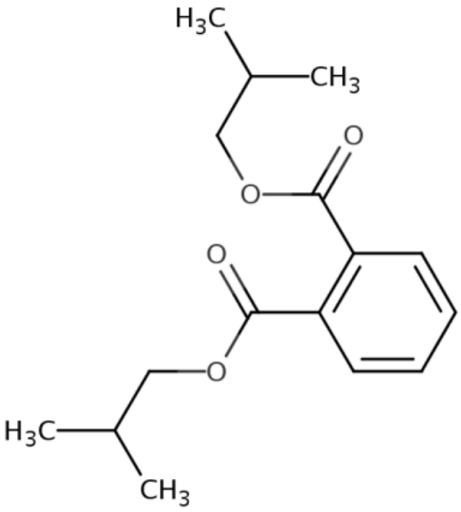
United States
Environmental Protection Agency

July 2025
Office of Chemical Safety and
Pollution Prevention

Data Quality Evaluation Information for Environmental Hazard for Di-isobutyl Phthalate (DIBP) (1,2-Benzenedicarboxylic acid, 1,2-bis(2-methylpropyl) ester)

Systematic Review Support Document for the Draft Risk Evaluation

CASRN: 84-69-5



July 2025

This supplemental file contains information regarding the data quality evaluation results relevant to the analysis of environmental hazard for the *Draft Environmental Hazard Assessment for Diisobutyl Phthalate (DIBP)*. Due to data gaps identified for DIBP, a read-across analog approach was employed and DBP data were used to fill data gaps in the *Draft Environmental Hazard Assessment for Diisobutyl Phthalate (DIBP)*. 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* (also referred to as '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 Diisobutyl Phthalate (DIBP) - Systematic Review Protocol*.

Different data quality evaluation forms were used depending on the organism 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. Data quality evaluation results were organized by first presenting the data for the target compound (DIBP) followed by a separate section for analog data (DBP). The table of contents lists references based on chemical (target chemical followed by analog chemical), and study details and respective endpoints are organized by first the relevant habitat (*i.e.*, aquatic, 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
Diisobutyl Phthalate		
Habitat: Aquatic (freshwater)		
Taxa: Vertebrates		
Pimephales promelas		
11581733	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 (Pimephales promelas) – The potential application of omics data in risk evaluations under TSCA (internal use only).	**8**
Habitat: Aquatic (marine)		
Taxa: Plants (Non-vascular)		
Karenia brevis		
3230225	Liu, N., Wen, F., Li, F., Zheng, X., Liang, Z., Zheng, H. (2016). Inhibitory mechanism of phthalate esters on Karenia brevis. Chemosphere 155:498-508.	**20**
Habitat: Aquatic (brackish)		
Taxa: Invertebrates		
Nitocra spinipes		
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 (Alburnus alburnus) and the harpacticoid Nitocra spinipes. Chemosphere 8(11-12):843-851.	**23**
Habitat: Terrestrial		
Taxa: Invertebrates		
Caenorhabditis elegans		
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 Caenorhabditis elegans. PLoS ONE 8(12):e82657.	**25**
Lasius niger		

2347468 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. **33**

Taxa: Plants (Vascular)

Nicotiana tabacum

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. Pedosphere 27(6):1073-1082. **35**

Nicotinana tobacum

792357 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. **39**

Analog Chemical Data

Habitat: Aquatic (freshwater)

Taxa: Vertebrates

Cyprinodon variegatus

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. Environmental Toxicology and Chemistry 14(9):1569-1574. **47**

Danio rerio

2298079 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. **49**

Lepomis macrochirus

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. Environmental Toxicology and Chemistry 14(9):1569-1574. **51**

1316201 Bionomics,, EG&G (1983). Exhibit III: Acute toxicity of thirteen phthalate esters to bluegill (Lepomis macrochirus). **53**

18064 Buccafusco, R. J., Ells, S. J., Leblanc, G. A. (1981). Acute toxicity of priority pollutants to bluegill (Lepomis macrochirus). Bulletin of Environmental Contamination and Toxicology 26(4):446-452. **55**

Oncorhynchus mykiss

6571362 EnviroSystem, (1991). Early life-stage toxicity of di-n-butyl phthalate (DnBP) to the rainbow trout (Oncorhynchus mykiss) under flow-through conditions. **57**

6571362 EnviroSystem, (1991). Early life-stage toxicity of di-n-butyl phthalate (DnBP) to the rainbow trout (Oncorhynchus mykiss) under flow-through conditions. **59**

Oncorhynchus mykiss (Salmo gairdneri)

5530771 Bionomics,, EG&G (1983). Acute toxicity of fourteen phthalate esters to rainbow trout (Salmo gairdneri) under flow-through conditions (final report) report no BW-83-3-1373. **63**

Oryzias latipes

10064186 EAG Laboratories, (2018). Dibutyl phthalate: Medaka extended one generation reproduction test (final report). **65**

10064186 EAG Laboratories, (2018). Dibutyl phthalate: Medaka extended one generation reproduction test (final report). **69**

Oryzias melastigma

2298079 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. **85**

Pimephales promelas

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. Environmental Toxicology and Chemistry 14(9):1569-1574. **87**

1336024 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. **91**

1336024 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. **93**

Salmo mykiss

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. Environmental Toxicology and Chemistry 14(9):1569-1574. **95**

Taxa: Invertebrates

Chironomus plumosus

1332972 Streufort, J. M. (1978). Some effects of two phthalic acid esters on the life cycle of the midge (Chironomus plumosus). **97**

1332972 Streufort, J. M. (1978). Some effects of two phthalic acid esters on the life cycle of the midge (Chironomus plumosus). **101**

Chironomus tentans

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. Environmental Toxicology and Chemistry 20(8):1805-1815. **103**

Daphnia magna

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. Environmental Toxicology and Chemistry 14(9):1569-1574. **107**

1336024 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. **109**

4829279 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 Daphnia magna. Bulletin of Environmental Contamination and Toxicology 101(2):214-221. **111**

1336024 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. **113**

4829279 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 Daphnia magna. Bulletin of Environmental Contamination and Toxicology 101(2):214-221. **119**

Hyalella azteca

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. Environmental Toxicology and Chemistry 20(8):1805-1815. **123**

Paratanytarsus parthenogenetica

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. Environmental Toxicology and Chemistry 14(9):1569-1574. **127**

Paratanytarsus parthenogenica

1316219 Bionomics,, EG&G (1984). Acute toxicity of twelve phthalate esters to Paratanytarsus parthenogenica (final report) report no BW-83-6-1424. **129**

Taxa: Plants (Non-vascular)

Selenastrum capricornutum

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. Environmental Toxicology and Chemistry 14(9):1569-1574. **131**

Habitat: Aquatic (marine)

Taxa: Invertebrates

Animalia

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. **133**

Annelida

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. **137**

Arthropoda

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. **141**

Chordata

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. **145**

Coelenterata

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. **149**

Echinodermata

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. **153**

Mollusca

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. **157**

Mysidopsis bahia

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. Environmental Toxicology and Chemistry 14(9):1569-1574. **161**

Rhynchocoela

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. **163**

Habitat: Aquatic (brackish)

Taxa: Invertebrates

Nitocra spinipes

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 (Alburnus alburnus) and the harpacticoid Nitocra spinipes. Chemosphere 8(11-12):843-851. **165**

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 (Pimephales promelas) – 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; *Pimephales promelas*; Larvae
Health Outcome:	Behavioral
Chemical:	Diisobutyl Phthalate- Parent compound
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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Taxa, Species, Age:	Vertebrate; Fish; *Pimephales promelas*; Larvae
Health Outcome:	Behavioral
Chemical:	Diisobutyl Phthalate- Parent compound
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 for 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 behavior 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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Taxa, Species, Age:	Vertebrate; Fish; *Pimephales promelas*; Larvae
Health Outcome:	Behavioral
Chemical:	Diisobutyl Phthalate- Parent compound
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	Calculation of all points of departure measurements was described in the text (methods and results) and Table 6 shows the bevavior-based POD value for DBP. However, actual 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 data evaluation is for the calculation of the behavior point of departure as part of the behavior analysis following exposure to DIBP.

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 (Pimephales promelas) – The potential application of omics data in risk evaluations under TSCA (internal use only).
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Taxa, Species, Age:	Vertebrate; Fish; *Pimephales promelas*; Larvae
Health Outcome:	Mechanistic-Cell signaling/function
Chemical:	Diisobutyl Phthalate- Parent compound
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 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 transcriptomic changes.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups (11 treatment groups) and spacing were adequate for the transcriptomics analysis and POD derivation.
	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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Taxa, Species, Age:	Vertebrate; Fish; *Pimephales promelas*; Larvae
Health Outcome:	Mechanistic-Cell signaling/function
Chemical:	Diisobutyl Phthalate- Parent compound
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 (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 RNA 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 vessel 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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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 (Pimephales promelas) – 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; *Pimephales promelas*; Larvae
Health Outcome:	Mechanistic-Cell signaling/function
Chemical:	Diisobutyl Phthalate- Parent compound
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 DIBP.

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 (Pimephales promelas) – The potential application of omics data in risk evaluations under TSCA (internal use only).
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Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Pimephales promelas*; Larvae
Health Outcome:	Mechanistic-Cell signaling/function
Chemical:	Diisobutyl Phthalate- Parent compound
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 minoow 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 (11 treatment groups) and spacing were adequate to obtain changes in metabolomics data.

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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; *Pimephales promelas*; Larvae
Health Outcome:	Mechanistic-Cell signaling/function
Chemical:	Diisobutyl Phthalate- Parent compound
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 vessel 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 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 (Pimephales promelas) – 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; *Pimephales promelas*; Larvae
Health Outcome:	Mechanistic-Cell signaling/function
Chemical:	Diisobutyl Phthalate- Parent compound
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 DiBP.

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 (Pimephales promelas) – 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; *Pimephales promelas*; Larvae
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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 was 0% total mortality 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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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 (Pimephales promelas) – 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; *Pimephales promelas*; Larvae
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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 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 was described in detail in the supplental 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 (2C) 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 evaluation is for the mortality assessment with DiBP.

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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 (Pimephales promelas) – 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; *Pimephales promelas*; Larvae
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
HERO ID:	11581733

Domain	Metric	Rating	Comments
Overall Quality Determination		**High**	

Study Citation:	Liu, N., Wen, F., Li, F., Zheng, X., Liang, Z., Zheng, H. (2016). Inhibitory mechanism of phthalate esters on Karenia brevis. Chemosphere 155:498-508.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
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; *Karenia brevis*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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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Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
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; *Karenia brevis*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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 (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 $\pm$ 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 appear to be consistent across groups. 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. Details of EC50 calculations were not provided.

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Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
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; *Karenia brevis*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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 observed for DIBP 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:	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 (Alburnus alburnus) and the harpacticoid Nitocra spinipes. 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; *Nitocra spinipes*; Adult
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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.
	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.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	At least six concentrations were tested, but a range of levels 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 (3.0 mg/L) was below the water solubility given in the Final Scope for DIBP, 6.2 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	There were 20 organisms with no replicates used per treatment.
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 (Alburnus alburnus) and the harpacticoid Nitocra spinipes. 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; *Nitocra spinipes*; Adult
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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 the table.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: None

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 Caenorhabditis elegans. 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); *Caenorhabditis elegans*; wild type; Larvae
Health Outcome:	Behavioral
Chemical:	Diisobutyl Phthalate- Parent compound
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 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	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

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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 Caenorhabditis elegans. 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); *Caenorhabditis elegans*; wild type; Larvae
Health Outcome:	Behavioral
Chemical:	Diisobutyl Phthalate- Parent compound
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: locomotor and thermotaxis

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 Caenorhabditis elegans. 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); *Caenorhabditis elegans*; wild type; Larvae
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)
Chemical:	Diisobutyl Phthalate- Parent compound
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 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 "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 Caenorhabditis elegans. 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); *Caenorhabditis elegans*; wild type; Larvae
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)
Chemical:	Diisobutyl Phthalate- Parent compound
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: 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 Caenorhabditis elegans. 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); *Caenorhabditis elegans*; wild type; Larvae
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)
Chemical:	Diisobutyl Phthalate- Parent compound
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 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 "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 Caenorhabditis elegans. 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); *Caenorhabditis elegans*; wild type; Larvae
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)
Chemical:	Diisobutyl Phthalate- Parent compound
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 Caenorhabditis elegans. 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); *Caenorhabditis elegans*; DA 1267; Larvae
Health Outcome:	Mechanistic-Neurotoxicology-Ocular and Sensory
Chemical:	Diisobutyl Phthalate- Parent compound
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 ”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, and 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.
	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 Caenorhabditis elegans. 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); *Caenorhabditis elegans*; DA 1267; Larvae
Health Outcome:	Mechanistic-Neurotoxicology-Ocular and Sensory
Chemical:	Diisobutyl Phthalate- Parent compound
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: This form is for AFD thermosensory neurons.

Overall Quality Determination — High

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; *Lasius niger*; Adult
Health Outcome:	ADME (biotransformation)
Chemical:	Diisobutyl Phthalate- Parent compound
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	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 source of the 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; *Lasius niger*; Adult
Health Outcome:	ADME (biotransformation)
Chemical:	Diisobutyl Phthalate- Parent compound
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 there were few details 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:	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. Pedosphere 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; *Nicotiana tabacum*; cv K326; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Reproductive/Teratogenic
Chemical:	Diisobutyl Phthalate- Parent compound
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.

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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. Pedosphere 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; *Nicotiana tabacum*; cv K326; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Reproductive/Teratogenic
Chemical:	Diisobutyl Phthalate- Parent compound
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: This form is for 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. Pedosphere 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; *Nicotiana tabacum*; cv K326; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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. Pedosphere 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; *Nicotiana tabacum*; cv K326; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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; *Nicotinana tobacum*; G168; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Reproductive/Teratogenic
Chemical:	Diisobutyl Phthalate- Parent compound
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 (6.2 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				

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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; *Nicotinana tobacum*; G168; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Reproductive/Teratogenic
Chemical:	Diisobutyl Phthalate- Parent compound
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: Germination

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; *Nicotinana tobacum*; Hong da; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Reproductive/Teratogenic
Chemical:	Diisobutyl Phthalate- Parent compound
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 (6.2 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; *Nicotinana tobacum*; Hong da; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Reproductive/Teratogenic
Chemical:	Diisobutyl Phthalate- Parent compound
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

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; *Nicotinana tobacum*; Hong da; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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 (6.2 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; *Nicotinana tobacum*; Hong da; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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; *Nicotinana tobacum*; G168; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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 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 (6.2 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; *Nicotinana tobacum*; G168; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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: This form is for growth (Development-Slowed, Retarded, Delayed or Non-development, Response Site: Not reported).

Overall Quality Determination Low

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; *Cyprinodon variegatus*; Juvenile
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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	Medium	Sample extracts were analyzed by gas chromatography at start and end of 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 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.

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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; *Cyprinodon variegatus*; Juvenile
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
HERO ID:	1321996; Linked HERO ID(s): 1321996, 1316224

Domain	Metric		Rating	Comments
	Metric 17:	Outcome Assessment Methodology	High	Intended outcomes reported.
	Metric 18:	Consistency of Outcome Assessment	High	Outcome assessment consistent across groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	Environmental conditions were consistant 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:	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; *Danio rerio*; AB Strain; Embryo
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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; *Danio rerio*; AB Strain; Embryo
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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:	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; *Lepomis macrochirus*; Juvenile
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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; *Lepomis macrochirus*; Juvenile
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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 (Lepomis macrochirus).
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; *Lepomis macrochirus*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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 labortory.
	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 (Lepomis macrochirus).
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; *Lepomis macrochirus*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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 (Lepomis macrochirus). 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; *Lepomis macrochirus*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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 (Lepomis macrochirus). 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; *Lepomis macrochirus*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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:	EnviroSystem, (1991). Early life-stage toxicity of di-n-butyl phthalate (DnBP) to the rainbow trout (Oncorhynchus mykiss) 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; *Oncorhynchus mykiss*; Juvenile
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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 (Oncorhynchus mykiss) 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; *Oncorhynchus mykiss*; Juvenile
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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 (Oncorhynchus mykiss) 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; *Oncorhynchus mykiss*; Embryo
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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.

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Study Citation:	EnviroSystem, (1991). Early life-stage toxicity of di-n-butyl phthalate (DnBP) to the rainbow trout (Oncorhynchus mykiss) 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; *Oncorhynchus mykiss*; Embryo
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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 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:	EnviroSystem, (1991). Early life-stage toxicity of di-n-butyl phthalate (DnBP) to the rainbow trout (Oncorhynchus mykiss) 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; *Oncorhynchus mykiss*; Embryo
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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.
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 (Oncorhynchus mykiss) 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; *Oncorhynchus mykiss*; Embryo
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
HERO ID:	6571362

Domain	Metric		Rating	Comments
	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:	Bionomics,, EG&G (1983). Acute toxicity of fourteen phthalate esters to rainbow trout (Salmo gairdneri) 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; *Oncorhynchus mykiss (Salmo gairdneri)*; Adult
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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 (Salmo gairdneri) 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; *Oncorhynchus mykiss (Salmo gairdneri)*; Adult
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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:	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; *Oryzias latipes*; Adult
Health Outcome:	Reproductive/Teratogenic
Chemical:	Diisobutyl Phthalate- Parent compound
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; *Oryzias latipes*; Adult
Health Outcome:	Reproductive/Teratogenic
Chemical:	Diisobutyl Phthalate- Parent compound
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; *Oryzias latipes*; Adult
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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; *Oryzias latipes*; Adult
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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; *Oryzias latipes*; Adult
Health Outcome:	Reproductive/Teratogenic
Chemical:	Diisobutyl Phthalate- Parent compound
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

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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; *Oryzias latipes*; Adult
Health Outcome:	Reproductive/Teratogenic
Chemical:	Diisobutyl Phthalate- Parent compound
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: 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; *Oryzias latipes*; Adult
Health Outcome:	Skin & Connective Tissue
Chemical:	Diisobutyl Phthalate- Parent compound
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)
	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; *Oryzias latipes*; Adult
Health Outcome:	Skin & Connective Tissue
Chemical:	Diisobutyl Phthalate- Parent compound
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: 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; *Oryzias latipes*; Adult
Health Outcome:	Hepatic/Liver
Chemical:	Diisobutyl Phthalate- Parent compound
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; *Oryzias latipes*; Adult
Health Outcome:	Hepatic/Liver
Chemical:	Diisobutyl Phthalate- Parent compound
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; *Oryzias latipes*; Adult
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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; *Oryzias latipes*; Adult
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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; *Oryzias latipes*; Adult
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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; *Oryzias latipes*; Adult
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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; *Oryzias latipes*; Adult
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Endocrine toxicity-Reproductive/Teratogenic
Chemical:	Diisobutyl Phthalate- Parent compound
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; *Oryzias latipes*; Adult
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Endocrine toxicity-Reproductive/Teratogenic
Chemical:	Diisobutyl Phthalate- Parent compound
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; *Oryzias latipes*; Adult
Health Outcome:	Endocrine
Chemical:	Diisobutyl Phthalate- Parent compound
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; *Oryzias latipes*; Adult
Health Outcome:	Endocrine
Chemical:	Diisobutyl Phthalate- Parent compound
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; *Oryzias latipes*; Adult
Health Outcome:	Renal/Kidney
Chemical:	Diisobutyl Phthalate- Parent compound
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; *Oryzias latipes*; Adult
Health Outcome:	Renal/Kidney
Chemical:	Diisobutyl Phthalate- Parent compound
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:	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; *Oryzias melastigma*; ChgH-EGFP; Larvae
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Receptor binding/ regulation of receptor activity-Endocrine toxicity-Reproductive/Teratogenic
Chemical:	Diisobutyl Phthalate- Parent compound
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; *Oryzias melastigma*; ChgH-EGFP; Larvae
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Receptor binding/ regulation of receptor activity-Endocrine toxicity-Reproductive/Teratogenic
Chemical:	Diisobutyl Phthalate- Parent compound
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; *Pimephales promelas*; Juvenile
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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.

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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; *Pimephales promelas*; Juvenile
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
HERO ID:	1321996; Linked HERO ID(s): 1321996, 1316224

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	High	Outcome assessment consistent across groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	Environmental conditions were consistant 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:	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; *Pimephales promelas*; Juvenile
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions were appropriate for the test.

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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; *Pimephales promelas*; Juvenile
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
HERO ID:	1321996; Linked HERO ID(s): 1321996, 1316224

Domain	Metric		Rating	Comments
	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:	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; *Pimephales promelas*; Embryo
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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; *Pimephales promelas*; Embryo
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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 morality 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 morality 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:	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; *Pimephales promelas*; Embryo
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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; *Pimephales promelas*; Embryo
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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 morality 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 morality in the early life-stage test.

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; *Salmo mykiss*; Juvenile
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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	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	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; *Salmo mykiss*; Juvenile
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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 consistant 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:	Streufort, J. M. (1978). Some effects of two phthalic acid esters on the life cycle of the midge (Chironomus plumosus).
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; *Chironomus plumosus*; Larvae
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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				

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Study Citation:	Streufort, J. M. (1978). Some effects of two phthalic acid esters on the life cycle of the midge (Chironomus plumosus).
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; *Chironomus plumosus*; Larvae
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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 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	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:	Streufort, J. M. (1978). Some effects of two phthalic acid esters on the life cycle of the midge (Chironomus plumosus).
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; *Chironomus plumosus*; Larvae
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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				
	Metric 16:	Adequacy of Test Conditions	Medium	DO, 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 (Chironomus plumosus).
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; *Chironomus plumosus*; Larvae
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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	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 (Chironomus plumosus).
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; *Chironomus plumosus*; Larvae
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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 hydrosoil 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 (Chironomus plumosus).
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; *Chironomus plumosus*; Larvae
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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; *Chironomus tentans*; Larvae
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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; *Chironomus tentans*; Larvae
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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 usingthe trimmed Spearman–Karber method. Dry weight datawere analyzed by one-way analysis of variance and Dunnett'sprocedure using a SigmaStatt 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; *Chironomus tentans*; Larvae
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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; *Chironomus tentans*; Larvae
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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 usingthe trimmed Spearman–Karber method. Dry weight datawere analyzed by one-way analysis of variance and Dunnett'sprocedure using a SigmaStatt 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:	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; *Daphnia magna*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Immobilization
Chemical:	Diisobutyl Phthalate- Parent compound
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	The 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	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; *Daphnia magna*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Immobilization
Chemical:	Diisobutyl Phthalate- Parent compound
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:	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; *Daphnia magna*; Juvenile
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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; *Daphnia magna*; Juvenile
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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 estimatedusing 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:	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 Daphnia magna. 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; *Daphnia magna*; Juvenile
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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	Chemical purity was 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. 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 Daphnia magna. 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; *Daphnia magna*; Juvenile
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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:	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; *Daphnia magna*; Juvenile
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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				

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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; *Daphnia magna*; Juvenile
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
HERO ID:	1336024

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	Low	Results for this criterion should be related to the solvent control.There was statistically-significant morality 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; *Daphnia magna*; Juvenile
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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; *Daphnia magna*; Juvenile
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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 morality 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 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:	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; *Daphnia magna*; Juvenile
Health Outcome:	Reproductive/Teratogenic
Chemical:	Diisobutyl Phthalate- Parent compound
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				
	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: 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; *Daphnia magna*; Juvenile
Health Outcome:	Reproductive/Teratogenic
Chemical:	Diisobutyl Phthalate- Parent compound
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 morality 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 clean control.

Additional Comments: This form is for D. manga 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:	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 Daphnia magna. 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; *Daphnia magna*; Adult
Health Outcome:	Reproductive/Teratogenic
Chemical:	Diisobutyl Phthalate- Parent compound
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				

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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 Daphnia magna. 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; *Daphnia magna*; Adult
Health Outcome:	Reproductive/Teratogenic
Chemical:	Diisobutyl Phthalate- Parent compound
HERO ID:	4829279

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	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:	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 Daphnia magna. 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; *Daphnia magna*; Adult
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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	Chemical purity was 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, 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				
	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:	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 Daphnia magna. 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; *Daphnia magna*; Adult
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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: molting frequency, generational effects also reported

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; *Hyalella azteca*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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.

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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; *Hyalella azteca*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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 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 usingthe trimmed Spearman–Karber method. Dry weight datawere analyzed by one-way analysis of variance and Dunnett'sprocedure using a SigmaStatt 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., 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; *Hyalella azteca*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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.
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; *Hyalella azteca*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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 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 usingthe trimmed Spearman–Karber method. Dry weight datawere analyzed by one-way analysis of variance and Dunnett'sprocedure using a SigmaStatt 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:	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; *Paratanytarsus parthenogenetica*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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; *Paratanytarsus parthenogenetica*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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 consistant 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 Paratanytarsus parthenogenica (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; *Paratanytarsus parthenogenica*; Larvae
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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 Paratanytarsus parthenogenica (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; *Paratanytarsus parthenogenica*; Larvae
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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:	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; *Selenastrum capricornutum*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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; *Selenastrum capricornutum*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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 consistant 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:	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.
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); *Animalia*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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.

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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. Aquatic Toxicology 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); *Animalia*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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. Aquatic Toxicology 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); *Animalia*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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.
	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. Aquatic Toxicology 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); *Animalia*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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. Aquatic Toxicology 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); *Annelida*; multiple species; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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.

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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. Aquatic Toxicology 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); *Annelida*; multiple species; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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. Aquatic Toxicology 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); *Annelida*; multiple species; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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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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); *Annelida*; multiple species; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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

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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; *Arthropoda*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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.
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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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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.
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Taxa, Species, Age:	Invertebrate; Other Invertebrate (e.g., sea urchins, ciliates, rotifers); *Chordata*; multiple species; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
HERO ID:	5495608

Domain	Metric		Rating	Comments
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Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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.
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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.
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Additional Comments: None

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Chemical:	Diisobutyl Phthalate- Parent compound
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Domain	Metric		Rating	Comments
Domain 1: Test Substance				
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Domain 2: Test Design				
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Chemical:	Diisobutyl Phthalate- Parent compound
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Domain 6: Confounding / Variable Control				
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Overall Quality Determination — Medium

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Taxa, Species, Age:	Invertebrate; Other Invertebrate (e.g., sea urchins, ciliates, rotifers); *Coelenterata*; Actinaria; Not Applicable (e.g., fungi or algae studies) or Not Reported
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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); *Coelenterata*; Actinaria; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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. Aquatic Toxicology 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); *Echinodermata*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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. Aquatic Toxicology 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); *Echinodermata*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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. Aquatic Toxicology 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); *Echinodermata*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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. Aquatic Toxicology 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); *Echinodermata*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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. Aquatic Toxicology 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; *Mollusca*; multiple species; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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. Aquatic Toxicology 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; *Mollusca*; multiple species; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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. Aquatic Toxicology 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; *Mollusca*; multiple species; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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. Aquatic Toxicology 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; *Mollusca*; multiple species; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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; *Mysidopsis bahia*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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. 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; *Mysidopsis bahia*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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 consistant 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:	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.
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); *Rhynchocoela*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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. Aquatic Toxicology 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); *Rhynchocoela*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	Diisobutyl Phthalate- Parent compound
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:	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 (Alburnus alburnus) and the harpacticoid Nitocra spinipes. 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; *Nitocra spinipes*; Adult
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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 (Alburnus alburnus) and the harpacticoid Nitocra spinipes. 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; *Nitocra spinipes*; Adult
Health Outcome:	Mortality
Chemical:	Diisobutyl Phthalate- Parent compound
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