutyl phthalate and phthalic-acid on chlorosis recovery in iron-deficiency stressed sorghum cultivars. Journal of Plant Nutrition 9(12):1543-1551.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Sorghum bicolor</i> ; cv. 2077-5; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5433174			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	Low	The source was not reported.
	Metric 3:	Test Substance Purity	Low	Purity or grade of test substance was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using a concurrent negative control group
	Metric 5:	Negative Control Response	Low	A picture of the control response was all that was reported
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided no details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Low	Only general methods of exposure administration were reported so assessment was difficult to determine
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	Medium	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	Only one concentration tested
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test seeds was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of test organisms or replicates was not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	Low	Root growth was not quantitatively reported
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported

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Study Citation:	Kannan, S. (1986). Effects of dibutyl phthalate and phthalic-acid on chlorosis recovery in iron-deficiency stressed sorghum cultivars. Journal of Plant Nutrition 9(12):1543-1551.		
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days		
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake		
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Sorghum bicolor</i> ; cv. 2077-5; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Development/Growth		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	5433174		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	Uninformative	Statistical analysis was not conducted.
Metric 22:	Reporting of Data	Uninformative	Only figures were presented
Metric 23:	Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability
Additional Comments: recovery from chlorosis was noted but not quantified			
Overall Quality Determination		Uninformative	

Study Citation:	Dueck, T. A., Dijk, Van, C. J., David, F., Scholz, N., Vanwalleghe, F. (2003). Chronic effects of vapour phase di-n-butyl phthalate (DBP) on six plant species. Chemosphere 53(8):911-920.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Trifolium repens</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1302103			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The chemical was identified by name and CAS#.
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity or grade of the test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Study authors reported using an appropriate concurrent negative control group, note that all controls had some DBP detected. Control group DBP concentrations are reported in Table 1.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail. The chambers were constructed of "hardened glass" and aluminum.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups based on timed samplings.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies. Samples were taken frequently: "Actual DBP concentrations were measured by taking air samples from each chamber using an automated gas sampling device (GS-1 gas sampler, Gerstel, M €ulheim, Germany). Sampling was performed with Tenax TA tubes (2,6-diphenylphenylene oxide polymer) at 250ml min1 for 20 min. During the fumigation experiment, each chamber was sampled in duplicate on a weekly basis during the complete exposure period (76 days)."
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	The number of exposure groups and spacing of exposure levels were suitable. Concentration gradient is reported as both actual and nominal within table 1.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via fumigation in a continually renewed chamber.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The crop and herbaceous species originated from germinated seeds, but source was not explicitly reported.

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Study Citation:	Dueck, T. A., Dijk, Van, C. J., David, F., Scholz, N., Vanwalleghem, F. (2003). Chronic effects of vapour phase di-n-butyl phthalate (DBP) on six plant species. Chemosphere 53(8):911-920.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Trifolium repens</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1302103			
Domain	Metric	Rating	Comments	
	Metric 14: Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions.	
	Metric 15: Number of Organisms and Replicates per Group	Low	There were four plants per treatment and no replicates reported. "Three to four weeks after sowing, when the first mature leaves appeared, four uniform individuals per species including the tree species were placed in each fumigation chamber (the maximum number physically possible) for exposure to DBP."	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	
	Metric 18: Consistency of Outcome Assessment	Medium	Outcomes were assessed consistently across study groups but few details were reported.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.	
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.	
	Metric 23: Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.	
Additional Comments:	This form represents growth outcomes associated with Dry Weight reported for Trifolium shoot and roots within Figure 3 on page 6/10.			

Overall Quality Determination**High**

Study Citation:	Dueck, T. A., Dijk, Van, C. J., David, F., Scholz, N., Vanwalleghem, F. (2003). Chronic effects of vapour phase di-n-butyl phthalate (DBP) on six plant species. Chemosphere 53(8):911-920.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Trifolium repens</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1302103			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Study authors reported using an appropriate concurrent negative control group, note that all controls had some DBP detected. Control group DBP concentrations are reported in Table 1.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail. The chambers were constructed of "hardened glass" and aluminum.
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups based on timed samplings.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies. Samples were taken frequently: "Actual DBP concentrations were measured by taking air samples from each chamber using an automated gas sampling device (GS-1 gas sampler, Gerstel, M €ulheim, Germany). Sampling was performed with Tenax TA tubes (2,6-diphenylphenylene oxide polymer) at 250ml min ⁻¹ for 20 min. During the fumigation experiment, each chamber was sampled in duplicate on a weekly basis during the complete exposure period (76 days)."
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	The number of exposure groups and spacing of exposure levels were suitable. Concentration gradient is reported as both actual and nominal within table 1.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via fumigation in a continually renewed chamber.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The crop and herbaceous species originated from germinated seeds, but source was not explicitly reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions.
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Study Citation:	Dueck, T. A., Dijk, Van, C. J., David, F., Scholz, N., Vanwallegthem, F. (2003). Chronic effects of vapour phase di-n-butyl phthalate (DBP) on six plant species. Chemosphere 53(8):911-920.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Air; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Trifolium repens</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	1302103			
Domain	Metric	Rating	Comments	
	Metric 15: Number of Organisms and Replicates per Group	Low	Four plants per treatment and no replicates reported, "Three to four weeks after sowing, when the first mature leaves appeared, four uniform individuals per species including the tree species were placed in each fumigation chamber (the maximum number physically possible) for exposure to DBP.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	
	Metric 18: Consistency of Outcome Assessment	Medium	Outcomes were assessed consistently across study groups but few details were reported.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	High	Statistical methods were adequately described.	
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group in Table 2.	
	Metric 23: Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.	
Additional Comments:	This form is for ADME (biotransformation) assessment of DBP concentration in leaf tissue.			
Overall Quality Determination		High		

Study Citation:	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. Frontiers of Environmental Science & Engineering 9(2):259-268.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Triticum aestivum</i> ; Embryo			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2915866			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	Chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	Source was reported, the test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity reported as 99.6%	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported in Table 1 and adequate.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type (168 hours of cultivation time for Allium Sepa and 72 hours for the other species).	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were justified for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Seeds exposed via soil.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test seeds were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed seeds.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.	
Domain 5: Outcome Assessment				
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Study Citation:	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. Frontiers of Environmental Science & Engineering 9(2):259-268.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Triticum aestivum</i> ; Embryo
Health Outcome:	Reproductive/Teratogenic
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	2915866

Domain	Metric	Rating	Comments
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	Nothing reported to indicate there were differences among groups that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Section 2.7 (page 3/10) does not present the statistical tests used to perform analysis other than the program used and p-value for significance.
	Metric 22: Reporting of Data	High	Percent germination was presented as a mean among the four treatment replicates for each treatment and control group were presented.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This form represents the germination rate results presented in Table 1 for *Triticum aestivum* with DBP exposure.

Overall Quality Determination
High

Study Citation:	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. Frontiers of Environmental Science & Engineering 9(2):259-268.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Triticum aestivum</i> ; Embryo			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2915866			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	Chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	Source was reported, the test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity reported as 99.6%	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was adequate.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were justified for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Seeds exposed via soil.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test seeds were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed seeds.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.	

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Study Citation:	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. Frontiers of Environmental Science & Engineering 9(2):259-268.		
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)		
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Triticum aestivum</i> ; Embryo		
Health Outcome:	Development/Growth		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	2915866		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20: Outcomes Unrelated to Exposure	Medium	Nothing reported to indicate there were differences among groups that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Section 2.7 (page 3/10) does not present the statistical tests used to perform analysis other than the program used and p-value for significance.
	Metric 22: Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group and were adequate to determine values for the endpoint.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments:	"DnBP and DEHP at a range of concentrations in the experimental soil showed no discernible effect on the germination rate of the seven test plant species. However, they did exert effects on root elongation, seedling growth and biomass to different extents, indicating the potential applicability of seedling growth in the evaluation of the phytotoxicity of PAE compounds."		
Overall Quality Determination		High	

Study Citation:	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. Frontiers of Environmental Science & Engineering 9(2):259-268.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Triticum aestivum</i> ; Embryo			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Photosynthesis			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2915866			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity was reported as 99.6%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was adequate.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were justified for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Seeds were exposed via soil.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test seeds were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed seeds.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.	
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Study Citation:	Ma, T., Teng, Y., Christie, P., Luo, Y. (2015). Phytotoxicity in seven higher plant species exposed to di-n-butyl phthalate or bis (2-ethylhexyl) phthalate. Frontiers of Environmental Science & Engineering 9(2):259-268.		
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)		
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Triticum aestivum</i> ; Embryo		
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Photosynthesis		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	2915866		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
Metric 20:	Outcomes Unrelated to Exposure	Medium	Nothing was reported to indicate there were differences among groups that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	Low	Section 2.7 (page 3/10) does not present the statistical tests used to perform analysis other than the program used and p-value for significance.
Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group and were adequate to determine values for the endpoint.
Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments:	This is for pigment content."The results indicate that longer periods of cultivation of the test plants may make it easier to interpret the changes in pigment contents."		
Overall Quality Determination		High	

Study Citation:	Gao, M., Guo, Z., Dong, Y., Song, Z. (2019). Effects of di-n-butyl phthalate on photosynthetic performance and oxidative damage in different growth stages of wheat in cinnamon soils. Environmental Pollution 250:357-365.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Triticum aestivum</i> L.; Embryo			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5495646			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The DBP was identified by CASRN.
	Metric 2:	Test Substance Source	Low	The source was reported to be J & K Scientific Ltd. in Beijing, China. However, it was not reported if the DBP was analytically verified.
	Metric 3:	Test Substance Purity	Low	
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of a concurrent negative control.
	Metric 5:	Negative Control Response	High	The negative control response was reported in Figure 1 and was adequate for the outcomes of interest.
	Metric 6:	Randomized Allocation	Low	It was not reported how the seeds were allocated into study groups. The study was, however, reported to have a completely randomized design.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The test media was prepared by dissolving DBP into methanol to obtain 3 different concentrations of stock solution. Stock solutions were added to cinnamon soil and stirred completely. They were then mixed with the appropriate amount of untreated soil to achieve the desired concentrations. The methanol was then allowed to evaporate with 5 day soil equilibration period.
	Metric 8:	Consistency of Exposure Administration	High	
	Metric 9:	Measurement of Test Substance Concentration	Low	It was not reported if the test concentrations were measured at any point.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration was reported to be for 40 days with samples were taken at 14d, 24d and 40d. This was adequate to observe a response.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	There were only 3 exposure groups, which is lower than is typical, but the spacing was adequate to see a response.
	Metric 12:	Testing at or Below Solubility Limit	N/A	The exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The wheat seeds were from the Agro-Environmental Protection Institute, Ministry of Agriculture, China.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the seeds were acclimated.

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Study Citation:	Gao, M., Guo, Z., Dong, Y., Song, Z. (2019). Effects of di-n-butyl phthalate on photosynthetic performance and oxidative damage in different growth stages of wheat in cinnamon soils. Environmental Pollution 250:357-365.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Triticum aestivum</i> L.; Embryo
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	5495646

Domain	Metric	Rating	Comments
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were 16 seeds per test chamber. There were 3 replicates for each treatment.
Domain 5: Outcome Assessment			
	Metric 16: Adequacy of Test Conditions	Medium	The plants were fertilized on a regular basis. They were kept in a greenhouse at 25C during the day and 15C at night in cinnamon soil. Little other information was provided. It would have been beneficial to have information on photoperiod and relative humidity.
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—plant biomass.
	Metric 18: Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups—at each sample period, plants were rinsed and dried with filter paper and separated into roots, stems, and leaves. Dry weights were then taken.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	The statistical methods were described in the "statistical analysis" section of the paper.
	Metric 22: Reporting of Data	High	The exposure and control outcomes were reported in Figure 1, and they were adequate for the outcomes of interest.
	Metric 23: Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability is reported in Figure 1.

Additional Comments: This portion of the evaluation is on the effect of DBP on wheat biomass. Development/growth was selected as the outcome of interest.

Overall Quality Determination

High

Study Citation:	Gao, M., Guo, Z., Dong, Y., Song, Z. (2019). Effects of di-n-butyl phthalate on photosynthetic performance and oxidative damage in different growth stages of wheat in cinnamon soils. Environmental Pollution 250:357-365.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Triticum aestivum</i> L.; Embryo			
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)-Photosynthesis			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5495646			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The DBP was identified by CASRN.
	Metric 2:	Test Substance Source	Low	The source was reported to be J & K Scientific Ltd. in Beijing, China. However, it was not reported if the DBP was analytically verified.
	Metric 3:	Test Substance Purity	Low	The purity of the DBP was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of a concurrent negative control.
	Metric 5:	Negative Control Response	High	The negative control responses were reported in Figures 2-4 and in Table 1, and were adequate for the outcomes of interest.
	Metric 6:	Randomized Allocation	Low	It was not reported how the seeds were allocated into study groups. The study was, however, reported to have a completely randomized design.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The test media was prepared by dissolving DBP into methanol to obtain three different concentrations of stock solution. Stock solutions were added to cinnamon soil and stirred completely. They were then mixed with the appropriate amount of untreated soil to achieve the desired concentrations. The methanol was then allowed to evaporate with a 5-day soil equilibration period.
	Metric 8:	Consistency of Exposure Administration	High	All test containers were 18x16x22cm pots with 16 seeds each. All contained the same amount of treated soil or control soil under similar conditions.
	Metric 9:	Measurement of Test Substance Concentration	Low	It was not reported if the test concentrations were measured at any point.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration was reported to be for 40 days with samples taken at 14d, 24d and 40d. This was adequate to observe a response.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	There were only three exposure groups, which is lower than is typical, but the spacing was adequate to see a response.
	Metric 12:	Testing at or Below Solubility Limit	N/A	The exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The wheat seeds were from the Agro-Environmental Protection Institute, Ministry of Agriculture, China.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the seeds were acclimated.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	There were 16 seeds per test chamber. There were three replicates for each treatment.
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Study Citation:	Gao, M., Guo, Z., Dong, Y., Song, Z. (2019). Effects of di-n-butyl phthalate on photosynthetic performance and oxidative damage in different growth stages of wheat in cinnamon soils. Environmental Pollution 250:357-365.		
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days		
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake		
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Triticum aestivum</i> L.; Embryo		
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)-Photosynthesis		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	5495646		
Domain	Metric	Rating	Comments
Domain 5: Outcome Assessment			
Metric 16:	Adequacy of Test Conditions	Medium	The plants were fertilized on a regular basis. They were kept in a greenhouse at 25C during the day and 15C at night in cinnamon soil. Little other information was provided. It would have been beneficial to have information on photoperiod and relative humidity.
Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—changes in photosynthesis and oxidative responses.
Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	High	The statistical methods were described in the "statistical analysis" section of the paper.
Metric 22:	Reporting of Data	High	The exposure and control outcomes were reported in Figures 2-4 and in Table 1, and they were adequate for the outcomes of interest.
Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability is reported in the figures and table.
Additional Comments:	This portion of the evaluation is on the effect of DBP on photosynthesis and oxidative stress mechanisms in wheat. Mechanistic outcomes of photosynthesis and oxidative stress were selected as the outcomes of interest.		
Overall Quality Determination		High	

Study Citation:	Gao, M., Qi, Y., Song, W., Xu, H. (2016). Effects of di-n-butyl phthalate and di (2-ethylhexyl) phthalate on the growth, photosynthesis, and chlorophyll fluorescence of wheat seedlings. Chemosphere 151:76-83.		
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days		
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake		
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Triticum sp</i> ; Jinnong 7; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Mechanistic-Photosynthesis		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	3350318		
Domain	Metric	Rating	Comments
Domain 1: Test Substance			
	Metric 1: Test Substance Identity	High	Chemical was identified by name and CAS#.
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3: Test Substance Purity	High	Chemical purity reported as 96.8%.
Domain 2: Test Design			
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent solvent control group.
	Metric 5: Negative Control Response	High	The biological response of the negative control group was suitable
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization			
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.
	Metric 12: Testing at or Below Solubility Limit	Medium	The solvent concentration slightly exceeded an appropriate concentration, but the biological response of the solvent control was acceptable.
Domain 4: Test Organism			
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms (8 seedlings per concentration) were reported and sufficient to characterize toxicological effects. Replicates were not reported but it was stated that the experiment was repeated five times.
Domain 5: Outcome Assessment			
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Study Citation:	Gao, M., Qi, Y., Song, W., Xu, H. (2016). Effects of di-n-butyl phthalate and di (2-ethylhexyl) phthalate on the growth, photosynthesis, and chlorophyll fluorescence of wheat seedlings. Chemosphere 151:76-83.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Triticum sp</i> ; Jinnong 7; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Photosynthesis			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3350318			
Domain	Metric	Rating	Comments	
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health. "The experiments were conducted in an artificial climate chamber. The seedlings were cultured under a cycle of 12-h days at 25 ± 1oC and 12-h nights at 20 ± 1 oC at 60% relative humidity level."	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodologies for chlorophyll content and photosynthetic parameters were reported in detail.	
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.	
Domain 6: Confounding / Variable Control				
	Metric 19: Confounding Variables in Test Design and Procedures	Medium	Data on attrition and/or outcomes unrelated to controlled variables for each study group were not reported, but these are unlikely to have a substantial impact on results.	
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.	
Domain 7: Data Presentation and Analysis				
	Metric 21: Statistical Methods	Low	Statistical analysis was performed but calculations and measures of significance were not provided, so no conclusions about a dose response could be made. Results of multiple comparisons were not provided.	
	Metric 22: Reporting of Data	High	Data for exposure-related findings were shown for each treatment and control group.	
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.	
Additional Comments:	The author's discussion of the effects of DBP and DEHP on growth indices and mechanistic endpoints of wheat seedlings was not clear, and their conclusions were made without incorporating any discussion of statistical significance. Results of multiple comparisons were not provided.			
Overall Quality Determination		High		

Study Citation:	Gao, M., Qi, Y., Song, W., Xu, H. (2016). Effects of di-n-butyl phthalate and di (2-ethylhexyl) phthalate on the growth, photosynthesis, and chlorophyll fluorescence of wheat seedlings. Chemosphere 151:76-83.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Triticum sp</i> ; Jinnong 7; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3350318			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	High	The chemical was identified by name and CAS#.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity was reported as 96.8%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent solvent control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was suitable.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.	
	Metric 8: Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	Medium	The solvent concentration slightly exceeded an appropriate concentration, but the biological response of the solvent control was acceptable.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms (eight seedlings per concentration) were reported and sufficient to characterize toxicological effects. Replicates were not reported, but it was stated that the experiment was repeated five times.	
Domain 5: Outcome Assessment				
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Study Citation:	Gao, M., Qi, Y., Song, W., Xu, H. (2016). Effects of di-n-butyl phthalate and di (2-ethylhexyl) phthalate on the growth, photosynthesis, and chlorophyll fluorescence of wheat seedlings. Chemosphere 151:76-83.			
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Triticum sp</i> ; Jinnong 7; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	3350318			
Domain	Metric	Rating	Comments	
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health. "The experiments were conducted in an artificial climate chamber. The seedlings were cultured under a cycle of 12-h days at 25 ± 1oC and 12-h nights at 20 ± 1 oC at 60% relative humidity level."
	Metric 17:	Outcome Assessment Methodology	Medium	The outcome assessment methodologies for plant height, fresh weights of shoots and roots, and dry weight of shoots and roots were reported but not in sufficient detail.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups at days 7 and 14.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Medium	Data on attrition and/or outcomes unrelated to controlled variables for each study group were not reported, but these are unlikely to have a substantial impact on results. Experiments were conducted in an artificial climate chamber.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but calculations and measures of significance were not provided, so no conclusions about a dose response could be made. Results of multiple comparisons were not provided.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were shown for each treatment and control group (Table 1).
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments:	The author's discussion of the effects of DBP and DEHP on growth indices of wheat seedlings was not clear, and their conclusions were made without incorporating any discussion of statistical significance. Results of multiple comparisons were not provided.			

Overall Quality Determination**Medium**

Study Citation:	Ma, T. T., Christie, P., Luo, Y. M., Teng, Y. (2014). Physiological and antioxidant responses of germinating mung bean seedlings to phthalate esters in soil. <i>Pedosphere</i> 24(1):107-115.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Vigna radiata</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Reproductive/Teratogenic			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2510954			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	Chemical was identified by name only	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory	
	Metric 3: Test Substance Purity	High	Chemical purity reported as 99.1%	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group	
	Metric 5: Negative Control Response	Low	The biological response of the negative control groups was not reported	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail	
	Metric 8: Consistency of Exposure Administration	Medium	exposures were administered consistently across study groups, albeit with few details	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type	
	Metric 11: Number of Exposure Groups/Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were adequate for a dose response	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via soil.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	
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Study Citation:	Ma, T. T., Christie, P., Luo, Y. M., Teng, Y. (2014). Physiological and antioxidant responses of germinating mung bean seedlings to phthalate esters in soil. <i>Pedosphere</i> 24(1):107-115.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Vigna radiata</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Reproductive/Teratogenic
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	2510954

Domain	Metric	Rating	Comments
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
	Metric 20: Outcomes Unrelated to Exposure	Medium	Nothing reported to indicate there were differences among groups that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	High	Statistical methods were adequately described
	Metric 22: Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group, but results were described in the text.
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: This form is to assess the germination outcome.

Overall Quality Determination

Medium

Study Citation:	Ma, T. T., Christie, P., Luo, Y. M., Teng, Y. (2014). Physiological and antioxidant responses of germinating mung bean seedlings to phthalate esters in soil. Pedosphere 24(1):107-115.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Vigna radiata</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2510954			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The chemical was identified by name only.	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.	
	Metric 3: Test Substance Purity	High	Chemical purity was reported as 99.1%.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.	
	Metric 5: Negative Control Response	High	The biological response of the negative control group was reported and adequate.	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.	
	Metric 8: Consistency of Exposure Administration	Medium	Exposures were administered consistently across study groups, albeit with few details.	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were adequate for a dose response.	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via soil.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of the test system were conducive to maintenance of organism health.	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.	
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Study Citation:	Ma, T. T., Christie, P., Luo, Y. M., Teng, Y. (2014). Physiological and antioxidant responses of germinating mung bean seedlings to phthalate esters in soil. <i>Pedosphere</i> 24(1):107-115.		
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)		
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)		
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Vigna radiata</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported		
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)		
Chemical:	Dibutyl phthalate (DBP)		
HERO ID:	2510954		
Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
Metric 20:	Outcomes Unrelated to Exposure	High	There were no differences among groups that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	High	Statistical methods were adequately described.
Metric 22:	Reporting of Data	High	Data for exposure-related findings were shown for each treatment and control group.
Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments:	Mechanistic endpoints are: malondialdehyde (MDA) Polyphenol oxidase (PPO)ascorbate peroxidase (APX)Superoxide dismutase (SOF)The glutathione (GSH)peroxidase (POD)Proline content		
Overall Quality Determination		High	

Study Citation:	Ma, T. T., Christie, P., Luo, Y. M., Teng, Y. (2014). Physiological and antioxidant responses of germinating mung bean seedlings to phthalate esters in soil. <i>Pedosphere</i> 24(1):107-115.			
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)			
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Vigna radiata</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	2510954			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	Chemical was identified by name only	
	Metric 2: Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory	
	Metric 3: Test Substance Purity	High	Chemical purity reported as 99.1%	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported using an appropriate concurrent negative control group	
	Metric 5: Negative Control Response	Low	The biological response of the negative control groups was not reported	
	Metric 6: Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail	
	Metric 8: Consistency of Exposure Administration	Medium	exposures were administered consistently across study groups, albeit with few details	
	Metric 9: Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured	
	Metric 10: Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were adequate for a dose response	
	Metric 12: Testing at or Below Solubility Limit	N/A	Exposure was via soil.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health	
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.	
	Metric 18: Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.	
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Study Citation:	Ma, T. T., Christie, P., Luo, Y. M., Teng, Y. (2014). Physiological and antioxidant responses of germinating mung bean seedlings to phthalate esters in soil. <i>Pedosphere</i> 24(1):107-115.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Vigna radiata</i> ; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	2510954

Domain	Metric	Rating	Comments
Domain 6: Confounding / Variable Control			
Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
Metric 20:	Outcomes Unrelated to Exposure	Medium	Nothing reported to indicate there were differences among groups that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis			
Metric 21:	Statistical Methods	High	Statistical methods were adequately described
Metric 22:	Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group, but results were described in the text.
Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes
Additional Comments:	Seedling root and shoot lengths were measured with a millimeter ruler, and the biomass (fresh weight, FW) in each dish was determined by weighing. Root length was defined as the length from root tip to root radicle.		

Overall Quality Determination**Medium**

Study Citation:	Wang, S. G., Lin, X. G., Yin, R., Hou, Y. L. (2003). Effects of di-n-butyl phthalate on mycorrhizal and non-mycorrhizal cowpea plants. <i>Biologia Plantarum</i> 47(4):637-639.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Vigna sinensis</i> ; Juvenile			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5495799			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The test substance was identified by name only.
	Metric 2:	Test Substance Source	Low	The source of the DBP was not reported.
	Metric 3:	Test Substance Purity	Low	The purity/grade of the DBP was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of a negative control in which no DBP was added.
	Metric 5:	Negative Control Response	High	The negative control response was reported in Table 2 and was adequate for the outcome of interest.
	Metric 6:	Randomized Allocation	Low	It was not reported how the seedlings were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	Little information was provided on the preparation of the test media.
	Metric 8:	Consistency of Exposure Administration	Low	All exposure occurred for 60 days with samples taken at 20, 40, and 60 days. Test pot dimensions were not described. More information needed to be reported.
	Metric 9:	Measurement of Test Substance Concentration	Low	It was not reported if exposure concentrations were measured.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration was reported to be 60 days. This was adequate to observe a response.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	There were only two exposure levels, but the spacing was adequate to see a response.
	Metric 12:	Testing at or Below Solubility Limit	N/A	The exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	Three-day old cowpea seedlings were used in the study, but the source of the cowpeas was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the plants were acclimated.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	There were two cowpeas per test chamber, and there were three replicates per treatment.
Domain 5: Outcome Assessment				
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Study Citation:	Wang, S. G., Lin, X. G., Yin, R., Hou, Y. L. (2003). Effects of di-n-butyl phthalate on mycorrhizal and non-mycorrhizal cowpea plants. <i>Biologia Plantarum</i> 47(4):637-639.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Vigna sinensis</i> ; Juvenile			
Health Outcome:	Development/Growth			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5495799			
Domain	Metric	Rating	Comments	
	Metric 16:	Adequacy of Test Conditions	Medium	The plants were kept on a 16L:8D photoperiod with temperatures at 22/18C for day/night. No information was provided on fertilizing or other environmental conditions.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—changes in root and shoot biomass.
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the assessment of root and shoot biomass were limited.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. It was not reported if the plants were acclimated.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistics were performed but not described adequately.
	Metric 22:	Reporting of Data	High	Data for the control response and exposure response were reported in Table 2 and were appropriate for the outcomes of interest.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in the table.
Additional Comments:	This portion of the evaluation was on the effect of DBP on cowpea root and shoot biomass. Development/growth was selected as the outcome of interest.			
Overall Quality Determination		Low		

Study Citation:	Wang, S. G., Lin, X. G., Yin, R., Hou, Y. L. (2003). Effects of di-n-butyl phthalate on mycorrhizal and non-mycorrhizal cowpea plants. <i>Biologia Plantarum</i> 47(4):637-639.			
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days			
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake			
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Vigna sinensis</i> ; Juvenile			
Health Outcome:	ADME (biotransformation)			
Chemical:	Dibutyl phthalate (DBP)			
HERO ID:	5495799			
Domain	Metric	Rating	Comments	
Domain 1: Test Substance				
	Metric 1: Test Substance Identity	Low	The test substance was identified by name only.	
	Metric 2: Test Substance Source	Low	The source of the DBP was not reported.	
	Metric 3: Test Substance Purity	Low	The purity/grade of the DBP was not reported.	
Domain 2: Test Design				
	Metric 4: Negative Controls	High	Study authors reported the use of a negative control in which no DBP was added.	
	Metric 5: Negative Control Response	High	The negative control response was reported in Table 3 and was adequate for the outcome of interest.	
	Metric 6: Randomized Allocation	Low	It was not reported how the seedlings were allocated into study groups.	
Domain 3: Exposure Characterization				
	Metric 7: Experimental System/Test Media Preparation	Low	Little information was provided on the preparation of the test media.	
	Metric 8: Consistency of Exposure Administration	Low	All exposure occurred for 60 days with samples taken at 20, 40, and 60 days. Test pot dimensions were not described. More information needed to be reported.	
	Metric 9: Measurement of Test Substance Concentration	Low	It was not reported if exposure concentrations were measured.	
	Metric 10: Exposure Duration and Frequency	High	The exposure duration was reported to be 60 days. This was adequate to observe a response.	
	Metric 11: Number of Exposure Groups/ Spacing of Exposure Levels	Low	There were only 2 exposure levels, but the spacing was adequate to see a response.	
	Metric 12: Testing at or Below Solubility Limit	N/A	The exposure was via soil.	
Domain 4: Test Organism				
	Metric 13: Test Organism Characteristics	Low	3 day old cowpea seedlings were used in the study, but the source of the cowpeas was not reported.	
	Metric 14: Acclimatization and Pretreatment Conditions	Low	It was not reported if the plants were acclimated.	
	Metric 15: Number of Organisms and Replicates per Group	Medium	There were two cowpeas per test chamber, and there were 3 replicates per treatment.	
Domain 5: Outcome Assessment				
	Metric 16: Adequacy of Test Conditions	Medium	The plants were kept on a 16L:8D photoperiod with temperatures at 22/18C for day/night. No information was provided on fertilizing or other environmental conditions.	

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Study Citation:	Wang, S. G., Lin, X. G., Yin, R., Hou, Y. L. (2003). Effects of di-n-butyl phthalate on mycorrhizal and non-mycorrhizal cowpea plants. <i>Biologia Plantarum</i> 47(4):637-639.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake
Taxa, Species, Age:	Vegetation; Vascular Plants; <i>Vigna sinensis</i> ; Juvenile
Health Outcome:	ADME (biotransformation)
Chemical:	Dibutyl phthalate (DBP)
HERO ID:	5495799

Domain	Metric	Rating	Comments
	Metric 17: Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest—DBP accumulation in plant tissues.
	Metric 18: Consistency of Outcome Assessment	High	The shoots and roots were oven dried and ground up for DBP analysis. Gas chromatography was used to determine DBP levels in the tissues.
Domain 6: Confounding / Variable Control			
	Metric 19: Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. It was not reported if the plants were acclimated.
	Metric 20: Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis			
	Metric 21: Statistical Methods	Low	Statistics were performed but not described adequately.
	Metric 22: Reporting of Data	High	Data for the control response and exposure response were reported in Table 3 and were appropriate for the outcomes of interest.
	Metric 23: Explanation of Unexpected Outcomes	High	Study authors did not report and unexpected outcomes. Variability was reported in the table.

Additional Comments: This portion of the evaluation was on DBP accumulation in the roots and shoots of cowpeas. ADME was selected as the outcome of interest.

Overall Quality Determination

Medium