United States
Environmental Protection Agency

May 2026
Office of Chemical Safety and
Pollution Prevention

Draft Data Quality Evaluation Information for Environmental Hazard for 4,4'-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)

Systematic Review Support Document for the Draft Risk Evaluation

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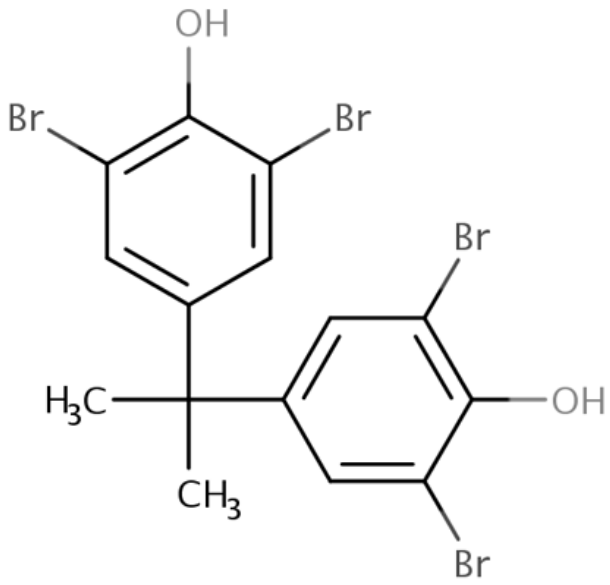

May 2026

This supplemental file contains information regarding the data quality evaluation results relevant to the analysis of environmental hazard for the *Draft Human Health and Environmental Hazard Assessment for 4,4'-(1-Methylethylidene)bis[2, 6-dibromophenol] (TBBPA)*. 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 performs data quality evaluation as a part of the TSCA systematic review process described in the *Draft Systematic Review Protocol Supporting TSCA Risk Evaluations for Chemical Substances* (referred to hereafter as the '2021 Draft Systematic Review Protocol'). The systematic review steps are further described in the *Draft Systematic Review Protocol for 4,4'-(1-Methylethylidene)bis[2, 6-dibromophenol] (TBBPA)*.

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

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HERO ID	Reference	Page
TBBPA		
Habitat: Aquatic (freshwater)		
Taxa: Vertebrates		
Carassius auratus		
2562857	Yang, S. W., Yan, Z. G., Xu, F. F., Wang, S. R., Wu, F. C. (2012). Development of freshwater aquatic life criteria for tetrabromobisphenol A in China. Environmental Pollution 169:59-63.	**19**
3350555	He, Q., Wang, X., Sun, P., Wang, Z., Wang, L. (2015). Acute and chronic toxicity of tetrabromobisphenol A to three aquatic species under different pH conditions. Aquatic Toxicology 164:145-154.	**21**
2562857	Yang, S. W., Yan, Z. G., Xu, F. F., Wang, S. R., Wu, F. C. (2012). Development of freshwater aquatic life criteria for tetrabromobisphenol A in China. Environmental Pollution 169:59-63.	**27**
Danio rerio		
5518140	Carlsson, G., Norrgren, L. (2014). Comparison of embryo toxicity using two classes of aquatic vertebrates. Environmental Toxicology and Pharmacology 37(1):24-7. [Environmental toxicology and pharmacology].	**29**
1401639	Chan, W. K., Chan, K. M. (2012). Disruption of the hypothalamic-pituitary-thyroid axis in zebrafish embryo-larvae following waterborne exposure to BDE-47, TBBPA and BPA. Aquatic Toxicology 108:106-111.	**35**
4933778	Chen, J., Tanguay, R. L., Xiao, Y., Haggard, D. E., Ge, X., Jia, Y., Zheng, Y., Dong, Q., Huang, C., Lin, K. (2016). TBBPA exposure during a sensitive developmental window produces neurobehavioral changes in larval zebrafish. Environmental Pollution 216:53-63.	**41**
5804500	Chow, W. S., Chan, W., Chan, K. M. (2013). Toxicity assessment and vitellogenin expression in zebrafish (Danio rerio) embryos and larvae acutely exposed to bisphenol A, endosulfan, heptachlor, methoxychlor and tetrabromobisphenol A. Journal of Applied Toxicology 33(7):670-678.	**57**
3858252	Godfrey, A., Abdel-Moneim, A., Sepúlveda, M. S. (2017). Acute mixture toxicity of halogenated chemicals and their next generation counterparts on zebrafish embryos. Chemosphere 181:710-712.	**65**
1927732	Hu, J., Liang, Y., Chen, M., Wang, X. (2009). Assessing the toxicity of TBBPA and HBCD by zebrafish embryo toxicity assay and biomarker analysis. Environmental Toxicology 24(4):334-342.	**67**
4180931	Jarema, K. A., Hunter, D. L., Shaffer, R. M., Behl, M., Padilla, S. (2015). Acute and developmental behavioral effects of flame retardants and related chemicals in zebrafish. Neurotoxicology and Teratology 52(Pt B):194-209.	**73**
3981891	Kalasekar, S. M., Zacharia, E., Kessler, N., Ducharme, N. A., Gustafsson, J. Å., Kakadiaris, I. A., Bondesson, M. (2015). Identification of environmental chemicals that induce yolk malabsorption in zebrafish using automated image segmentation. Reproductive Toxicology 55:20-29.	**77**

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5518890	Lee, S. K., Freitag, D., Steinberg, C., Kettrup, A., Kim, Y. H. (1993). Effects of dissolved humic materials on acute toxicity of some organic chemicals to aquatic organisms. Water Research 27(2):199-204.	**81**
5489020	Mccormick, J. M. (2010). Microbial transformations of tetrabromobisphenol a and its metabolites, and their impact on toxicity to the developing zebrafish (Danio rerio) embryo. Doctoral Dissertation:191.	**83**
1294687	Mccormick, J. M., Paiva, M. S., Häggblom, M. M., Cooper, K. R., White, L. A. (2010). Embryonic exposure to tetrabromobisphenol A and its metabolites, bisphenol A and tetrabromobisphenol A dimethyl ether disrupts normal zebrafish (Danio rerio) development and matrix metalloproteinase expression. Aquatic Toxicology 100(3):255-262.	**85**
5496817	Parsons, A., Lange, A., Hutchinson, T. H., Miyagawa, S., Iguchi, T., Kudoh, T., Tyler, C. R. (2019). Molecular mechanisms and tissue targets of brominated flame retardants, BDE-47 and TBBPA, in embryo-larval life stages of zebrafish (Danio rerio). Aquatic Toxicology 209:99-112.	**89**
2754370	Song, M., Liang, D., Liang, Y., Chen, M., Wang, F., Wang, H., Jiang, G. (2014). Assessing developmental toxicity and estrogenic activity of halogenated bisphenol A on zebrafish (Danio rerio). Chemosphere 112:275-281.	**93**
4162140	Yang, J., Chan, K. M. (2015). Evaluation of the toxic effects of brominated compounds (BDE-47, 99, 209, TBBPA) and bisphenol A (BPA) using a zebrafish liver cell line, ZFL. Aquatic Toxicology 159:138-147.	**97**
5490833	Yang, S., Wang, S., Sun, F., Zhang, M., Wu, F., Xu, F., Ding, Z. (2015). Protective effects of puerarin against tetrabromobisphenol a-induced apoptosis and cardiac developmental toxicity in zebrafish embryo-larvae. Environmental Toxicology 30(9):1014-1023.	**103**
4199233	Baumann, L., Ros, A., Rehberger, K., Neuhauss, S. C., Segner, H. (2016). Thyroid disruption in zebrafish (Danio rerio) larvae: Different molecular response patterns lead to impaired eye development and visual functions. Aquatic Toxicology 172:44-55.	**111**
3479540	Behl, M., Hsieh, J. H., Shafer, T. J., Mundy, W. R., Rice, J. R., Boyd, W. A., Freedman, J. H., Hunter, E. S., Jarema, K. A., Padilla, S., Tice, R. R. (2015). Use of alternative assays to identify and prioritize organophosphorus flame retardants for potential developmental and neurotoxicity. Neurotoxicology and Teratology 52(Pt B):181-193.	**117**
5549558	Fraser, K., T.W., Khezri, A., Jusdado, H., J.G., Lewandowska-Sabat, A. M., Henry, T., Ropstad, E. (2017). Toxicant induced behavioural aberrations in larval zebrafish are dependent on minor methodological alterations. Toxicology Letters 276:62-68.	**121**
4167836	Godfrey, A., Hooser, B., Abdelmoneim, A., Horzmann, K. A., Freemanc, J. L., Sepúlveda, M. S. (2017). Thyroid disrupting effects of halogenated and next generation chemicals on the swim bladder development of zebrafish. Aquatic Toxicology 193:228-235.	**127**
5500466	Li, L., Bonneton, F., Tohme, M., Bernard, L., Chen, X. Y., Laudet, V. (2016). In Vivo Screening Using Transgenic Zebrafish Embryos Reveals New Effects of HDAC Inhibitors Trichostatin A and Valproic Acid on Organogenesis. PLoS ONE 11(2):e0149497.	**129**
1294687	Mccormick, J. M., Paiva, M. S., Häggblom, M. M., Cooper, K. R., White, L. A. (2010). Embryonic exposure to tetrabromobisphenol A and its metabolites, bisphenol A and tetrabromobisphenol A dimethyl ether disrupts normal zebrafish (Danio rerio) development and matrix metalloproteinase expression. Aquatic Toxicology 100(3):255-262.	**131**
2953504	Noyes, P. D., Haggard, D. E., Gonnerman, G. D., Tanguay, R. L. (2015). Advanced morphological - behavioral test platform reveals neurodevelopmental defects in embryonic zebrafish exposed to comprehensive suite of halogenated and organophosphate flame retardants. Toxicological Sciences 145(1):177-195.	**141**
5496817	Parsons, A., Lange, A., Hutchinson, T. H., Miyagawa, S., Iguchi, T., Kudoh, T., Tyler, C. R. (2019). Molecular mechanisms and tissue targets of brominated flame retardants, BDE-47 and TBBPA, in embryo-larval life stages of zebrafish (Danio rerio). Aquatic Toxicology 209:99-112.	**148**
2754370	Song, M., Liang, D., Liang, Y., Chen, M., Wang, F., Wang, H., Jiang, G. (2014). Assessing developmental toxicity and estrogenic activity of halogenated bisphenol A on zebrafish (Danio rerio). Chemosphere 112:275-281.	**150**

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Gambusia affinis

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Lepomis macrochirus

5549811 Union Carbide Corporation Environmental Services, (1978). Acute toxicity of FMBP4A (tetrabromobisphenol A) to the bluegill sunfish, Lepomis macrochirus rafinesque. **258**

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1294788 Debenest, T., Gagné, F., Petit, A. N., André, C., Kohli, M., Blaise, C. (2010). Ecotoxicity of a brominated flame retardant (tetrabromobisphenol A) and its derivatives to aquatic organisms. Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 152(4):407-412. **266**

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Oryzias melastigma

5555779 Ye, G., Chen, Y., Wang, H. O., Ye, T., Lin, Y., Huang, Q., Chi, Y., Dong, S. (2016). Metabolomics approach reveals metabolic disorders and potential biomarkers associated with the developmental toxicity of tetrabromobisphenol A and tetrachlorobisphenol A. Scientific Reports 6:35257. **282**

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Pimephelas promelas

10064188 Battelle, (2018). Fish short-term reproduction assay of 3,3',5,5'-tetrabromobisphenol A (TBBPA) in Pimephales promelas. **301**

Pseudacris regilla

1414891 Veldhoen, N., Boggs, A., Walzak, K., Helbing, C. C. (2006). Exposure to tetrabromobisphenol-A alters TH-associated gene expression and tadpole metamorphosis in the Pacific tree frog Pseudacris regilla. Aquatic Toxicology 78(3):292-302. **313**

Pseudorasbora parva

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Rana arvalis

5518140 Carlsson, G., Norrgren, L. (2014). Comparison of embryo toxicity using two classes of aquatic vertebrates. Environmental Toxicology and Pharmacology 37(1):24-7. [Environmental toxicology and pharmacology]. **319**

Rana limnocharis

2562857 Yang, S. W., Yan, Z. G., Xu, F. F., Wang, S. R., Wu, F. C. (2012). Development of freshwater aquatic life criteria for tetrabromobisphenol A in China. Environmental Pollution 169:59-63. **323**

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Rana rugosa

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Salmo gairdneri

5549807 Union Carbide Corporation Environmental Services, (1978). Acute toxicity of FMBP4A (tetrabromobisphenol A) to the rainbow trout, Salmo gairdneri Richardson. **331**

Salvelinus namaycush

2159104 Jurgella, G. F., Marwah, A., Malison, J. A., Peterson, R., Barry, T. P. (2006). Effects of xenobiotics and steroids on renal and hepatic estrogen metabolism in lake trout. General and Comparative Endrocrinology 148(2):273-281. **335**

Silurana tropicalis

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Xenopus laevis

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1298685 Jagnytsch, O., Opitz, R., Lutz, I., Kloas, W. (2006). Effects of tetrabromobisphenol A on larval development and thyroid hormone-regulated biomarkers of the amphibian Xenopus laevis. Environmental Research 101(3):340-348. **371**

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Xenopus tropicalis

5518140 Carlsson, G., Norrgren, L. (2014). Comparison of embryo toxicity using two classes of aquatic vertebrates. Environmental Toxicology and Pharmacology 37(1):24-7. [Environmental toxicology and pharmacology]. **381**

1403499 Shi, H., Qian, L., Guo, S., Zhang, X., Liu, J., Cao, Q. (2010). Teratogenic effects of tetrabromobisphenol A on Xenopus tropicalis embryos. Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 152(1):62-68. **385**

Taxa: Invertebrates

Caenorhabditis elegans

3479540 Behl, M., Hsieh, J. H., Shafer, T. J., Mundy, W. R., Rice, J. R., Boyd, W. A., Freedman, J. H., Hunter, E. S., Jarema, K. A., Padilla, S., Tice, R. R. (2015). Use of alternative assays to identify and prioritize organophosphorus flame retardants for potential developmental and neurotoxicity. Neurotoxicology and Teratology 52(Pt B):181-193. **391**

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5497653 Ju, J., Lieke, T., Saul, N., Pu, Y., Yin, L., Kochan, C., Putschew, A., Baberschke, N., Steinberg, C. E. (2014). Neurotoxic evaluation of two organobromine model compounds and natural AOBr-containing surface water samples by a Caenorhabditis elegans test. Ecotoxicology and Environmental Safety 104:194-201. **399**

5497487 Liu, F., Zaman, W. Q., Peng, H., Li, C., Cao, X., Huang, K., Cui, C., Zhang, W., Lin, K., Luo, Q. (2019). Ecotoxicity of Caenorhabditis elegans following a step and repeated chronic exposure to tetrabromobisphenol A. Ecotoxicology and Environmental Safety 169:273-281. **401**

5497487 Liu, F., Zaman, W. Q., Peng, H., Li, C., Cao, X., Huang, K., Cui, C., Zhang, W., Lin, K., Luo, Q. (2019). Ecotoxicity of Caenorhabditis elegans following a step and repeated chronic exposure to tetrabromobisphenol A. Ecotoxicology and Environmental Safety 169:273-281. **405**

Chironomus plumosus

2562857 Yang, S. W., Yan, Z. G., Xu, F. F., Wang, S. R., Wu, F. C. (2012). Development of freshwater aquatic life criteria for tetrabromobisphenol A in China. Environmental Pollution 169:59-63. **425**

Chironomus riparius

7415562 Ltd,, W.I. (2005). Tetrabromobisphenol-A (TBBPA): A 28-day sediment toxicity test with Chironomus riparius using spiked sediment. **427**

Chironomus tentans

5547095 Springborn Laboratories, (1989). The subchronic toxicity of sediment-sorbed tetrabromobisphenol A to Chironomus tentans under flow-through conditions (final report) with cover sheet and letter dated 101689. **433**

Daphnia magna

3350555 He, Q., Wang, X., Sun, P., Wang, Z., Wang, L. (2015). Acute and chronic toxicity of tetrabromobisphenol A to three aquatic species under different pH conditions. Aquatic Toxicology 164:145-154. **455**

5518890 Lee, S. K., Freitag, D., Steinberg, C., Kettrup, A., Kim, Y. H. (1993). Effects of dissolved humic materials on acute toxicity of some organic chemicals to aquatic organisms. Water Research 27(2):199-204. **457**

5352358 Ltd,, W.I. (2003). Tetrabromobisphenol A: A 48-hour flow-through acute toxicity test with the cladoceran (Daphnia magna). **459**

5546496 Union Carbide Corporation Environmental Services, (1978). Acute toxicity of FMBP4A (Tetrabromobisphenol A) to the water flea Daphnia magna straus. **461**

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Dugesia japonica

10064285 Zhang, S., Hagstrom, D., Hayes, P., Graham, A., Collins, E. S. (2019). Multi-behavioral endpoint testing of an 87-chemical compound library in freshwater planarians. Toxicological Sciences 167(1):26-44. **479**

5469417 Zhang, S., Ireland, D., Sipes, N. S., Behl, M., Collins, E. S. (2019). Screening for neurotoxic potential of 15 flame retardants using freshwater planarians. Neurotoxicology and Teratology 73:54-66. **481**

10064285 Zhang, S., Hagstrom, D., Hayes, P., Graham, A., Collins, E. S. (2019). Multi-behavioral endpoint testing of an 87-chemical compound library in freshwater planarians. Toxicological Sciences 167(1):26-44. **496**

5469417 Zhang, S., Ireland, D., Sipes, N. S., Behl, M., Collins, E. S. (2019). Screening for neurotoxic potential of 15 flame retardants using freshwater planarians. Neurotoxicology and Teratology 73:54-66. **504**

Hyalella azteca

7415563 Ltd,, W.I. (2006). Tetrabromobisphenol-A (TBBPA): A prolonged sediment toxicity test with Hyalella azteca using spiked sediment. **516**

Hydra attenuata a

1294788 Debenest, T., Gagné, F., Petit, A. N., André, C., Kohli, M., Blaise, C. (2010). Ecotoxicity of a brominated flame retardant (tetrabromobisphenol A) and its derivatives to aquatic organisms. Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 152(4):407-412. **520**

Limnodrilus hoffmeisteri

3350555 He, Q., Wang, X., Sun, P., Wang, Z., Wang, L. (2015). Acute and chronic toxicity of tetrabromobisphenol A to three aquatic species under different pH conditions. Aquatic Toxicology 164:145-154. **522**

2562857 Yang, S. W., Yan, Z. G., Xu, F. F., Wang, S. R., Wu, F. C. (2012). Development of freshwater aquatic life criteria for tetrabromobisphenol A in China. Environmental Pollution 169:59-63. **524**

Lumbriculus variegatus

7769074 Ltd,, W.I. (2002). Tetrabromobisphenol A: A prolonged sediment toxicity test with Lumbriculus variegatus using spiked sediment with 2% total organic carbon. **526**

7769200 Ltd,, W.I. (2002). Tetrabromobisphenol A: A prolonged sediment toxicity test with Lumbriculus variegatus using spiked sediment with 5% total organic carbon. **530**

Macrobrachium nipponense

2562857 Yang, S. W., Yan, Z. G., Xu, F. F., Wang, S. R., Wu, F. C. (2012). Development of freshwater aquatic life criteria for tetrabromobisphenol A in China. Environmental Pollution 169:59-63. **534**

Pristina longiseta

12812456 Souza, C.d., T. T., Castro, G. B., Bernegossi, A. C., Felipe, M. C., Pinheiro, F. R., Colombo-Corbi, V., Girolli, D. A., Gorni, G. R., Corbi, J. J. (2023). Pristina longiseta reproduction test: Chronic exposure to environmental contaminants. Environmental Science and Pollution Research 30(9):23578-23588. **536**

Thamnocephalus platyurus

1294788 Debenest, T., Gagné, F., Petit, A. N., André, C., Kohli, M., Blaise, C. (2010). Ecotoxicity of a brominated flame retardant (tetrabromobisphenol A) and its derivatives to aquatic organisms. Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 152(4):407-412. **539**

Taxa: Plants (Non-vascular)

Chlamydomonas reinhardtii

5496905 Debenest, T., Gagné, F., Petit, A. N., Kohli, M., Eullafroy, P., Blaise, C. (2010). Monitoring of a flame retardant (tetrabromobisphenol A) toxicity on different microalgae assessed by flow cytometry. Journal of Environmental Monitoring 12(10):1918-1923. **541**

Chlorella pyrenoidosa

2523760 Peng, F.,uQ, Ying, G., Yang, B.,in, Liu, Y., Lai, H., Zhou, G., Chen, J.,un, Zhao, J. L. (2014). Biotransformation of the flame retardant tetrabromobisphenol-a (TBBPA) by freshwater microalgae. Environmental Toxicology and Chemistry 33(8):1705-1711. **545**

2523760 Peng, F.,uQ, Ying, G., Yang, B.,in, Liu, Y., Lai, H., Zhou, G., Chen, J.,un, Zhao, J. L. (2014). Biotransformation of the flame retardant tetrabromobisphenol-a (TBBPA) by freshwater microalgae. Environmental Toxicology and Chemistry 33(8):1705-1711. **547**

Coelastrum sphaericum

2523760 Peng, F.,uQ, Ying, G., Yang, B.,in, Liu, Y., Lai, H., Zhou, G., Chen, J.,un, Zhao, J. L. (2014). Biotransformation of the flame retardant tetrabromobisphenol-a (TBBPA) by freshwater microalgae. Environmental Toxicology and Chemistry 33(8):1705-1711. **551**

Nitzschia palea

5496905 Debenest, T., Gagné, F., Petit, A. N., Kohli, M., Eullafroy, P., Blaise, C. (2010). Monitoring of a flame retardant (tetrabromobisphenol A) toxicity on different microalgae assessed by flow cytometry. Journal of Environmental Monitoring 12(10):1918-1923. **553**

4509862 Debenest, T., Petit, A. N., Gagné, F., Kohli, M., Nguyen, N., Blaise, C. (2011). Comparative toxicity of a brominated flame retardant (tetrabromobisphenol A) on microalgae with single and multi-species bioassays. Chemosphere 85(1):50-55. **557**

Pseudokirchneriella subcapitata

1294788 Debenest, T., Gagné, F., Petit, A. N., André, C., Kohli, M., Blaise, C. (2010). Ecotoxicity of a brominated flame retardant (tetrabromobisphenol A) and its derivatives to aquatic organisms. Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 152(4):407-412. **559**

5496905 Debenest, T., Gagné, F., Petit, A. N., Kohli, M., Eullafroy, P., Blaise, C. (2010). Monitoring of a flame retardant (tetrabromobisphenol A) toxicity on different microalgae assessed by flow cytometry. Journal of Environmental Monitoring 12(10):1918-1923. **561**

4509862 Debenest, T., Petit, A. N., Gagné, F., Kohli, M., Nguyen, N., Blaise, C. (2011). Comparative toxicity of a brominated flame retardant (tetrabromobisphenol A) on microalgae with single and multi-species bioassays. Chemosphere 85(1):50-55. **565**

2523760 Peng, F.,uQ, Ying, G., Yang, B.,in, Liu, Y., Lai, H., Zhou, G., Chen, J.,un, Zhao, J. L. (2014). Biotransformation of the flame retardant tetrabromobisphenol-a (TBBPA) by freshwater microalgae. Environmental Toxicology and Chemistry 33(8):1705-1711. **567**

5497680 Sazawa, K., Furuhashi, Y., Hata, N., Taguchi, S., Fukushima, M., Kuramitz, H. (2013). Evaluation of the toxicity of tetrabromobisphenol A and some of its oxidation products using a micro-scale algal growth inhibition test. Toxicological and Environmental Chemistry 95(3):472-482. **569**

Scenedesmus acuminatus

2523760 Peng, F.,uQ, Ying, G., Yang, B.,in, Liu, Y., Lai, H., Zhou, G., Chen, J.,un, Zhao, J. L. (2014). Biotransformation of the flame retardant tetrabromobisphenol-a (TBBPA) by freshwater microalgae. Environmental Toxicology and Chemistry 33(8):1705-1711. **571**

Scenedesmus obliquus

5497133 Meng, Y., Wang, S., Wang, Z., Ye, N., Fang, H. (2018). Algal toxicity of binary mixtures of zinc oxide nanoparticles and tetrabromobisphenol A: Roles of dissolved organic matters. Environmental Toxicology and Pharmacology 64:78-85. **573**

2523760 Peng, F.,uQ, Ying, G., Yang, B.,in, Liu, Y., Lai, H., Zhou, G., Chen, J.,un, Zhao, J. L. (2014). Biotransformation of the flame retardant tetrabromobisphenol-a (TBBPA) by freshwater microalgae. Environmental Toxicology and Chemistry 33(8):1705-1711. **577**

2523760 Peng, F.,uQ, Ying, G., Yang, B.,in, Liu, Y., Lai, H., Zhou, G., Chen, J.,un, Zhao, J. L. (2014). Biotransformation of the flame retardant tetrabromobisphenol-a (TBBPA) by freshwater microalgae. Environmental Toxicology and Chemistry 33(8):1705-1711. **579**

Scenedesmus quadricanda

2562857 Yang, S. W., Yan, Z. G., Xu, F. F., Wang, S. R., Wu, F. C. (2012). Development of freshwater aquatic life criteria for tetrabromobisphenol A in China. Environmental Pollution 169:59-63. **583**

Scenedesmus quadricauda

2523760 Peng, F.,uQ, Ying, G., Yang, B.,in, Liu, Y., Lai, H., Zhou, G., Chen, J.,un, Zhao, J. L. (2014). Biotransformation of the flame retardant tetrabromobisphenol-a (TBBPA) by freshwater microalgae. Environmental Toxicology and Chemistry 33(8):1705-1711. **585**

Selenastrum capricornutum

5547583 Springborn Laboratories, (1988). Acute toxicity of tetrabromobisphenol A to fathead minnow (Pimephales promelas) under flow-through conditions with attachments and cover letter dated 111788. **587**

Taxa: Plants (Vascular)

Ceratophyllum demersum

4181234 Sun, Y., Guo, H., Yu, H., Wang, X., Wu, J., Xue, Y. (2008). Bioaccumulation and physiological effects of tetrabromobisphenol A in coontail Ceratophyllum demersum L. Chemosphere 70(10):1787-1795. **589**

Lemna gibba

12155422 Agroscience,, E.E., LLC, (2023). TBBPA: A 7-day static-renewal toxicity test with duckweed (Lemna gibba G3). **593**

Spinacia oleracea

1294788 Debenest, T., Gagné, F., Petit, A. N., André, C., Kohli, M., Blaise, C. (2010). Ecotoxicity of a brominated flame retardant (tetrabromobisphenol A) and its derivatives to aquatic organisms. Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 152(4):407-412. **600**

Habitat: Aquatic (marine)

Taxa: Vertebrates

Oryzias melastigma

5499513 Huang, Q., Chen, Y., Lin, L., Liu, Y., Chi, Y., Lin, Y., Ye, G., Zhu, H., Dong, S. (2017). Different effects of bisphenol a and its halogenated derivatives on the reproduction and development of Oryzias melastigma under environmentally relevant doses. Science of the Total Environment 595:752-758. **602**

5499513 Huang, Q., Chen, Y., Lin, L., Liu, Y., Chi, Y., Lin, Y., Ye, G., Zhu, H., Dong, S. (2017). Different effects of bisphenol a and its halogenated derivatives on the reproduction and development of Oryzias melastigma under environmentally relevant doses. Science of the Total Environment 595:752-758. **610**

Taxa: Invertebrates

Acartia pacifica

3350491 Gong, W., Zhu, L., Hao, Y. (2016). Lethal and sublethal toxicity comparison of BFRs to three marine planktonic copepods: Effects on survival, metabolism and ingestion. PLoS ONE 11(1):e0147790. **622**

Acartia tonsa

2819235 Wollenberger, L., Dinan, L., Breitholtz, M. (2005). Brominated flame retardants: Activities in a crustacean development test and in an ecdysteroid screening assay. Environmental Toxicology and Chemistry 24(2):400-407. **628**

2819235 Wollenberger, L., Dinan, L., Breitholtz, M. (2005). Brominated flame retardants: Activities in a crustacean development test and in an ecdysteroid screening assay. Environmental Toxicology and Chemistry 24(2):400-407. **630**

Crassostrea virginica

5547102 Springborn Laboratories, (1989). Acute toxicity of tetrabromobisphenol A to eastern oysters (Crassostrea virginica) under flow-through conditions with cover letters dated 021489 and 021789. **634**

5547108 Springborn Laboratories, (1989). Bioconcentration and elimination of C-residues by eastern oysters (Crassostrea virginica) exposed to tetrabromobisphenol A (final report) with attached studies and letter 081889. **636**

Mytilus edulis

6311092 Brixham Laboratory, (2005). TBBPA: Determination of effects on growth of the common mussel Mytilus edulis. **638**

Mytilus galloprovincialis

3981747 Fabbri, R., Montagna, M., Balbi, T., Raffo, E., Palumbo, F., Canesi, L. (2014). Adaptation of the bivalve embryotoxicity assay for the high throughput screening of emerging contaminants in Mytilus galloprovincialis. Marine Environmental Research 99:1-8. **642**

5497037 Ji, C., Li, F., Wang, Q., Zhao, J., Sun, Z., Wu, H. (2016). An integrated proteomic and metabolomic study on the gender-specific responses of mussels Mytilus galloprovincialis to tetrabromobisphenol A (TBBPA). Chemosphere 144:527-539. **644**

Oithona similis

3350491 Gong, W., Zhu, L., Hao, Y. (2016). Lethal and sublethal toxicity comparison of BFRs to three marine planktonic copepods: Effects on survival, metabolism and ingestion. PLoS ONE 11(1):e0147790. **650**

Psammechinus miliaris

1274149 Anselmo, R., H.M., Koerting, L., Devito, S., Berg, v.d., J., J.H., Dubbeldam, M., Kwadijk, C., Murk, A. J. (2011). Early life developmental effects of marine persistent organic pollutants on the sea urchin Psammechinus miliaris. Ecotoxicology and Environmental Safety 74(8):2182-2192. **656**

Pseudodiaptomus inopinus

3350491 Gong, W., Zhu, L., Hao, Y. (2016). Lethal and sublethal toxicity comparison of BFRs to three marine planktonic copepods: Effects on survival, metabolism and ingestion. PLoS ONE 11(1):e0147790. **664**

Pseudodiaptomus inopinus.

5519030 Gong, W.,j, Zhu, L.,y, Jiang, T.,t, Han, C. (2017). The occurrence and spatial-temporal distribution of tetrabromobisphenol A in the coastal intertidal zone of Qingdao in China, with a focus on toxicity assessment by biological monitoring. Chemosphere 185:462-467. **670**

Ruditapes phillipinarum

5635811 Jiang, S., Miao, J., Wang, X., Liu, P., Pan, L. (2019). Inhibition of growth in juvenile manila clam Ruditapes philippinarum: Potential adverse outcome pathway of TBBPA. Chemosphere 224:588-596. **674**

5635811 Jiang, S., Miao, J., Wang, X., Liu, P., Pan, L. (2019). Inhibition of growth in juvenile manila clam Ruditapes philippinarum: Potential adverse outcome pathway of TBBPA. Chemosphere 224:588-596. **677**

5635811 Jiang, S., Miao, J., Wang, X., Liu, P., Pan, L. (2019). Inhibition of growth in juvenile manila clam Ruditapes philippinarum: Potential adverse outcome pathway of TBBPA. Chemosphere 224:588-596. **681**

Taxa: Plants (Non-vascular)

Skeletonema costatum

1927837 Walsh, G. E., Yoder, M. J., Mclaughlin, L. L., Lores, E. M. (1987). Responses of marine unicellular algae to brominated organic compounds in six growth media. Ecotoxicology and Environmental Safety 14(3):215-222. **683**

Thalassiosira pseudonana

1927837 Walsh, G. E., Yoder, M. J., Mclaughlin, L. L., Lores, E. M. (1987). Responses of marine unicellular algae to brominated organic compounds in six growth media. Ecotoxicology and Environmental Safety 14(3):215-222. **698**

Habitat: Aquatic (brackish)

Taxa: Vertebrates

Oryzias melastigma

5555779 Ye, G., Chen, Y., Wang, H. O., Ye, T., Lin, Y., Huang, Q., Chi, Y., Dong, S. (2016). Metabolomics approach reveals metabolic disorders and potential biomarkers associated with the developmental toxicity of tetrabromobisphenol A and tetrachlorobisphenol A. Scientific Reports 6:35257. **716**

Platichthys flesus

1412802 Kuiper, R. V., Cantón, R. F., Leonards, P. E., Jenssen, B. M., Dubbeldam, M., Wester, P. W., Berg, v.d., M., Vos, J. G., Vethaak, A. D. (2007). Long-term exposure of European flounder (Platichthys flesus) to the flame-retardants tetrabromobisphenol A (TBBPA) and hexabromocyclododecane (HBCD). Ecotoxicology and Environmental Safety 67(3):349-360. **731**

Taxa: Invertebrates

Mysidopsis bahia

5757963 Goodman, L. R., Cripe, G. M., Moody, P. H., Halsell, D. G. (1988). Acute toxicity of malathion, tetrabromobisphenol-a, and tirbutyltin chloride to mysids (mysidopsis bahia) of three ages. Bulletin of Environmental Contamination and Toxicology 41(5):746. **747**

Habitat: Terrestrial

Taxa: Vertebrates

Coturnix japonica

1416717 Halldin, K., Berg, C., Bergman, A., Brandt, I., Brunström, B. (2001). Distribution of bisphenol A and tetrabromobisphenol A in quail eggs, embryos and laying birds and studies on reproduction variables in adults following in ovo exposure. Archives of Toxicology 75(10):597-603. **753**

1416717 Halldin, K., Berg, C., Bergman, A., Brandt, I., Brunström, B. (2001). Distribution of bisphenol A and tetrabromobisphenol A in quail eggs, embryos and laying birds and studies on reproduction variables in adults following in ovo exposure. Archives of Toxicology 75(10):597-603. **755**

1416653 Berg, C., Halldin, K., Brunström, B. (2001). Effects of bisphenol A and tetrabromobisphenol A on sex organ development in quail and chicken embryos. Environmental Toxicology and Chemistry 20(12):2836-2840. **759**

1416717 Halldin, K., Berg, C., Bergman, A., Brandt, I., Brunström, B. (2001). Distribution of bisphenol A and tetrabromobisphenol A in quail eggs, embryos and laying birds and studies on reproduction variables in adults following in ovo exposure. Archives of Toxicology 75(10):597-603. **763**

1416717 Halldin, K., Berg, C., Bergman, A., Brandt, I., Brunström, B. (2001). Distribution of bisphenol A and tetrabromobisphenol A in quail eggs, embryos and laying birds and studies on reproduction variables in adults following in ovo exposure. Archives of Toxicology 75(10):597-603. **765**

Danio rerio

1293799 Fini, J. B., Riu, A., Debrauwer, L., Hillenweck, A., Mével, Le, S., Chevolleau, S., Boulahtouf, A., Palmier, K., Balaguer, P., Cravedi, J. P., Demeneix, B. A., Zalko, D. (2012). Parallel biotransformation of tetrabromobisphenol A in Xenopus laevis and mammals: Xenopus as a model for endocrine perturbation studies. Toxicological Sciences 125(2):359-367. **769**

Gallus domesticus

1416653 Berg, C., Halldin, K., Brunström, B. (2001). Effects of bisphenol A and tetrabromobisphenol A on sex organ development in quail and chicken embryos. Environmental Toxicology and Chemistry 20(12):2836-2840. **772**

Mus musculus

3479540 Behl, M., Hsieh, J. H., Shafer, T. J., Mundy, W. R., Rice, J. R., Boyd, W. A., Freedman, J. H., Hunter, E. S., Jarema, K. A., Padilla, S., Tice, R. R. (2015). Use of alternative assays to identify and prioritize organophosphorus flame retardants for potential developmental and neurotoxicity. Neurotoxicology and Teratology 52(Pt B):181-193. **776**

Rana nigromaculata

5547039 Zhang, H., Liu, W., Chen, B., He, J., Chen, F., Shan, X., Du, Q., Li, N., Jia, X., Tang, J. (2018). Differences in reproductive toxicity of TBBPA and TCBPA exposure in male Rana nigromaculata. Environmental Pollution 243(Pt A):394-403. **778**

Rattus norvegicus

3479540 Behl, M., Hsieh, J. H., Shafer, T. J., Mundy, W. R., Rice, J. R., Boyd, W. A., Freedman, J. H., Hunter, E. S., Jarema, K. A., Padilla, S., Tice, R. R. (2015). Use of alternative assays to identify and prioritize organophosphorus flame retardants for potential developmental and neurotoxicity. Neurotoxicology and Teratology 52(Pt B):181-193. **782**

Xenopus laevis

1293799 Fini, J. B., Riu, A., Debrauwer, L., Hillenweck, A., Mével, Le, S., Chevolleau, S., Boulahtouf, A., Palmier, K., Balaguer, P., Cravedi, J. P., Demeneix, B. A., Zalko, D. (2012). Parallel biotransformation of tetrabromobisphenol A in Xenopus laevis and mammals: Xenopus as a model for endocrine perturbation studies. Toxicological Sciences 125(2):359-367. **786**

Taxa: Invertebrates

Caenorhabditis elegans

11365410 Li, J., Dai, L., Feng, Y., Cao, Z., Ding, Y., Xu, H., Xu, A., Du, H. (2024). Multigenerational effects and mutagenicity of three flame retardants on germ cells in Caenorhabditis elegans. Ecotoxicology and Environmental Safety 269:115815. **789**

5555762 Lieke, T., Steinberg, C. E., Ju, J., Saul, N. (2015). Natural Marine and Synthetic Xenobiotics Get on Nematode's Nerves: Neuro-Stimulating and Neurotoxic Findings in Caenorhabditis elegans. Marine Drugs 13(5):2785-812. [Marine drugs]. **797**

5497487 Liu, F., Zaman, W. Q., Peng, H., Li, C., Cao, X., Huang, K., Cui, C., Zhang, W., Lin, K., Luo, Q. (2019). Ecotoxicity of Caenorhabditis elegans following a step and repeated chronic exposure to tetrabromobisphenol A. Ecotoxicology and Environmental Safety 169:273-281. **815**

5549514 Saul, N., Baberschke, N., Chakrabarti, S., Stürzenbaum, S. R., Lieke, T., Menzel, R., Jonáš, A., Steinberg, C. E. (2014). Two organobromines trigger lifespan, growth, reproductive and transcriptional changes in Caenorhabditis elegans. Environmental Science and Pollution Research 21(17):10419-10431. **819**

5549514 Saul, N., Baberschke, N., Chakrabarti, S., Stürzenbaum, S. R., Lieke, T., Menzel, R., Jonáš, A., Steinberg, C. E. (2014). Two organobromines trigger lifespan, growth, reproductive and transcriptional changes in Caenorhabditis elegans. Environmental Science and Pollution Research 21(17):10419-10431. **824**

5549514 Saul, N., Baberschke, N., Chakrabarti, S., Stürzenbaum, S. R., Lieke, T., Menzel, R., Jonáš, A., Steinberg, C. E. (2014). Two organobromines trigger lifespan, growth, reproductive and transcriptional changes in Caenorhabditis elegans. Environmental Science and Pollution Research 21(17):10419-10431. **828**

11365410 Li, J., Dai, L., Feng, Y., Cao, Z., Ding, Y., Xu, H., Xu, A., Du, H. (2024). Multigenerational effects and mutagenicity of three flame retardants on germ cells in Caenorhabditis elegans. Ecotoxicology and Environmental Safety 269:115815. **831**

5555762 Lieke, T., Steinberg, C. E., Ju, J., Saul, N. (2015). Natural Marine and Synthetic Xenobiotics Get on Nematode's Nerves: Neuro-Stimulating and Neurotoxic Findings in Caenorhabditis elegans. Marine Drugs 13(5):2785-812. [Marine drugs]. **835**

Eisenia fetida

6775143 Gu, J., Chen, X., Wang, Y., Wang, L., Szlavecz, K., Ma, Y., Ji, R. (2020). Bioaccumulation, physiological distribution, and biotransformation of tetrabromobisphenol a (TBBPA) in the geophagous earthworm Metaphire guillelmi - hint for detoxification strategy. Journal of Hazardous Materials 388:122027. **847**

5115416 Chen, X., Gu, X., Zhao, X., Ma, X., Pan, Y., Wang, X., Ji, R. (2018). Species-dependent toxicity, accumulation, and subcellular partitioning of cadmium in combination with tetrabromobisphenol A in earthworms. Chemosphere 210:1042-1050. **849**

2965902 Shi, Y. J., Xu, X. B., Zheng, X. Q., Lu, Y. L. (2015). Responses of growth inhibition and antioxidant gene expression in earthworms (Eisenia fetida) exposed to tetrabromobisphenol A, hexabromocyclododecane and decabromodiphenyl ether. Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 174-175:32-38. **853**

7415561 Ltd,, W.I. (2003). Effect of tetrabromobisphenol A on the survival and reproduction of the earthworm, Eisenia fetida. **857**

6311100 Warbritton, R., Kendall, T. Z., Nixon, W. B. (2005). Effect of tetrabromobisphenol A on the reproduction of the earthworm, Eisenia fetida. **873**

Enchytraeus crypticus

938766 Sverdrup, L. E., Hartnik, T., Mariussen, E., Jensen, J. (2006). Toxicity of three halogenated flame retardants to nitrifying bacteria, red clover (Trifolium pratense), and a soil invertebrate (Enchytraeus crypticus). Chemosphere 64(1):96-103. **881**

Metaphire guillelmi

5115416 Chen, X., Gu, X., Zhao, X., Ma, X., Pan, Y., Wang, X., Ji, R. (2018). Species-dependent toxicity, accumulation, and subcellular partitioning of cadmium in combination with tetrabromobisphenol A in earthworms. Chemosphere 210:1042-1050. **883**

6775143 Gu, J., Chen, X., Wang, Y., Wang, L., Szlavecz, K., Ma, Y., Ji, R. (2020). Bioaccumulation, physiological distribution, and biotransformation of tetrabromobisphenol a (TBBPA) in the geophagous earthworm Metaphire guillelmi - hint for detoxification strategy. Journal of Hazardous Materials 388:122027. **889**

Taxa: Plants (Non-vascular)

Cucurbita sp.

10115348 Hou, X., Wei, L., Tang, Y., Kong, W., Liu, J., Schnoor, J. L., Jiang, G. (2021). Two typical glycosylated metabolites of tetrabromobisphenol A formed in plants: Excretion and deglycosylation in plant root zones. Environmental Science & Technology Letters 8(4):313-319. **897**

Taxa: Plants (Vascular)

Allium cepa

6391028 Ltd,, W.I. (2002). Tetrabromobisphenol-A: A toxicity test to determine the effects of the test substance on seedling emergence of six species of plants. **900**

Avicennia marina

10113700 Jiang, Y., Lu, H., Wang, Y., Hong, H., Wang, Q., Liu, J., Yan, C. (2020). Uptake, biotransformation and physiological response of TBBPA in mangrove plants after hydroponics exposure. Marine Pollution Bulletin 151:110832. **904**

Cucumis sativa

6391028 Ltd,, W.I. (2002). Tetrabromobisphenol-A: A toxicity test to determine the effects of the test substance on seedling emergence of six species of plants. **913**

Glycine max

4687686 Ge, H., Zhang, F. (2017). Effects of Tetrabromobisphenol A Stress on Growth and Physiological Characteristics of Soybean Seedling. Bulletin of Environmental Contamination and Toxicology 98(1):141-146. **917**

6391028 Ltd,, W.I. (2002). Tetrabromobisphenol-A: A toxicity test to determine the effects of the test substance on seedling emergence of six species of plants. **921**

Kandelia obovata

10113700 Jiang, Y., Lu, H., Wang, Y., Hong, H., Wang, Q., Liu, J., Yan, C. (2020). Uptake, biotransformation and physiological response of TBBPA in mangrove plants after hydroponics exposure. Marine Pollution Bulletin 151:110832. **925**

Lolium perenne

6391028 Ltd,, W.I. (2002). Tetrabromobisphenol-A: A toxicity test to determine the effects of the test substance on seedling emergence of six species of plants. **934**

Lycopersicon esculentum

6391028 Ltd,, W.I. (2002). Tetrabromobisphenol-A: A toxicity test to determine the effects of the test substance on seedling emergence of six species of plants. **938**

Trifolium pratense

938766 Sverdrup, L. E., Hartnik, T., Mariussen, E., Jensen, J. (2006). Toxicity of three halogenated flame retardants to nitrifying bacteria, red clover (Trifolium pratense), and a soil invertebrate (Enchytraeus crypticus). Chemosphere 64(1):96-103. **942**

Triticum aestivum

5728923 Dogan, M., Korkunc, M., Yumrutas, O. (2012). Effects of bisphenol A and tetrabromobisphenol A on bread and durum wheat varieties. Ekoloji Dergisi 21(85):114-122. **944**

3416156 Li, Y., Zhou, Q., Li, F., Liu, X., Luo, Y.,i (2008). Effects of tetrabromobisphenol A as an emerging pollutant on wheat (Triticum aestivum) at biochemical levels. Chemosphere 74(1):119-124. **950**

Triticum durum

5728923 Dogan, M., Korkunc, M., Yumrutas, O. (2012). Effects of bisphenol A and tetrabromobisphenol A on bread and durum wheat varieties. Ekoloji Dergisi 21(85):114-122. **952**

Triticum sp. (Wheat reported by author)

3009733 Ge, H. L., Zhang, F. (2015). The effects of composite photosynthetic bacterial inoculant PS21 on the biochemical characteristics of wheat seedlings under tetrabromobisphenol A stress. Biotechnology and Biotechnological Equipment 29(2):289-298. **958**

Zea mays

6391028 Ltd,, W.I. (2002). Tetrabromobisphenol-A: A toxicity test to determine the effects of the test substance on seedling emergence of six species of plants. **960**

5123648 Wang, Y., Zhang, M., Zhao, L., Zhang, W., Zhao, T., Chu, J., Qiu, Y., Gong, H., Li, R. (2019). Effects of tetrabromobisphenol A on maize (Zea mays L.) physiological indexes, soil enzyme activity, and soil microbial biomass. Ecotoxicology 28(1):1-12. **964**

11520674 Xu, X., Han, W. (2023). Analysis of tetrabromobisphenol A and bisphenol A in plant sample—method optimization and identification of the derivatives. Environmental Science and Pollution Research 30(34):82770-82779. **970**

Study Citation:	Yang, S. W., Yan, Z. G., Xu, F. F., Wang, S. R., Wu, F. C. (2012). Development of freshwater aquatic life criteria for tetrabromobisphenol A in China. Environmental Pollution 169:59-63.
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; *Carassius auratus*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2562857

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%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using appropriate concurrent negative control groups
	Metric 5:	Negative Control Response	High	Mortality was less than 8 percent
	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	The study provided few details on exposure administration
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and 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 at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	Exposure concentrations above the solubility limit listed in the Final TBBPA Scope (4.15 mg/L) were included, however an ethanol solvent was used and a solvent control was indicated.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed

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Study Citation:	Yang, S. W., Yan, Z. G., Xu, F. F., Wang, S. R., Wu, F. C. (2012). Development of freshwater aquatic life criteria for tetrabromobisphenol A in China. Environmental Pollution 169:59-63.
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; *Carassius auratus*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2562857

Domain	Metric		Rating	Comments
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the outcome assessment were not reported.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	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
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: None

Overall Quality Determination Medium

Study Citation:	He, Q., Wang, X., Sun, P., Wang, Z., Wang, L. (2015). Acute and chronic toxicity of tetrabromobisphenol A to three aquatic species under different pH conditions. Aquatic Toxicology 164:145-154.
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; *Carassius auratus*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Hepatic/Liver
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3350555

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	High	The test substance identity was not analytically verified by the performing laboratory but source was clearly identified.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was shown in Figure 5A.
	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 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 but few details were provided
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	The number of exposure groups and spacing of exposure levels was lower than expected for a dose response
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms,
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of test organisms and replicates was lower than typical
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Few details provided so difficult to assess
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest

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Study Citation:	He, Q., Wang, X., Sun, P., Wang, Z., Wang, L. (2015). Acute and chronic toxicity of tetrabromobisphenol A to three aquatic species under different pH conditions. Aquatic Toxicology 164:145-154.
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; *Carassius auratus*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Hepatic/Liver
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3350555

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	High	the execution of the study protocol for outcome assessment was reported.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	pH was varied across experiments but was consistent across treatments
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups within an exposure
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group in Figure 5.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcome

Additional Comments: This evaluation is for the assessment of liver histopathology.

Overall Quality Determination High

Study Citation:	He, Q., Wang, X., Sun, P., Wang, Z., Wang, L. (2015). Acute and chronic toxicity of tetrabromobisphenol A to three aquatic species under different pH conditions. Aquatic Toxicology 164:145-154.
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; *Carassius auratus*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3350555

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	High	The test substance identity was not analytically verified by the performing laboratory but source was clearly identified.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	Medium	The biological response of the negative control groups was reported for oxidative stress
	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 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 but few details were provided
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	The number of exposure groups and spacing of exposure levels was lower than expected for a dose response
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms,
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of test organisms and replicates was lower than typical
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Few details provided so difficult to assess
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	the execution of the study protocol for outcome assessment was reported.

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Study Citation:	He, Q., Wang, X., Sun, P., Wang, Z., Wang, L. (2015). Acute and chronic toxicity of tetrabromobisphenol A to three aquatic species under different pH conditions. Aquatic Toxicology 164:145-154.
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; *Carassius auratus*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3350555

Domain	Metric		Rating	Comments
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	pH was varied across experiments but was consistent across treatments
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups within an exposure
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcome

Additional Comments: pH effects and mixture results with DMSO were reported as was histological assessment of some exposed organisms

Overall Quality Determination High

Study Citation:	He, Q., Wang, X., Sun, P., Wang, Z., Wang, L. (2015). Acute and chronic toxicity of tetrabromobisphenol A to three aquatic species under different pH conditions. Aquatic Toxicology 164:145-154.
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; *Carassius auratus*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Respiratory
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3350555

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	High	The test substance identity was not analytically verified by the performing laboratory but source was clearly identified.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was shown in Figure 4A.
	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 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 but few details were provided
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	The number of exposure groups and spacing of exposure levels was lower than expected for a dose response
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms,
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of test organisms and replicates was lower than typical
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Few details provided so difficult to assess
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	the execution of the study protocol for outcome assessment was reported.

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Study Citation:	He, Q., Wang, X., Sun, P., Wang, Z., Wang, L. (2015). Acute and chronic toxicity of tetrabromobisphenol A to three aquatic species under different pH conditions. Aquatic Toxicology 164:145-154.
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; *Carassius auratus*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Respiratory
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3350555

Domain	Metric		Rating	Comments
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	pH was varied across experiments but was consistent across treatments
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups within an exposure
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group in Figure 4.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcome

Additional Comments: This evaluation is for the assessment of gill histopathology.

Overall Quality Determination High

Study Citation:	Yang, S. W., Yan, Z. G., Xu, F. F., Wang, S. R., Wu, F. C. (2012). Development of freshwater aquatic life criteria for tetrabromobisphenol A in China. Environmental Pollution 169:59-63.
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; *Carassius auratus*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2562857

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%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using appropriate concurrent negative control groups
	Metric 5:	Negative Control Response	High	Mortality was less than 8 percent
	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	The study provided few details on exposure administration
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and 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 above the solubility limit listed in the Final TBBPA Scope (4.15 mg/L) were included, however an ethanol solvent was used and a solvent control was indicated.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	There are minor reservations about the source of test organisms
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	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	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed

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Study Citation:	Yang, S. W., Yan, Z. G., Xu, F. F., Wang, S. R., Wu, F. C. (2012). Development of freshwater aquatic life criteria for tetrabromobisphenol A in China. Environmental Pollution 169:59-63.
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; *Carassius auratus*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2562857

Domain	Metric		Rating	Comments
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the outcome assessment were not reported.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	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
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: None

Overall Quality Determination Medium

Study Citation:	Carlsson, G., Norrgren, L. (2014). Comparison of embryo toxicity using two classes of aquatic vertebrates. Environmental Toxicology and Pharmacology 37(1):24-7. [Environmental toxicology and pharmacology].
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*; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5518140

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity 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	Low	The biological response of the negative control groups was not reported.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Medium	Reporting omissions were unlikely to have a substantial impact on results
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure 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	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 replicates was not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Reported environmental conditions were not sufficient to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported

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Study Citation:	Carlsson, G., Norrgren, L. (2014). Comparison of embryo toxicity using two classes of aquatic vertebrates. Environmental Toxicology and Pharmacology 37(1):24-7. [Environmental toxicology and pharmacology].
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*; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5518140

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22:	Reporting of Data	Low	Data were only reported for some outcomes
	Metric 23:	Explanation of Unexpected Outcomes	Low	insufficient information was provided to determine if excessive variability or unexpected outcomes occurred.

Additional Comments: None

Overall Quality Determination Medium

Study Citation:	Carlsson, G., Norrgren, L. (2014). Comparison of embryo toxicity using two classes of aquatic vertebrates. Environmental Toxicology and Pharmacology 37(1):24-7. [Environmental toxicology and pharmacology].
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*; Embryo
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5518140

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity 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	Low	The biological response of the negative control groups was not reported.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Medium	Reporting omissions were unlikely to have a substantial impact on results
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure 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	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 replicates was not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Reported environmental conditions were not sufficient to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported.

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Study Citation:	Carlsson, G., Norrgren, L. (2014). Comparison of embryo toxicity using two classes of aquatic vertebrates. Environmental Toxicology and Pharmacology 37(1):24-7. [Environmental toxicology and pharmacology].
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*; Embryo
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5518140

Domain	Metric		Rating	Comments
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22:	Reporting of Data	Low	Data were only reported for some outcomes
	Metric 23:	Explanation of Unexpected Outcomes	Low	insufficient information was provided to determine if excessive variability or unexpected outcomes occurred.

Additional Comments: None

Overall Quality Determination Medium

Study Citation:	Carlsson, G., Norrgren, L. (2014). Comparison of embryo toxicity using two classes of aquatic vertebrates. Environmental Toxicology and Pharmacology 37(1):24-7. [Environmental toxicology and pharmacology].
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*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5518140

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity 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	Low	The biological response of the negative control groups was not reported.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Medium	Reporting omissions were unlikely to have a substantial impact on results
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure 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	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 replicates was not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Reported environmental conditions were not sufficient to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported.

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Study Citation:	Carlsson, G., Norrgren, L. (2014). Comparison of embryo toxicity using two classes of aquatic vertebrates. Environmental Toxicology and Pharmacology 37(1):24-7. [Environmental toxicology and pharmacology].
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*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5518140

Domain	Metric		Rating	Comments
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22:	Reporting of Data	Low	Data were only reported for some outcomes
	Metric 23:	Explanation of Unexpected Outcomes	Low	insufficient information was provided to determine if excessive variability or unexpected outcomes occurred.

Additional Comments: None

Overall Quality Determination Medium

Study Citation:	Chan, W. K., Chan, K. M. (2012). Disruption of the hypothalamic-pituitary-thyroid axis in zebrafish embryo-larvae following waterborne exposure to BDE-47, TBBPA and BPA. Aquatic Toxicology 108:106-111.
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*; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1401639

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	analytical-grade chemical was used
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 reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of 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:	Chan, W. K., Chan, K. M. (2012). Disruption of the hypothalamic-pituitary-thyroid axis in zebrafish embryo-larvae following waterborne exposure to BDE-47, TBBPA and BPA. Aquatic Toxicology 108:106-111.
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*; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1401639

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
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: None

Overall Quality Determination High

Study Citation:	Chan, W. K., Chan, K. M. (2012). Disruption of the hypothalamic-pituitary-thyroid axis in zebrafish embryo-larvae following waterborne exposure to BDE-47, TBBPA and BPA. Aquatic Toxicology 108:106-111.
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*; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1401639

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	analytical-grade chemical was used
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 reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of 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:	Chan, W. K., Chan, K. M. (2012). Disruption of the hypothalamic-pituitary-thyroid axis in zebrafish embryo-larvae following waterborne exposure to BDE-47, TBBPA and BPA. Aquatic Toxicology 108:106-111.
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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; *Danio rerio*; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1401639

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
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: expression

Overall Quality Determination High

Study Citation:	Chan, W. K., Chan, K. M. (2012). Disruption of the hypothalamic-pituitary-thyroid axis in zebrafish embryo-larvae following waterborne exposure to BDE-47, TBBPA and BPA. Aquatic Toxicology 108:106-111.
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*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1401639

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	analytical-grade chemical was used
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 reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of 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:	Chan, W. K., Chan, K. M. (2012). Disruption of the hypothalamic-pituitary-thyroid axis in zebrafish embryo-larvae following waterborne exposure to BDE-47, TBBPA and BPA. Aquatic Toxicology 108:106-111.
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*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1401639

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
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: hatch

Overall Quality Determination High

Study Citation:	Chen, J., Tanguay, R. L., Xiao, Y., Haggard, D. E., Ge, X., Jia, Y., Zheng, Y., Dong, Q., Huang, C., Lin, K. (2016). TBBPA exposure during a sensitive developmental window produces neurobehavioral changes in larval zebrafish. Environmental Pollution 216:53-63.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wild type; Embryo
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4933778

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >97%
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 reported and reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The experimental system and methods for preparation of test media were described with few details
	Metric 8:	Consistency of Exposure Administration	Medium	Exposures were administered consistently across study groups but few details were provided
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured, fish were measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	Only one concentration tested
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms,
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Reported environmental conditions were not sufficient to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest

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Study Citation:	Chen, J., Tanguay, R. L., Xiao, Y., Haggard, D. E., Ge, X., Jia, Y., Zheng, Y., Dong, Q., Huang, C., Lin, K. (2016). TBBPA exposure during a sensitive developmental window produces neurobehavioral changes in larval zebrafish. Environmental Pollution 216:53-63.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wild type; Embryo
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4933778

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	Medium	outcomes were assessed consistently across study groups, few details were provided
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Medium	There were no reported differences among the study groups in environmental conditions, few details were provided
	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	Medium	There were no unexpected outcomes, few details were provided

Additional Comments: None

Overall Quality Determination High

Study Citation:	Chen, J., Tanguay, R. L., Xiao, Y., Haggard, D. E., Ge, X., Jia, Y., Zheng, Y., Dong, Q., Huang, C., Lin, K. (2016). TBBPA exposure during a sensitive developmental window produces neurobehavioral changes in larval zebrafish. Environmental Pollution 216:53-63.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wild type; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4933778

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >97%
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 reported and reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The experimental system and methods for preparation of test media were described with few details
	Metric 8:	Consistency of Exposure Administration	Medium	Exposures were administered consistently across study groups but few details were provided
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	There were deficiencies regarding the number of exposure groups. The range of concentrations produced no response at the low level, and a large response at the high level. Intermediate concentrations would have been helpful to determine a more robust dose-response relationship.
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms,
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Reported environmental conditions were not sufficient to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest

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Study Citation:	Chen, J., Tanguay, R. L., Xiao, Y., Haggard, D. E., Ge, X., Jia, Y., Zheng, Y., Dong, Q., Huang, C., Lin, K. (2016). TBBPA exposure during a sensitive developmental window produces neurobehavioral changes in larval zebrafish. Environmental Pollution 216:53-63.
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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; *Danio rerio*; AB wild type; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4933778

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	Medium	outcomes were assessed consistently across study groups, few details were provided
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Medium	There were no reported differences among the study groups in environmental conditions, few details were provided
	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	Medium	There were no unexpected outcomes, few details were provided

Additional Comments: None

Overall Quality Determination High

Study Citation:	Chen, J., Tanguay, R. L., Xiao, Y., Haggard, D. E., Ge, X., Jia, Y., Zheng, Y., Dong, Q., Huang, C., Lin, K. (2016). TBBPA exposure during a sensitive developmental window produces neurobehavioral changes in larval zebrafish. Environmental Pollution 216:53-63.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wild type; Embryo
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4933778

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >97%
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 reported and reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The experimental system and methods for preparation of test media were described with few details
	Metric 8:	Consistency of Exposure Administration	Medium	Exposures were administered consistently across study groups but few details were provided
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured, fish were measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	Only one concentration tested
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms,
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Reported environmental conditions were not sufficient to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	Medium	outcomes were assessed consistently across study groups, few details were provided

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Study Citation:	Chen, J., Tanguay, R. L., Xiao, Y., Haggard, D. E., Ge, X., Jia, Y., Zheng, Y., Dong, Q., Huang, C., Lin, K. (2016). TBBPA exposure during a sensitive developmental window produces neurobehavioral changes in larval zebrafish. Environmental Pollution 216:53-63.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wild type; Embryo
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4933778

Domain	Metric		Rating	Comments
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Medium	There were no reported differences among the study groups in environmental conditions, few details were provided
	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	Medium	There were no unexpected outcomes, few details were provided

Additional Comments: activity, speed

Overall Quality Determination — High

Study Citation:	Chen, J., Tanguay, R. L., Xiao, Y., Haggard, D. E., Ge, X., Jia, Y., Zheng, Y., Dong, Q., Huang, C., Lin, K. (2016). TBBPA exposure during a sensitive developmental window produces neurobehavioral changes in larval zebrafish. Environmental Pollution 216:53-63.
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 wild type; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Cytotoxicity
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4933778

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >97%
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 reported and reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The experimental system and methods for preparation of test media were described with few details
	Metric 8:	Consistency of Exposure Administration	Medium	Exposures were administered consistently across study groups but few details were provided
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured, fish were measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	Only one concentration tested
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms,
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Reported environmental conditions were not sufficient to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	Medium	outcomes were assessed consistently across study groups, few details were provided

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Study Citation:	Chen, J., Tanguay, R. L., Xiao, Y., Haggard, D. E., Ge, X., Jia, Y., Zheng, Y., Dong, Q., Huang, C., Lin, K. (2016). TBBPA exposure during a sensitive developmental window produces neurobehavioral changes in larval zebrafish. Environmental Pollution 216:53-63.
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 wild type; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Cytotoxicity
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4933778

Domain	Metric		Rating	Comments
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Medium	There were no reported differences among the study groups in environmental conditions, few details were provided
	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	Medium	There were no unexpected outcomes, few details were provided

Additional Comments: mRNA expression

Overall Quality Determination — High

Study Citation:	Chen, J., Tanguay, R. L., Xiao, Y., Haggard, D. E., Ge, X., Jia, Y., Zheng, Y., Dong, Q., Huang, C., Lin, K. (2016). TBBPA exposure during a sensitive developmental window produces neurobehavioral changes in larval zebrafish. Environmental Pollution 216:53-63.
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 wild type; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Cytotoxicity
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4933778

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >97%
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 reported and reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The experimental system and methods for preparation of test media were described with few details
	Metric 8:	Consistency of Exposure Administration	Medium	Exposures were administered consistently across study groups but few details were provided
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured, fish were measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	Only one concentration tested
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms,
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Reported environmental conditions were not sufficient to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	Medium	outcomes were assessed consistently across study groups, few details were provided

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Study Citation:	Chen, J., Tanguay, R. L., Xiao, Y., Haggard, D. E., Ge, X., Jia, Y., Zheng, Y., Dong, Q., Huang, C., Lin, K. (2016). TBBPA exposure during a sensitive developmental window produces neurobehavioral changes in larval zebrafish. Environmental Pollution 216:53-63.
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 wild type; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Cytotoxicity
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4933778

Domain	Metric		Rating	Comments
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Medium	There were no reported differences among the study groups in environmental conditions, few details were provided
	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	Medium	There were no unexpected outcomes, few details were provided

Additional Comments: apoptosis

Overall Quality Determination High

Study Citation:	Chen, J., Tanguay, R. L., Xiao, Y., Haggard, D. E., Ge, X., Jia, Y., Zheng, Y., Dong, Q., Huang, C., Lin, K. (2016). TBBPA exposure during a sensitive developmental window produces neurobehavioral changes in larval zebrafish. Environmental Pollution 216:53-63.
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*; NBT; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4933778

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >97%
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 reported and reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The experimental system and methods for preparation of test media were described with few details
	Metric 8:	Consistency of Exposure Administration	Medium	Exposures were administered consistently across study groups but few details were provided
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured, fish were measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	Only one concentration tested
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms,
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Reported environmental conditions were not sufficient to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	Medium	outcomes were assessed consistently across study groups, few details were provided

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Study Citation:	Chen, J., Tanguay, R. L., Xiao, Y., Haggard, D. E., Ge, X., Jia, Y., Zheng, Y., Dong, Q., Huang, C., Lin, K. (2016). TBBPA exposure during a sensitive developmental window produces neurobehavioral changes in larval zebrafish. Environmental Pollution 216:53-63.
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*; NBT; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4933778

Domain	Metric		Rating	Comments
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Medium	There were no reported differences among the study groups in environmental conditions, few details were provided
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical methods were adequately described for ventral axon length not for neuron patterns
	Metric 22:	Reporting of Data	Medium	Data for exposure-related findings for ventral axon length were presented for each treatment and control group not for neuron patterns
	Metric 23:	Explanation of Unexpected Outcomes	Medium	There were no unexpected outcomes, few details were provided

Additional Comments: neuron, fiber development

Overall Quality Determination Medium

Study Citation:	Chen, J., Tanguay, R. L., Xiao, Y., Haggard, D. E., Ge, X., Jia, Y., Zheng, Y., Dong, Q., Huang, C., Lin, K. (2016). TBBPA exposure during a sensitive developmental window produces neurobehavioral changes in larval zebrafish. Environmental Pollution 216:53-63.
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*; Islet; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4933778

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >97%
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 reported and reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The experimental system and methods for preparation of test media were described with few details
	Metric 8:	Consistency of Exposure Administration	Medium	Exposures were administered consistently across study groups but few details were provided
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured, fish were measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	Only one concentration tested
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms,
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Reported environmental conditions were not sufficient to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	Medium	outcomes were assessed consistently across study groups, few details were provided

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Study Citation:	Chen, J., Tanguay, R. L., Xiao, Y., Haggard, D. E., Ge, X., Jia, Y., Zheng, Y., Dong, Q., Huang, C., Lin, K. (2016). TBBPA exposure during a sensitive developmental window produces neurobehavioral changes in larval zebrafish. Environmental Pollution 216:53-63.
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*; Islet; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4933778

Domain	Metric		Rating	Comments
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Medium	There were no reported differences among the study groups in environmental conditions, few details were provided
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Uninformative	Statistical methods were not used for ventral axon length or neuron patterns
	Metric 22:	Reporting of Data	Low	Data for exposure-related findings were not reported for ventral axon length or neuron patterns
	Metric 23:	Explanation of Unexpected Outcomes	Medium	There were no unexpected outcomes, few details were provided

Additional Comments: neuron, fiber development

Overall Quality Determination Uninformative

Study Citation:	Chen, J., Tanguay, R. L., Xiao, Y., Haggard, D. E., Ge, X., Jia, Y., Zheng, Y., Dong, Q., Huang, C., Lin, K. (2016). TBBPA exposure during a sensitive developmental window produces neurobehavioral changes in larval zebrafish. Environmental Pollution 216:53-63.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wild type; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4933778

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >97%
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 reported and reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The experimental system and methods for preparation of test media were described with few details
	Metric 8:	Consistency of Exposure Administration	Medium	Exposures were administered consistently across study groups but few details were provided
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	There were deficiencies regarding the number of exposure groups
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms,
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Reported environmental conditions were not sufficient to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	Medium	outcomes were assessed consistently across study groups, few details were provided

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Study Citation:	Chen, J., Tanguay, R. L., Xiao, Y., Haggard, D. E., Ge, X., Jia, Y., Zheng, Y., Dong, Q., Huang, C., Lin, K. (2016). TBBPA exposure during a sensitive developmental window produces neurobehavioral changes in larval zebrafish. Environmental Pollution 216:53-63.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wild type; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4933778

Domain	Metric		Rating	Comments
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Medium	There were no reported differences among the study groups in environmental conditions, few details were provided
	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	Medium	There were no unexpected outcomes, few details were provided

Additional Comments: malformations

Overall Quality Determination High

Study Citation:	Chow, W. S., Chan, W., Chan, K. M. (2013). Toxicity assessment and vitellogenin expression in zebrafish (Danio rerio) embryos and larvae acutely exposed to bisphenol A, endosulfan, heptachlor, methoxychlor and tetrabromobisphenol A. Journal of Applied Toxicology 33(7):670-678.
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*; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5804500

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	CASN and name reported, no structure listed.
	Metric 2:	Test Substance Source	Low	Sigma Aldrich was reported as the source. The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Authors reported that the purity was Analytical grade and highest available (98.7% as reported on Sigma website).
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Study authors reported using an appropriate concurrent negative control group. Solvent controls with DMSO were used. However, it was reported that TBBPA was dissolved in methanol.
	Metric 5:	Negative Control Response	Medium	The control responses are not in the narrative but can be viewed within the figures for dose-reposes on figure 1e and 2e for embryos and larvae, respectively.
	Metric 6:	Randomized Allocation	Low	Allocation not reported as random.
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. Test media methods did not detail the final concentration of methanol for the preparation of initial stock solutions. The solvent control was reported as using DMSO, so some inconsistencies in reporting. Bioassays conducted as static non-renewal for 96 h duration, which might no account for loss from evaporation throughout time. Concentrations of test substance were not measured during the study.
	Metric 8:	Consistency of Exposure Administration	High	Exposure administration appears to be consistent among treatments.
	Metric 9:	Measurement of Test Substance Concentration	Low	Concentrations reported as nominal.
	Metric 10:	Exposure Duration and Frequency	High	96h duration is appropriate for this acute bioassay.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The concentrations are spaced well but not directly reported in the narrative, they can be viewed in Figure 1e.
	Metric 12:	Testing at or Below Solubility Limit	High	The exposure concentrations (from Figure 1e) were at or under solubility reported in the Final Scope for TBBPA (4.15 mg/L).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Authors reported using embryos that were within 2 hours post fertilization. Source of embryos were from in house broodfish.

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Study Citation:	Chow, W. S., Chan, W., Chan, K. M. (2013). Toxicity assessment and vitellogenin expression in zebrafish (Danio rerio) embryos and larvae acutely exposed to bisphenol A, endosulfan, heptachlor, methoxychlor and tetrabromobisphenol A. Journal of Applied Toxicology 33(7):670-678.
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*; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5804500

Domain	Metric		Rating	Comments
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Conditions of broodfish were adequately reported on page 2/9. All pretreatment conditions were the same for control and exposed groups.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Triplicate groups of embryos and larvae with 10 individuals per replicate.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	The conditions for acute toxicity bioassays for both embryos and larvae were appropriate. These were static assays for 96 hours and authors did not report water quality conditions (pH, Ammonia) at conclusion of test.
	Metric 17:	Outcome Assessment Methodology	Medium	Assessment methods were adequate, authors did not list detailed criteria for dead larvae or embryos.
	Metric 18:	Consistency of Outcome Assessment	High	Outcome assessment protocol was conducted consistently among treatments.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Medium	No differences reported to indicate environmental conditions or other factors to influence bioassay, but WQ parameters were not monitored or reported.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	Nothing presented to indicate differences from animal attrition or health.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	The authors reported using the USEPA probit program (MS DOS) for LC50 and CI estimation.
	Metric 22:	Reporting of Data	High	The authors present both a table and figures with dose-response information.
	Metric 23:	Explanation of Unexpected Outcomes	High	LC50s are presented with 95% confidence intervals. No unexpected results were encountered.

Additional Comments: The 96 hr EC50 values of TBBPA on hatching of zebrafish embryos was 1.091 mg/L with 95% confidence limits of 0.884 -1.293 mg/l.

Overall Quality Determination Medium

Study Citation:	Chow, W. S., Chan, W., Chan, K. M. (2013). Toxicity assessment and vitellogenin expression in zebrafish (Danio rerio) embryos and larvae acutely exposed to bisphenol A, endosulfan, heptachlor, methoxychlor and tetrabromobisphenol A. Journal of Applied Toxicology 33(7):670-678.
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*; Larvae
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5804500

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	CASRN and name reported.
	Metric 2:	Test Substance Source	Low	Sigma Aldrich was reported as the source. The test substance identity was NOT analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Authors reported that the purity was Analytical grade and highest available (98.7% as reported on Sigma website).
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Solvent controls with DMSO were used. However, stock solutions of TBBPA was made using methanol.
	Metric 5:	Negative Control Response	Medium	Control responses for vtg gene transcription are reported in figure 3E with control represented at 0% concentration of TBBPA and a fold induction of ~1 mean and SD.
	Metric 6:	Randomized Allocation	Low	Allocation not reported as random.
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. Test media methods did not detail the final concentration of methanol for the preparation of initial stock solutions. The solvent control was reported as using DMSO, so some inconsistencies in reporting. Bioassays conducted as static non-renewal for 96 h duration, which might no account for loss from evaporation throughout time. Concentrations of test substance were not measured during the study.
	Metric 8:	Consistency of Exposure Administration	High	Exposure administration appears to be consistent among treatments.
	Metric 9:	Measurement of Test Substance Concentration	Low	Concentrations reported as nominal.
	Metric 10:	Exposure Duration and Frequency	High	96h duration was performed for gene expression.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The concentrations for the gene transcription experiment were 0, 10, 25, 50 and 75% of the LC50 value for either embryos or larvae.
	Metric 12:	Testing at or Below Solubility Limit	High	For larvae, exposures were carried out at 0, 10, 25, 50, 75% proportions of the LC50 which was 5.27 mg/L. As a result, the highest treatment concentration for embryo vtg gene expression would be 3.95 mg/L, which is under the solubility reported in the final scope at 4.15 mg/L.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Larvae were 96 hpf and the source of embryos were from in house broodfish.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Conditions of broodfish were adequately reported on page 2/9. All pretreatment conditions were the same for control and exposed groups,

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Study Citation:	Chow, W. S., Chan, W., Chan, K. M. (2013). Toxicity assessment and vitellogenin expression in zebrafish (Danio rerio) embryos and larvae acutely exposed to bisphenol A, endosulfan, heptachlor, methoxychlor and tetrabromobisphenol A. Journal of Applied Toxicology 33(7):670-678.
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*; Larvae
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5804500

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Low	Triplicate groups of embryos and larvae with 10 individuals per replicate. The transcription data have pooled the 10 larvae into three samples, which looses some resolution for gene transcription variance.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	The conditions for gene transcription for both embryos and larvae were appropriate. These were static assays for 96 hours and authors did not report water quality conditions (pH, Ammonia) at conclusion of test.
	Metric 17:	Outcome Assessment Methodology	Medium	Assessment methods were adequate.
	Metric 18:	Consistency of Outcome Assessment	High	Outcome assessment protocol was conducted consistently among treatments.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Medium	No differences reported to indicate environmental conditions or other factors to influence bioassay, but WQ parameters were not monitored or reported.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	Nothing presented to indicate differences from animal attrition or health.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Gene transcription among control and treatment concentrations was assessed using ANOVA with Tukey's post hoc.
	Metric 22:	Reporting of Data	High	Figure 3E shows relative gene transcription of VTG in both embryo and larvae. Authors also calculate the NOEL and LOEL for gene expression.
	Metric 23:	Explanation of Unexpected Outcomes	High	Variance is represented in the gene transcription data with SD for each treatment and control. No unexpected results were encountered.

Additional Comments: The NOEL and LOEL values of TBBPA for vtg1 mRNA induction in larvae was determined as 2.64 and 3.95 mg/l respectively.

Overall Quality Determination Medium

Study Citation:	Chow, W. S., Chan, W., Chan, K. M. (2013). Toxicity assessment and vitellogenin expression in zebrafish (Danio rerio) embryos and larvae acutely exposed to bisphenol A, endosulfan, heptachlor, methoxychlor and tetrabromobisphenol A. Journal of Applied Toxicology 33(7):670-678.
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*; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5804500

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	CASRN and name reported.
	Metric 2:	Test Substance Source	Low	Sigma Aldrich was reported as the source .The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Authors reported that the purity was Analytical grade and highest available (98.7% as reported on Sigma website).
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Solvent controls with DMSO were used. But it was reported that stock solutions of TBBPA was made using methanol.
	Metric 5:	Negative Control Response	Medium	Control responses for vtg gene transcription are reported in figure 3E with control represented at 0% concentration of TBBPA and a fold induction of ~1 mean and SD.
	Metric 6:	Randomized Allocation	Low	Allocation not reported as random.
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. Test media methods did not detail the final concentration of methanol for the preparation of initial stock solutions. The solvent control was reported as using DMSO, so some inconsistencies in reporting. Bioassays conducted as static non-renewal for 96 h duration, which might not account for loss from evaporation throughout time. Concentrations of test substance were not measured during the study.
	Metric 8:	Consistency of Exposure Administration	High	Exposure administration appears to be consistent among treatments.
	Metric 9:	Measurement of Test Substance Concentration	Low	Concentrations reported as nominal.
	Metric 10:	Exposure Duration and Frequency	High	96h duration was performed for gene expression.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The concentrations for the gene transcription experiment were 0, 10, 25, 50 and 75% of the LC50 value for either embryos or larvae.
	Metric 12:	Testing at or Below Solubility Limit	High	For embryo, exposures were carried out at 0, 10, 25, 50, 75% proportions of the LC50 which was 1.09 mg/L. As a result, all treatment concentrations are under the solubility reported in the final scope at 4.15 mg/L.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Embryos were within 2 hours of fertilization and the source of embryos were from in house broodfish.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Conditions of broodfish were adequately reported on page 2/9. All pretreatment conditions were the same for control and exposed groups.

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Study Citation:	Chow, W. S., Chan, W., Chan, K. M. (2013). Toxicity assessment and vitellogenin expression in zebrafish (Danio rerio) embryos and larvae acutely exposed to bisphenol A, endosulfan, heptachlor, methoxychlor and tetrabromobisphenol A. Journal of Applied Toxicology 33(7):670-678.
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*; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5804500

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Low	Triplicate groups of embryos and larvae with 10 individuals per replicate. The transcription data have pooled the 10 larvae into three samples, which looses some resolution for gene transcription variance.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	The conditions for gene transcription for both embryos and larvae were appropriate. These were static assays for 96 hours and authors did not report water quality conditions (pH, Ammonia) at conclusion of test.
	Metric 17:	Outcome Assessment Methodology	Medium	Assessment methods were adequate.
	Metric 18:	Consistency of Outcome Assessment	High	Outcome assessment protocol was conducted consistently among treatments.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Medium	No differences reported to indicate environmental conditions or other factors to influence bioassay, but WQ parameters were not monitored or reported.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	Nothing presented to indicate differences from animal attrition or health.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Gene transcription among control and treatment concentrations was assessed using ANOVA with Tukey's post hoc.
	Metric 22:	Reporting of Data	High	Figure 3E shows relative gene transcription of VTG in both embryo and larvae. Authors also calculate the NOEL and LOEL for gene expression.
	Metric 23:	Explanation of Unexpected Outcomes	High	Variance is represented in the gene transcription data with SD for each treatment and control. No unexpected results were encountered.

Additional Comments: The NOEL and LOEL values of TBBPA for vtg1 mRNA induction in embryos was determined as 0.27 and 0.55 mg/l respectively.

Overall Quality Determination Medium

Study Citation:	Chow, W. S., Chan, W., Chan, K. M. (2013). Toxicity assessment and vitellogenin expression in zebrafish (Danio rerio) embryos and larvae acutely exposed to bisphenol A, endosulfan, heptachlor, methoxychlor and tetrabromobisphenol A. Journal of Applied Toxicology 33(7):670-678.
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*; Larvae
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5804500

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	CASRN and chemical name reported.
	Metric 2:	Test Substance Source	Low	Sigma Aldrich was reported as the source. The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Authors reported that the purity was Analytical grade and highest available (98.7% as reported on Sigma website).
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Study authors reported using an appropriate concurrent negative control group. Solvent controls with DMSO (1%) were used. However, stock solution of TBBPA was made using methanol.
	Metric 5:	Negative Control Response	Medium	The control responses are not in the narrative but can be viewed within the figures for dose-reposes on figure 1e and 2e for embryos and larvae, respectively.
	Metric 6:	Randomized Allocation	Low	Allocation not reported as random.
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. Test media methods did not detail the final concentration of methanol for the preparation of initial stock solutions. The solvent control was reported as using DMSO, so some inconsistencies in reporting. Bioassays conducted as static non-renewal for 96 h duration, which might no account for loss from evaporation throughout time. Concentrations of test substance were not measured during the study.
	Metric 8:	Consistency of Exposure Administration	High	Exposure administration appears to be consistent among treatments.
	Metric 9:	Measurement of Test Substance Concentration	Low	Concentrations reported as nominal.
	Metric 10:	Exposure Duration and Frequency	High	96h duration is appropriate for this acute bioassay.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The concentrations are spaced well but not directly reported in the narrative, they can be viewed in Figure 2e.
	Metric 12:	Testing at or Below Solubility Limit	Low	The exposure concentrations (from Figure 2e) at or over the solubility reported in the Final Scope for TBBPA (4.15 mg/L).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Source of embryos were from in house broodfish. Larvae used for this acute bioassay were 96 hpf.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Conditions of broodfish were adequately reported on page 2/9. All pretreatment conditions were the same for control and exposed groups.

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Study Citation:	Chow, W. S., Chan, W., Chan, K. M. (2013). Toxicity assessment and vitellogenin expression in zebrafish (Danio rerio) embryos and larvae acutely exposed to bisphenol A, endosulfan, heptachlor, methoxychlor and tetrabromobisphenol A. Journal of Applied Toxicology 33(7):670-678.
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*; Larvae
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5804500

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Triplicate groups of embryos and larvae with 10 individuals per replicate.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	The conditions for acute toxicity bioassays for both embryos and larvae were appropriate. These were static assays for 96 hours and authors did not report water quality conditions (pH, Ammonia) at conclusion of test.
	Metric 17:	Outcome Assessment Methodology	Medium	Assessment methods were adequate, authors did not list detailed criteria for dead larvae or embryos.
	Metric 18:	Consistency of Outcome Assessment	High	Outcome assessment protocol was conducted consistently among treatments.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Medium	No differences reported to indicate environmental conditions or other factors to influence bioassay, but WQ parameters were not monitored or reported.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	Nothing presented to indicate differences from animal attrition or health.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	The authors reported using the USEPA probit program (MS DOS) for LC50 and CI estimation.
	Metric 22:	Reporting of Data	High	The authors present both a table and figures with dose-response information Page 5/9.
	Metric 23:	Explanation of Unexpected Outcomes	High	LC50s are presented with 95% confidence intervals. No unexpected results were encountered.

Additional Comments: The 96 hr EC50 values of TBBPA on zebrafish larvae was 5.270 mg/L with 95% confidence limits of 4.931- 5.581 mg/l.

Overall Quality Determination Medium

Study Citation:	Godfrey, A., Abdel-Moneim, A., Sepúlveda, M. S. (2017). Acute mixture toxicity of halogenated chemicals and their next generation counterparts on zebrafish embryos. Chemosphere 181:710-712.
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 Wild type; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3858252

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#.
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent solvent control group.
	Metric 5:	Negative Control Response	High	The biological response of the 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 test media preparation methods were reported. 0.1 M NaOH was used as the solvent at a concentration <0.003%. Test solution was renewed daily. The cited reference was reviewed (Gao et al., 2015), which provided additional details.
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently on a daily basis.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	The range of concentrations tested (0.625 - 5 ppm) was adequate to allow identification of endpoint value but the number of exposure groups and spacing were not reported.
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate ((i.e., no effects on biological responses were observed in the solvent control and no interactions were expected between the solvent and test substance).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source. Embryos at gastrula stage (4.5 hours post fertilization) were obtained from adult zebrafish cultures maintained at Purdue University Aquatic Laboratory.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.

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Study Citation:	Godfrey, A., Abdel-Moneim, A., Sepúlveda, M. S. (2017). Acute mixture toxicity of halogenated chemicals and their next generation counterparts on zebrafish embryos. Chemosphere 181:710-712.
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 Wild type; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3858252

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms (20 embryos per concentration) and replicates were reported and sufficient to characterize toxicological effects. Only two replicates were used per concentration but exposures were repeated three times.
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	Medium	The outcome assessment methodology was reported in the cited reference for methods (Gao et al., 2015) but criteria used to asses dead embryos 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 in animal health outcomes.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods (including any calculations or data transformations) were clearly described.
	Metric 22:	Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group. Only LC 50 values were reported.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: LC50 value and associated 95% confidence intervals (CI) from zebrafish embryos exposed to TBBPA (0.625 -5 ppm) was 1.3 ppm (95%CI: 1.1-1.6mg/L). The number of exposure groups, spacing of exposure levels used and mortality data for each treatment group were not reported.

Overall Quality Determination High

Study Citation:	Hu, J., Liang, Y., Chen, M., Wang, X. (2009). Assessing the toxicity of TBBPA and HBCD by zebrafish embryo toxicity assay and biomarker analysis. Environmental Toxicology 24(4):334-342.
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; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1927732

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	All other chemicals were analytical grade but grade of test substance was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	Medium	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	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 their source 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 effect
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reported environmental conditions were not sufficient to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest

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Study Citation:	Hu, J., Liang, Y., Chen, M., Wang, X. (2009). Assessing the toxicity of TBBPA and HBCD by zebrafish embryo toxicity assay and biomarker analysis. Environmental Toxicology 24(4):334-342.
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; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1927732

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: None

Overall Quality Determination High

Study Citation:	Hu, J., Liang, Y., Chen, M., Wang, X. (2009). Assessing the toxicity of TBBPA and HBCD by zebrafish embryo toxicity assay and biomarker analysis. Environmental Toxicology 24(4):334-342.
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; Embryo
Health Outcome:	Mechanistic-Cell signaling/function-Oxidative stress (including redox biology)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1927732

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	All other chemicals were analytical grade but grade of test substance was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	Medium	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	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 their source 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 effect
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reported environmental conditions were not sufficient to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control				

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Study Citation:	Hu, J., Liang, Y., Chen, M., Wang, X. (2009). Assessing the toxicity of TBBPA and HBCD by zebrafish embryo toxicity assay and biomarker analysis. Environmental Toxicology 24(4):334-342.
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; Embryo
Health Outcome:	Mechanistic-Cell signaling/function-Oxidative stress (including redox biology)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1927732

Domain		Metric	Rating	Comments
	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: None

Overall Quality Determination High

Study Citation:	Hu, J., Liang, Y., Chen, M., Wang, X. (2009). Assessing the toxicity of TBBPA and HBCD by zebrafish embryo toxicity assay and biomarker analysis. Environmental Toxicology 24(4):334-342.
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; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1927732

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	All other chemicals were analytical grade but grade of test substance was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	Medium	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	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 their source 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 effect
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reported environmental conditions were not sufficient to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control				

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Study Citation:	Hu, J., Liang, Y., Chen, M., Wang, X. (2009). Assessing the toxicity of TBBPA and HBCD by zebrafish embryo toxicity assay and biomarker analysis. Environmental Toxicology 24(4):334-342.
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; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1927732

Domain	Metric		Rating	Comments
	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 for hatch, malformations were not statistically analyzed
	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: hatch and malformations

Overall Quality Determination High

Study Citation:	Jarema, K. A., Hunter, D. L., Shaffer, R. M., Behl, M., Padilla, S. (2015). Acute and developmental behavioral effects of flame retardants and related chemicals in zebrafish. Neurotoxicology and Teratology 52(Pt B):194-209.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; Embryo
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4180931

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	CASRN, structure, molecular weight, nomenclature all were reported.
	Metric 2:	Test Substance Source	Low	Lot number and source was reported but not analytically verified by the lab.
	Metric 3:	Test Substance Purity	High	99% purity reported from the manufacturer.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Controls with carrier (DMSO) were used. Positive controls were also employed.
	Metric 5:	Negative Control Response	High	Neurotypical control movement was well documented from control larvae (Figure 2).
	Metric 6:	Randomized Allocation	Medium	Random allocation of treatment concentrations and control groups was described by plate locations.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The preparation of test solutions is well described and appropriate.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently among control and treatment concentrations
	Metric 9:	Measurement of Test Substance Concentration	Medium	Exposure concentrations were not measured but renewal and protection from evaporation were conducted.
	Metric 10:	Exposure Duration and Frequency	High	This study utilized exposures to the test compound during developmental periods from fertilization to 48 hrs of embryo development.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	5 concentrations were employed and previous range finding was conducted.
	Metric 12:	Testing at or Below Solubility Limit	Medium	The highest test concentration 12 uMol is over the solubility limit as reported in the final scope at 4.1 mg/L.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Source of test organism were listed and reliable.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Test and pretreatment conditions were not different and were well detailed.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The number of embryos per treatment concentration are reported on page 22 for the exposure assay.
Domain 5: Outcome Assessment				

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Study Citation:	Jarema, K. A., Hunter, D. L., Shaffer, R. M., Behl, M., Padilla, S. (2015). Acute and developmental behavioral effects of flame retardants and related chemicals in zebrafish. Neurotoxicology and Teratology 52(Pt B):194-209.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; Embryo
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4180931

Domain	Metric		Rating	Comments
	Metric 16:	Adequacy of Test Conditions	High	Although no standard method listed, the culture conditions during the test are appropriate for zebrafish embryos.
	Metric 17:	Outcome Assessment Methodology	High	Outcome assessment was identified and appropriate.
	Metric 18:	Consistency of Outcome Assessment	High	Assessments were conducted consistency among treatments and control.
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	Medium	No information was reported to suggest differences among treatment groups in animal attrition.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	"The data were first assessed using a repeated-measures analysis of variance (ANOVA) with time and dose as the independent variables and locomotor activity (distance moved/time) as the dependent variable. All of the results of this repeated-measures ANOVA for both the developmental and acute (time of peak effect) assessments are presented in Supplemental Table 2."
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented.
	Metric 23:	Explanation of Unexpected Outcomes	High	Unexplained or unexpected outcomes were not encountered in the current study.

Additional Comments: A high-quality study that exposed zebrafish embryos to TBBPA at 5 concentrations and a control during development (0-48 hours post fertilization). Behavioral metrics based on movement (distance moved) are presented at 6 days post fertilization. Data are presented on Figure 3, page 22. TBBPA exposure during embryogenesis did not cause movement metrics to differ from control.

Overall Quality Determination High

Study Citation:	Jarema, K. A., Hunter, D. L., Shaffer, R. M., Behl, M., Padilla, S. (2015). Acute and developmental behavioral effects of flame retardants and related chemicals in zebrafish. Neurotoxicology and Teratology 52(Pt B):194-209.
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*; Larvae
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4180931

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	CASRN, structure, molecular weight, nomenclature all were reported.
	Metric 2:	Test Substance Source	Low	Lot number and source was reported but not analytically verified by the lab.
	Metric 3:	Test Substance Purity	High	99% purity reported from the manufacturer.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Controls with carrier (DMSO) were used. Positive controls were also employed.
	Metric 5:	Negative Control Response	High	Neurotypical control movement was well documented from control larvae (Figure 2).
	Metric 6:	Randomized Allocation	Medium	Random allocation of treatment concentrations and control groups was described by plate locations.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The preparation of test solutions is well described and appropriate.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently among control and treatment concentrations.
	Metric 9:	Measurement of Test Substance Concentration	Medium	Exposure concentrations were not measured but renewal and protection from evaporation were conducted.
	Metric 10:	Exposure Duration and Frequency	High	This study exposed 6 dpf larvae to an acute concentration of the test compound to observe acute behavioral responses.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	5 concentrations were employed and previous range finding was conducted.
	Metric 12:	Testing at or Below Solubility Limit	Medium	The highest test concentration 12 uMol is over the solubility limit as reported in the final scope at 4.1 mg/L.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Source of test organism were listed and reliable.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Test and pretreatment conditions were not different and were well detailed.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The number of larvae per treatment concentration are reported on page 25 for the acute exposure assay.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Although no standard method listed, the culture conditions during the test are appropriate for zebrafish embryos.
	Metric 17:	Outcome Assessment Methodology	High	Outcome assessment was identified and appropriate.

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Study Citation:	Jarema, K. A., Hunter, D. L., Shaffer, R. M., Behl, M., Padilla, S. (2015). Acute and developmental behavioral effects of flame retardants and related chemicals in zebrafish. Neurotoxicology and Teratology 52(Pt B):194-209.
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*; Larvae
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4180931

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	High	Assessments were conducted consistency among treatments and control.
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	Medium	No information was reported to suggest differences among treatment groups in animal attrition.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	"The data were first assessed using a repeated-measures analysis of variance (ANOVA) with time and dose as the independent variables and locomotor activity (distance moved/time) as the dependent variable. All of the results of this repeated-measures ANOVA for both the developmental and acute (time of peak effect) assessments are presented in Supplemental Table 2."
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented.
	Metric 23:	Explanation of Unexpected Outcomes	High	Unexplained or unexpected outcomes were not encountered in the current study.

Additional Comments: A high-quality study that exposed 6-dpf zebrafish larvae to TBBPA at 5 concentrations and a control as an acute exposure. Behavioral movement metrics were assessed from this acute exposure. Behavioral metrics based on movement (distance moved) are presented at 6 days post fertilization. Data are presented on Figure 4, page 25.

Overall Quality Determination High

Study Citation:	Kalasekar, S. M., Zacharia, E., Kessler, N., Ducharme, N. A., Gustafsson, J. Å., Kakadiaris, I. A., Bondesson, M. (2015). Identification of environmental chemicals that induce yolk malabsorption in zebrafish using automated image segmentation. Reproductive Toxicology 55:20-29.
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*; HGn50D; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3981891

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory; source reported as Sigma–Aldrich (St. Louis, MO).
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 97%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate solvent 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 no details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Medium	Reporting omissions were unlikely to have a substantial impact on results
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	Low	Exposure duration covered the yolk absorption portion of development but was shorter than usual
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	The number of exposure groups and spacing of exposure levels were not reported
	Metric 12:	Testing at or Below Solubility Limit	Low	The exposure concentration range was not reported
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	It was unclear if they used the transgenic strain for the toxicity test
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	pretreatment conditions were not reported
	Metric 15:	Number of Organisms and Replicates per Group	Low	The numbers of test organisms and replicates were not reported
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reported environmental conditions were not sufficient to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not reported

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Study Citation:	Kalasekar, S. M., Zacharia, E., Kessler, N., Ducharme, N. A., Gustafsson, J. Å., Kakadiaris, I. A., Bondesson, M. (2015). Identification of environmental chemicals that induce yolk malabsorption in zebrafish using automated image segmentation. Reproductive Toxicology 55:20-29.
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*; HGn50D; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3981891

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	Low	outcomes assessment methodology was not reported
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Medium	environmental conditions were not reported
	Metric 20:	Outcomes Unrelated to Exposure	Medium	there were no differences among groups but no details provided
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 presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	Medium	unexpected outcomes were not reported

Additional Comments: None

Overall Quality Determination Low

Study Citation:	Kalasekar, S. M., Zacharia, E., Kessler, N., Ducharme, N. A., Gustafsson, J. Å., Kakadiaris, I. A., Bondesson, M. (2015). Identification of environmental chemicals that induce yolk malabsorption in zebrafish using automated image segmentation. Reproductive Toxicology 55:20-29.
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*; HGn50D; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3981891

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory; source reported as Sigma–Aldrich (St. Louis, MO).
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 97%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate solvent control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The study provided limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Medium	Reporting omissions were unlikely to have a substantial impact on results
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	Medium	Exposure duration covered the yolk absorption portion of development
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Reported environmental conditions were not sufficient to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups

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Study Citation:	Kalasekar, S. M., Zacharia, E., Kessler, N., Ducharme, N. A., Gustafsson, J. Å., Kakadiaris, I. A., Bondesson, M. (2015). Identification of environmental chemicals that induce yolk malabsorption in zebrafish using automated image segmentation. Reproductive Toxicology 55:20-29.
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*; HGn50D; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3981891

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: yolk absorption

Overall Quality Determination High

Study Citation:	Lee, S. K., Freitag, D., Steinberg, C., Kettrup, A., Kim, Y. H. (1993). Effects of dissolved humic materials on acute toxicity of some organic chemicals to aquatic organisms. Water Research 27(2):199-204.
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*; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5518890

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Medium	Chemical was identified by name and structure
	Metric 2:	Test Substance Source	High	Sourced from "Kukdo Co, Korea". 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. "Technical grade" - no definition provided.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	use of appropriate controls was implied in Fig.1, but not stated directly.
	Metric 5:	Negative Control Response	Low	The biological response of the negative control groups was not reported.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided few details on exposure media preparation, and concentrations were not measured during the study.
	Metric 8:	Consistency of Exposure Administration	Medium	Reporting omissions were unlikely to have a substantial impact on results
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	exposure levels are not specified, there is only a range implied in Fig.1
	Metric 12:	Testing at or Below Solubility Limit	Low	Reporting omissions prevented determination of whether exposure concentrations exceeded the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test animals was not reported
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				

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Study Citation:	Lee, S. K., Freitag, D., Steinberg, C., Kettrup, A., Kim, Y. H. (1993). Effects of dissolved humic materials on acute toxicity of some organic chemicals to aquatic organisms. Water Research 27(2):199-204.
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*; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5518890

Domain	Metric		Rating	Comments
	Metric 16:	Adequacy of Test Conditions	Medium	Reported environmental conditions were not sufficient to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	One series with no added humic acid was used
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among group
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical analysis was performed and described adequately.
	Metric 22:	Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	Medium	There were no unexpected outcomes but few details were provided

Additional Comments: None

Overall Quality Determination Low

Study Citation:	Mccormick, J. M. (2010). Microbial transformations of tetrabromobisphenol a and its metabolites, and their impact on toxicity to the developing zebrafish (Danio rerio) embryo. Doctoral Dissertation:191.
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:	Mechanistic-Cell signaling/function-Receptor binding/ regulation of receptor activity
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5489020; Linked HERO ID(s): 5489020, 1294687

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The test substance was identified by chemical name and structure (Fig, 1.1, page 38/176).
	Metric 2:	Test Substance Source	Low	The source was reported but the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Percent purity was > 98%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group (i.e. all conditions equal except chemical exposure).
	Metric 5:	Negative Control Response	High	The biological responses of the negative control group were adequate.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	Experiment conducted in glass plates. Stock solutions were prepared in DMSO, which was also used as the vehicle control (0.1%). Concentration of test substance was not measured during the study.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study group and control.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure (72 hours post fertilization) was reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	There was only one exposure group but was justified by study authors and adequate to address the purpose of the study.
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate ((i.e., no effects on biological responses were observed in the solvent control and no interactions were expected between the solvent and test substance).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.

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Study Citation:	Mccormick, J. M. (2010). Microbial transformations of tetrabromobisphenol a and its metabolites, and their impact on toxicity to the developing zebrafish (Danio rerio) embryo. Doctoral Dissertation:191.
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:	Mechanistic-Cell signaling/function-Receptor binding/ regulation of receptor activity
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5489020; Linked HERO ID(s): 5489020, 1294687

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of test organisms and/or replicates was not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate and whether differences occurred between control and exposed population.
	Metric 17:	Outcome Assessment Methodology	Low	Outcome assessment methodology was not sensitive for the outcome of interest. Authors report that the data never fully replicated for the expression of cYP450 genes, so was not examined further. They also report that the trends in the protein data did not follow the trends in the RNA data and attribute this to analyses of total protein, which could have been confounded by widespread expression in the developing embryo.
	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 or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure (e.g., infection) that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Uninformative	Statistical analysis was not conducted. The authors did not have confidence in the data on CYP1A1, CYP1B1 expression and matrix metalloproteinases (Figures in the Appendix, A1 and A2), and excluded it from chapter 2 of the dissertation and the associated manuscript (HERO ID: 1294687).
	Metric 22:	Reporting of Data	Medium	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 evaluation form is only for the mechanistic data in the Appendix of the dissertation (Figures A1 and A2). The PECO relevant chapter of the dissertation (chapter 2 ; published in a journal (HERO ID:1294687)) has been reviewed independently.The authors did not have confidence in the data on CYP1A1, CYP1B1 expression and matrix metalloproteinases (Figures in the Appendix, A1 and A2), and excluded it from chapter 2 of the dissertation and the associated manuscript (HERO ID: 1294687).

Overall Quality Determination — Uninformative

Study Citation:	Mccormick, J. M., Paiva, M. S., Häggblom, M. M., Cooper, K. R., White, L. A. (2010). Embryonic exposure to tetrabromobisphenol A and its metabolites, bisphenol A and tetrabromobisphenol A dimethyl ether disrupts normal zebrafish (Danio rerio) development and matrix metalloproteinase expression. Aquatic Toxicology 100(3):255-262.
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:	Mechanistic-Biomarkers (exposure and effect)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1294687; Linked HERO ID(s): 5489020, 1294687

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 >98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent solvent control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was suitable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health

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Study Citation:	Mccormick, J. M., Paiva, M. S., Häggblom, M. M., Cooper, K. R., White, L. A. (2010). Embryonic exposure to tetrabromobisphenol A and its metabolites, bisphenol A and tetrabromobisphenol A dimethyl ether disrupts normal zebrafish (Danio rerio) development and matrix metalloproteinase expression. Aquatic Toxicology 100(3):255-262.
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:	Mechanistic-Biomarkers (exposure and effect)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1294687; Linked HERO ID(s): 5489020, 1294687

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	Medium	details of reported environmental conditions were not sufficient to evaluate if adequate
	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 clearly described
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: Mechanistic outcomes for 48h and 72h including gene expression and enzymes.

Overall Quality Determination High

Study Citation:	Mccormick, J. M., Paiva, M. S., Häggblom, M. M., Cooper, K. R., White, L. A. (2010). Embryonic exposure to tetrabromobisphenol A and its metabolites, bisphenol A and tetrabromobisphenol A dimethyl ether disrupts normal zebrafish (Danio rerio) development and matrix metalloproteinase expression. Aquatic Toxicology 100(3):255-262.
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:	Cardiovascular
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1294687; Linked HERO ID(s): 5489020, 1294687

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 >98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent solvent control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was suitable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of 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:	Mccormick, J. M., Paiva, M. S., Häggblom, M. M., Cooper, K. R., White, L. A. (2010). Embryonic exposure to tetrabromobisphenol A and its metabolites, bisphenol A and tetrabromobisphenol A dimethyl ether disrupts normal zebrafish (Danio rerio) development and matrix metalloproteinase expression. Aquatic Toxicology 100(3):255-262.
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:	Cardiovascular
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1294687; Linked HERO ID(s): 5489020, 1294687

Domain	Metric		Rating	Comments
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Medium	details of reported environmental conditions were not sufficient to evaluate if adequate
	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 clearly described
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: Heart rate responses for 48 and 27h exposures.

Overall Quality Determination High

Study Citation:	Parsons, A., Lange, A., Hutchinson, T. H., Miyagawa, S., Iguchi, T., Kudoh, T., Tyler, C. R. (2019). Molecular mechanisms and tissue targets of brominated flame retardants, BDE-47 and TBBPA, in embryo-larval life stages of zebrafish (Danio rerio). Aquatic Toxicology 209:99-112.
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*; casper(mitfa;roy) mutant strain; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5496817

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#.
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 97%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported via graph and was reasonable.
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided few details on exposure media preparation.
	Metric 8:	Consistency of Exposure Administration	Medium	No mention of irregularities in exposure administration.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	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. the test compound concentrations were all below the solubility listed in the final scope. (4.1 mg/L).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reported environmental conditions were not sufficient to evaluate if adequate.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.

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Study Citation:	Parsons, A., Lange, A., Hutchinson, T. H., Miyagawa, S., Iguchi, T., Kudoh, T., Tyler, C. R. (2019). Molecular mechanisms and tissue targets of brominated flame retardants, BDE-47 and TBBPA, in embryo-larval life stages of zebrafish (Danio rerio). Aquatic Toxicology 209:99-112.
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*; casper(mitfa;roy) mutant strain; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5496817

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There were no information presented 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: The percent of developmental deformities are reported on page 5.14, figure 2B.

Overall Quality Determination High

Study Citation:	Parsons, A., Lange, A., Hutchinson, T. H., Miyagawa, S., Iguchi, T., Kudoh, T., Tyler, C. R. (2019). Molecular mechanisms and tissue targets of brominated flame retardants, BDE-47 and TBBPA, in embryo-larval life stages of zebrafish (Danio rerio). Aquatic Toxicology 209:99-112.
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*; casper(mitfa;roy) mutant strain; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5496817

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#.
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 97%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported via graph and was reasonable.
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided few details on exposure media preparation.
	Metric 8:	Consistency of Exposure Administration	Medium	No mention of irregularities in exposure administration.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	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. the test compound concentrations were all below the solubility listed in the final scope. (4.1 mg/L).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reported environmental conditions were not sufficient to evaluate if adequate.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.

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Study Citation:	Parsons, A., Lange, A., Hutchinson, T. H., Miyagawa, S., Iguchi, T., Kudoh, T., Tyler, C. R. (2019). Molecular mechanisms and tissue targets of brominated flame retardants, BDE-47 and TBBPA, in embryo-larval life stages of zebrafish (Danio rerio). Aquatic Toxicology 209:99-112.
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*; casper(mitfa;roy) mutant strain; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5496817

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 were no information presented 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: Mortality data are presented on page 5/14.

Overall Quality Determination High

Study Citation:	Song, M., Liang, D., Liang, Y., Chen, M., Wang, F., Wang, H., Jiang, G. (2014). Assessing developmental toxicity and estrogenic activity of halogenated bisphenol A on zebrafish (Danio rerio). Chemosphere 112:275-281.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; Embryo
Health Outcome:	Cardiovascular
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2754370

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 >=98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control groups
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was 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 for making a stock solution
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The test organisms were adequately described unsure where they were obtained from
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest

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Study Citation:	Song, M., Liang, D., Liang, Y., Chen, M., Wang, F., Wang, H., Jiang, G. (2014). Assessing developmental toxicity and estrogenic activity of halogenated bisphenol A on zebrafish (Danio rerio). Chemosphere 112:275-281.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; Embryo
Health Outcome:	Cardiovascular
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2754370

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
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Unclear if statistical analysis was performed
	Metric 22:	Reporting of Data	Medium	Data (%) was shown in Table 1 and described in the text. No raw data was included.
	Metric 23:	Explanation of Unexpected Outcomes	Low	There were no measures of variability

Additional Comments: This evaluation is for the assessment of pericardial edema.

Overall Quality Determination High

Study Citation:	Song, M., Liang, D., Liang, Y., Chen, M., Wang, F., Wang, H., Jiang, G. (2014). Assessing developmental toxicity and estrogenic activity of halogenated bisphenol A on zebrafish (Danio rerio). Chemosphere 112:275-281.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2754370

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 >=98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control groups
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was 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 for making a stock solution
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The test organisms were adequately described unsure where they were obtained from
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups

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Study Citation:	Song, M., Liang, D., Liang, Y., Chen, M., Wang, F., Wang, H., Jiang, G. (2014). Assessing developmental toxicity and estrogenic activity of halogenated bisphenol A on zebrafish (Danio rerio). Chemosphere 112:275-281.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2754370

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
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Unclear if statistical analysis was performed
	Metric 22:	Reporting of Data	Low	Data for hatch was not shown for control group
	Metric 23:	Explanation of Unexpected Outcomes	Low	There were no measures of variability

Additional Comments: edema, hemorrhage

Overall Quality Determination High

Study Citation:	Yang, J., Chan, K. M. (2015). Evaluation of the toxic effects of brominated compounds (BDE-47, 99, 209, TBBPA) and bisphenol A (BPA) using a zebrafish liver cell line, ZFL. Aquatic Toxicology 159:138-147.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; ZFL cell line; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4162140

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The TBBPA CAS number was provided.
	Metric 2:	Test Substance Source	Low	TBBPA was purchased from Sigma-Aldrich Co. (St. Louis, USA). It was not reported if the test chemical was analytically verified or not.
	Metric 3:	Test Substance Purity	Medium	It was just mentioned that the chemical was of analytical grade with the highest purity available.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	The vehicle control was medium with 0.05% DMSO in the agonistic tests and 0.1 nM T3 in the antagonist tests.
	Metric 5:	Negative Control Response	High	The vehicle control response was shown in Figure 2 and appeared adequate for this outcome response.
	Metric 6:	Randomized Allocation	Low	Random allocation to the plates was not mentioned.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	Details on the test system were vague for the luciferase reporter gene system assay. Additionally, preparation of the test stock solutions was limited in detail.
	Metric 8:	Consistency of Exposure Administration	Medium	Due to some lack of details regarding the experimental system, there may be some inconsistencies in exposure administration.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	Cells were exposed for 24 hours. This was adequate to determine zfTRbeta agonistic or antagonist activity.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	Authors mentioned that test concentrations used were equivalent to 1%, 2.5%, 5%, 10%, 25%, and 37.5% of the 24-hour LC50 value. The actual test concentrations were not given.
	Metric 12:	Testing at or Below Solubility Limit	Low	Since the specific exposure concentrations were not provided, it is uncertain if they were below the solubility limit. However, DMSO solvent was used to prepare test solutions.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The zebrafish cell line was purchased from Cell Bank at the American Type Culture Collection (ATCC number CRL-2643TM). They were grown in a standard culture medium.

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Study Citation:	Yang, J., Chan, K. M. (2015). Evaluation of the toxic effects of brominated compounds (BDE-47, 99, 209, TBBPA) and bisphenol A (BPA) using a zebrafish liver cell line, ZFL. Aquatic Toxicology 159:138-147.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; ZFL cell line; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4162140

Domain	Metric		Rating	Comments
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	There wasn't any mention of acclimation prior to the luciferase reporter gene system assay. Authors just mentioned that cells were seeded in 48-well plates and incubated for 24 hours to achieve around 70% confluence before the start of the assay.
	Metric 15:	Number of Organisms and Replicates per Group	Low	There was no mention of replicates. Cells were seeded on the 48-well plate at a density of 2 x 10^4 cells per well.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	There was no mention of environmental conditions (hood parameters) throughout the assay.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment was described in section 2.5.
	Metric 18:	Consistency of Outcome Assessment	High	It appeared the outcome assessment was carried out consistently across treatment groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	There was no mention of acclimation prior to the assay start and there was no information provided on environmental conditions throughout the course of the assay.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information provided to suggest any differences in cells that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	All statistical analyses were carried out in the PRISM 6 programwith one-way ANOVA set at a 95% confidence level, and by Tukey'smultiple comparison tests on a personal computer.
	Metric 22:	Reporting of Data	High	Section 3.4 described the results of the luciferase reporter gene system assay and Figure 2 shows the results of the assay.
	Metric 23:	Explanation of Unexpected Outcomes	High	Measures of variability were shown in Figure 2.

Additional Comments: This previously supplemental reference satisfies a data need and therefore is evaluated here. A zebrafish liver cell line was used for exposure to TBBPA. This evaluation is for the luciferase reporter gene system assay.

Overall Quality Determination Medium

Study Citation:	Yang, J., Chan, K. M. (2015). Evaluation of the toxic effects of brominated compounds (BDE-47, 99, 209, TBBPA) and bisphenol A (BPA) using a zebrafish liver cell line, ZFL. Aquatic Toxicology 159:138-147.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; ZFL cell line; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4162140

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The TBBPA CAS number was provided.
	Metric 2:	Test Substance Source	Low	TBBPA was purchased from Sigma-Aldrich Co. (St. Louis, USA). It was not reported if the test chemical was analytically verified or not.
	Metric 3:	Test Substance Purity	Medium	It was just mentioned that the chemical was of analytical grade with the highest purity available.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	A vehicle control group (0.01% DMSO) was used.
	Metric 5:	Negative Control Response	High	The vehicle control response was shown in Figure 3C and appeared adequate for this outcome response.
	Metric 6:	Randomized Allocation	Low	Random allocation of the cells to treatment groups was not mentioned.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	Details on the test system were vague for the gene expression assay. Additionally, preparation of the test stock solutions was limited in detail.
	Metric 8:	Consistency of Exposure Administration	Medium	Due to some lack of details regarding the experimental system, there may be some inconsistencies in exposure administration.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	Cells were exposed for 24 hours. This was adequate to determine gene expression activity.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Three concentrations of the test chemical were used. They were either 10%, 25%, and 50% of the 24h or 96h LC50 values. The specific concentrations were not given.
	Metric 12:	Testing at or Below Solubility Limit	Low	Since the specific exposure concentrations were not provided, it is uncertain if they were below the solubility limit. However, DMSO solvent was used to prepare test solutions.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The zebrafish cell line was purchased from Cell Bank at the American Type Culture Collection (ATCC number CRL-2643TM). They were grown in a standard culture medium.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	There wasn't any mention of acclimation prior to the gene expression assay. Authors just mentioned that cells were cultured in 10cm dishes.
	Metric 15:	Number of Organisms and Replicates per Group	Low	There was no mention of replicates. There was no mention the amount of cells per dish.

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Study Citation:	Yang, J., Chan, K. M. (2015). Evaluation of the toxic effects of brominated compounds (BDE-47, 99, 209, TBBPA) and bisphenol A (BPA) using a zebrafish liver cell line, ZFL. Aquatic Toxicology 159:138-147.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; ZFL cell line; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4162140

Domain	Metric		Rating	Comments
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	There was no mention of environmental conditions (hood parameters) throughout the assay.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment was described in section 2.7.
	Metric 18:	Consistency of Outcome Assessment	High	It appeared the outcome assessment was carried out consistently across treatment groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	There was no mention of acclimation prior to the assay start and there was no information provided on environmental conditions throughout the course of the assay.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information provided to suggest any differences in cells that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	All statistical analyses were carried out in the PRISM 6 programwith one-way ANOVA set at a 95% confidence level, and by Tukey'smultiple comparison tests on a personal computer.
	Metric 22:	Reporting of Data	High	Section 3.6 described the results of the gene expression assay and Figure 3C shows the results of the assay.
	Metric 23:	Explanation of Unexpected Outcomes	High	Measures of variability were shown in Figure 3C.

Additional Comments: This previously supplemental reference satisfies a data need and therefore is evaluated here. A zebrafish liver cell line was used for exposure to TBBPA. This evaluation is for the gene expression assay.

Overall Quality Determination Medium

Study Citation:	Yang, J., Chan, K. M. (2015). Evaluation of the toxic effects of brominated compounds (BDE-47, 99, 209, TBBPA) and bisphenol A (BPA) using a zebrafish liver cell line, ZFL. Aquatic Toxicology 159:138-147.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; ZFL cell line; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Cytotoxicity
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4162140

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The TBBPA CAS number was provided.
	Metric 2:	Test Substance Source	Low	TBBPA was purchased from Sigma-Aldrich Co. (St. Louis, USA). It was not reported if the test chemical was analytically verified or not.
	Metric 3:	Test Substance Purity	Medium	It was just mentioned that the chemical was of analytical grade with the highest purity available.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	A control was used.
	Metric 5:	Negative Control Response	Medium	For the cytotoxicity test, the control response was not specifically shown but the cytotoxicity values presented in Figure 1 were calculated using fluorescence readings of the control and treatment groups. Also, study authors mentioned supplementary material can be found in an online link, so more data is likely shown there.
	Metric 6:	Randomized Allocation	Low	Random allocation to the plates was not mentioned.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	Details on the test system were vague for the cytotoxicity assay. Additionally, preparation of the test stock solutions was limited in detail. Exposure concentrations were not given.
	Metric 8:	Consistency of Exposure Administration	Low	Due to some lack of details regarding the experimental system, there may be some inconsistencies in exposure administration.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	Cells were exposed for 24 and 96 hours. This was adequate to determine LC50 values.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Uninformative	No information was provided on the number of exposure groups used in the cytotoxicity assay.
	Metric 12:	Testing at or Below Solubility Limit	Low	Since exposure concentrations were not provided, it is uncertain if they were below the solubility limit. However, DMSO solvent was used to prepare test solutions.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The zebrafish cell line was purchased from Cell Bank at the American Type Culture Collection (ATCC number CRL-2643TM). They were grown in a standard culture medium.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	There wasn't any mention of acclimation prior to the cytotoxicity assay. Authors just mentioned that cells were seeded on 96-well plates and incubated overnight to achieve around 80% confluence before the start of the assay.

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Study Citation:	Yang, J., Chan, K. M. (2015). Evaluation of the toxic effects of brominated compounds (BDE-47, 99, 209, TBBPA) and bisphenol A (BPA) using a zebrafish liver cell line, ZFL. Aquatic Toxicology 159:138-147.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; ZFL cell line; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Cytotoxicity
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4162140

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Low	There was no mention of replicates. Cells were seeded on the 96-well plate at a density of 1 x 10^4 cells per well.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	There was no mention of environmental conditions (hood parameters) throughout the assay.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment was described in section 2.3.
	Metric 18:	Consistency of Outcome Assessment	High	It appeared the outcome assessment was carried out consistently across treatment groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	There was no mention of acclimation prior to the assay start and there was no information provided on environmental conditions throughout the course of the assay.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information provided to suggest any differences in cells that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	All statistical analyses were carried out in the PRISM 6 programwith one-way ANOVA set at a 95% confidence level, and by Tukey'smultiple comparison tests on a personal computer.
	Metric 22:	Reporting of Data	High	Section 3.1 described the results of the cytotoxicity assay and Figures 1I and 1J show the cell death data at 24 and 96 hours.
	Metric 23:	Explanation of Unexpected Outcomes	High	Measures of variability were shown in Figure 1.

Additional Comments: This previously supplemental reference satisfies a data need and therefore is evaluated here. A zebrafish liver cell line was used for exposure to TBBPA. This evaluation is for the cytotoxicity assay. This got an unacceptable rating because there was no mention of the exposure groups used in the assay.

Overall Quality Determination Uninformative

Study Citation:	Yang, S., Wang, S., Sun, F., Zhang, M., Wu, F., Xu, F., Ding, Z. (2015). Protective effects of puerarin against tetrabromobisphenol a-induced apoptosis and cardiac developmental toxicity in zebrafish embryo-larvae. Environmental Toxicology 30(9):1014-1023.
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 wild type; Embryo
Health Outcome:	Mechanistic-Immune/Hematological
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5490833

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and structure.
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >= 99%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable (Figure 2).
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The study provided few details on exposure media preparation.
	Metric 8:	Consistency of Exposure Administration	Medium	Reporting omissions were unlikely to have a substantial impact on results.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not analyzed so nominal concentrations are reported. Reporting omissions were unlikely to have a substantial impact on results.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	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	Reporting omissions prevented determination of whether solvent concentration slightly exceeded an appropriate concentration. TBBPA concentrations were under the solubility listed in the final scope (4.1 mg/L).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Pretreatment and treatment conditions appear to be similar from the methods reported on page 2/10.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				

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Study Citation:	Yang, S., Wang, S., Sun, F., Zhang, M., Wu, F., Xu, F., Ding, Z. (2015). Protective effects of puerarin against tetrabromobisphenol a-induced apoptosis and cardiac developmental toxicity in zebrafish embryo-larvae. Environmental Toxicology 30(9):1014-1023.
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 wild type; Embryo
Health Outcome:	Mechanistic-Immune/Hematological
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5490833

Domain	Metric		Rating	Comments
	Metric 16:	Adequacy of Test Conditions	Medium	Reported environmental conditions were not sufficient to evaluate if adequate. incubation conditions are reported within the section titled ”detection of morphological indices”.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18:	Consistency of Outcome Assessment	Medium	few details regarding the execution of the study protocol for outcome assessment were provided. for example ”The morphological indices monitored in this test included blood flow disorders, pericardial edema, heart rate, malformation, spawn coagulation, and survival rate.”
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 information presented 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. Measures of variability were provided in the figures.

Additional Comments: This evaluation is for the assessment of blood flow.

Overall Quality Determination High

Study Citation:	Yang, S., Wang, S., Sun, F., Zhang, M., Wu, F., Xu, F., Ding, Z. (2015). Protective effects of puerarin against tetrabromobisphenol a-induced apoptosis and cardiac developmental toxicity in zebrafish embryo-larvae. Environmental Toxicology 30(9):1014-1023.
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 wild type; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5490833

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and structure.
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >= 99%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable (Figure 2).
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The study provided few details on exposure media preparation.
	Metric 8:	Consistency of Exposure Administration	Medium	Reporting omissions were unlikely to have a substantial impact on results.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not analyzed so nominal concentrations are reported. Reporting omissions were unlikely to have a substantial impact on results.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	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	Reporting omissions prevented determination of whether solvent concentration slightly exceeded an appropriate concentration. TBBPA concentrations were under the solubility listed in the final scope (4.1 mg/L).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Pretreatment and treatment conditions appear to be similar from the methods reported on page 2/10.
	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	Medium	Reported environmental conditions were not sufficient to evaluate if adequate. incubation conditions are reported within the section titled ”detection of morphological indices”.

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Study Citation:	Yang, S., Wang, S., Sun, F., Zhang, M., Wu, F., Xu, F., Ding, Z. (2015). Protective effects of puerarin against tetrabromobisphenol a-induced apoptosis and cardiac developmental toxicity in zebrafish embryo-larvae. Environmental Toxicology 30(9):1014-1023.
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 wild type; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5490833

Domain	Metric		Rating	Comments
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18:	Consistency of Outcome Assessment	Medium	few details regarding the execution of the study protocol for outcome assessment were provided. for example "The morphological indices monitored in this test included blood flow disorders, pericardial edema, heart rate, malformation, spawn coagulation, and survival rate."
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 information presented to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This evaluation is for malformations and pericardial edema.

Overall Quality Determination High

Study Citation:	Yang, S., Wang, S., Sun, F., Zhang, M., Wu, F., Xu, F., Ding, Z. (2015). Protective effects of puerarin against tetrabromobisphenol a-induced apoptosis and cardiac developmental toxicity in zebrafish embryo-larvae. Environmental Toxicology 30(9):1014-1023.
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 wild type; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Cytotoxicity-Oxidative stress (including redox biology)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5490833

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and structure.
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >= 99%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The study provided few details on exposure media preparation
	Metric 8:	Consistency of Exposure Administration	Medium	Reporting omissions were unlikely to have a substantial impact on results
	Metric 9:	Measurement of Test Substance Concentration	Low	Reporting omissions were unlikely to have a substantial impact on results
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	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	Reporting omissions prevented determination of whether solvent concentration slightly exceeded an appropriate concentration. TBBPA concentrations were under the solubility listed in the final scope (4.1 mg/L).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Pretreatment and treatment conditions appear to be similar from the methods reported on page 2/10.
	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	Medium	Reported environmental conditions were not sufficient to evaluate if adequate. incubation conditions are reported within the section titled ”detection of morphological indices”.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest

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Study Citation:	Yang, S., Wang, S., Sun, F., Zhang, M., Wu, F., Xu, F., Ding, Z. (2015). Protective effects of puerarin against tetrabromobisphenol a-induced apoptosis and cardiac developmental toxicity in zebrafish embryo-larvae. Environmental Toxicology 30(9):1014-1023.
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 wild type; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Cytotoxicity-Oxidative stress (including redox biology)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5490833

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	Medium	Few details regarding the execution of the study protocol for outcome assessment were provided. for example "The morphological indices monitored in this test included blood flow disorders, pericardial edema, heart rate, malformation, spawn coagulation, and survival rate."
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 information presented 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: apoptosis, ROS activity, gene expression, mixtures with puerarin also reported

Overall Quality Determination High

Study Citation:	Yang, S., Wang, S., Sun, F., Zhang, M., Wu, F., Xu, F., Ding, Z. (2015). Protective effects of puerarin against tetrabromobisphenol a-induced apoptosis and cardiac developmental toxicity in zebrafish embryo-larvae. Environmental Toxicology 30(9):1014-1023.
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 wild type; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5490833

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and structure.
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >= 99%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The study provided few details on exposure media preparation
	Metric 8:	Consistency of Exposure Administration	Medium	Reporting omissions were unlikely to have a substantial impact on results
	Metric 9:	Measurement of Test Substance Concentration	Low	Reporting omissions were unlikely to have a substantial impact on results
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	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	Reporting omissions prevented determination of whether solvent concentration slightly exceeded an appropriate concentration. TBBPA concentrations were under the solubility listed in the final scope (4.1 mg/L).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Pretreatment and treatment conditions appear to be similar from the methods reported on page 2/10.
	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	Medium	Reported environmental conditions were not sufficient to evaluate if adequate. incubation conditions are reported within the section titled ”detection of morphological indices”.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest

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Study Citation:	Yang, S., Wang, S., Sun, F., Zhang, M., Wu, F., Xu, F., Ding, Z. (2015). Protective effects of puerarin against tetrabromobisphenol a-induced apoptosis and cardiac developmental toxicity in zebrafish embryo-larvae. Environmental Toxicology 30(9):1014-1023.
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 wild type; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5490833

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	Medium	few details regarding the execution of the study protocol for outcome assessment were provided. for example "The morphological indices monitored in this test included blood flow disorders, pericardial edema, heart rate, malformation, spawn coagulation, and survival rate."
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 information presented 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: "Survival was also decreased significantly in the 0.5 and 1.0 mg L21 groups ($p<0.05$). " Page 3/10Figure 2, Page 4/10

Overall Quality Determination High

Study Citation:	Baumann, L., Ros, A., Rehberger, K., Neuhauss, S. C., Segner, H. (2016). Thyroid disruption in zebrafish (Danio rerio) larvae: Different molecular response patterns lead to impaired eye development and visual functions. Aquatic Toxicology 172:44-55.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wild type; Embryo
Health Outcome:	Ocular and Sensory
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4199233

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity 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 reported and adequate.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The experimental system and test media preparation methods were adequately reported. Unclear whether the same stock solution was used for all exposure solutions. Exposure solutions were renewed daily.
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups. Exposure solution volume was 2 ml and the same solvent (DMSO) was used across replicates and treatment groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	Embryos were exposed for 5 days post fertilization and was appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Exposure concentrations were 0, 100, 200, 300 and 400 ug/L forTBBPA and were adequate to show results relevant to the outcome of interest. "These concentrations were chosen based on range-finding tests to determine treatments that were sub-lethal."
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects. For eye diameter measurements and histological analysis, 24 euthanized larvae each were used.

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Study Citation:	Baumann, L., Ros, A., Rehberger, K., Neuhauss, S. C., Segner, H. (2016). Thyroid disruption in zebrafish (Danio rerio) larvae: Different molecular response patterns lead to impaired eye development and visual functions. Aquatic Toxicology 172:44-55.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wild type; Embryo
Health Outcome:	Ocular and Sensory
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4199233

Domain	Metric		Rating	Comments
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Organism housing and environmental conditions were reported and conducive to the maintenance of health. Zebrafish maintenance are described in section 2.2. ”Plates were incubated at 28 °C under a day/night cycle of 14/10 h.”
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest. Methods used for eye size measurements and histological analysis of eyes were provided in detail.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors.
	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. Data for eye size was given in Fig 5 and data for eye pigmentation was given in Fig 3.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: This evaluation form is for ocular outcomes (eye size and pigmentation) determined in zebrafish following exposure to 0, 100, 200, 300 and 400 ug/L of TBBPA.

Overall Quality Determination High

Study Citation:	Baumann, L., Ros, A., Rehberger, K., Neuhauss, S. C., Segner, H. (2016). Thyroid disruption in zebrafish (Danio rerio) larvae: Different molecular response patterns lead to impaired eye development and visual functions. Aquatic Toxicology 172:44-55.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wild type; Embryo
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4199233

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity 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 reported and adequate.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The experimental system and test media preparation methods were adequately reported. Unclear whether the same stock solution was used for all exposure solutions. Exposure solutions were renewed daily.
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups. Exposure solution volume was 2 ml and the same solvent (DMSO) was used across replicates and treatment groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	Embryos were exposed for 5 days post fertilization and was appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Exposure concentrations were 0, 100, 200, 300 and 400 ug/L forTBBPA and were adequate to show results relevant to the outcome of interest. ”These concentrations were chosen based on range-finding tests to determinetreatments that were sub-lethal.”
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects. For optokinetic response, ”single larvae were taken from exposure plates” for measuring responses of the eye to different contrasts, velocities and frequencies of moving stripes. Three independent replicates For phototactic behavior experiment, a total of 449 larvae (across 4 replicates) were used.

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Study Citation:	Baumann, L., Ros, A., Rehberger, K., Neuhauss, S. C., Segner, H. (2016). Thyroid disruption in zebrafish (Danio rerio) larvae: Different molecular response patterns lead to impaired eye development and visual functions. Aquatic Toxicology 172:44-55.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wild type; Embryo
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4199233

Domain	Metric		Rating	Comments
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Organism housing and environmental conditions were reported and conducive to the maintenance of health. Zebrafish maintenance are described in section 2.2. "Plates were incubated at 28 °C under a day/night cycle of 14/10 h."
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest. Methods for optokinetic response and phototactic swimming activity were given in detail.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors.
	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 for optokinetic response and phototactic swimming activity and light-dark preference were presented for each treatment and control group in figures 6 and 7.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This evaluation form is for behavior outcomes (optokinetic response and phototactic swimming activity and light-dark preference) measured in zebrafish following exposure to 0, 100, 200, 300 and 400 ug/L of TBBPA.

Overall Quality Determination High

Study Citation:	Baumann, L., Ros, A., Rehberger, K., Neuhauss, S. C., Segner, H. (2016). Thyroid disruption in zebrafish (Danio rerio) larvae: Different molecular response patterns lead to impaired eye development and visual functions. Aquatic Toxicology 172:44-55.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wild type; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4199233

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity 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 reported and reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The experimental system and test media preparation methods were adequately reported. Unclear whether the same stock solution was used for all exposure solutions. Exposure solutions were renewed daily.
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups. Exposure solution volume was 2 ml and the same solvent (DMSO) was used across replicates and treatment groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	Embryos were exposed for 5 days post fertilization and was appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Exposure concentrations were 0, 100, 200, 300 and 400 ug/L forTBBPA and were adequate to show results relevant to the outcome of interest. "These concentrations were chosen based on range-finding tests to determinetreatments that were sub-lethal."
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				

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Study Citation:	Baumann, L., Ros, A., Rehberger, K., Neuhauss, S. C., Segner, H. (2016). Thyroid disruption in zebrafish (Danio rerio) larvae: Different molecular response patterns lead to impaired eye development and visual functions. Aquatic Toxicology 172:44-55.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wild type; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4199233

Domain	Metric		Rating	Comments
	Metric 16:	Adequacy of Test Conditions	High	Organism housing and environmental conditions were reported and conducive to the maintenance of health. Zebrafish maintenance are described in section 2.2. "Plates were incubated at 28 °C under a day/night cycle of 14/10 h."
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology for mRNA expression 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 or other factors.
	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 the mechanistic outcome (mRNA expression) measured in zebrafish following exposure to 0, 100, 200, 300 and 400 ug/L of TBBPA.

Overall Quality Determination High

Study Citation:	Behl, M., Hsieh, J. H., Shafer, T. J., Mundy, W. R., Rice, J. R., Boyd, W. A., Freedman, J. H., Hunter, E. S., Jarema, K. A., Padilla, S., Tice, R. R. (2015). Use of alternative assays to identify and prioritize organophosphorus flame retardants for potential developmental and neurotoxicity. Neurotoxicology and Teratology 52(Pt B):181-193.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; wild type; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3479540

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent solvent control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided few details on exposure media preparation
	Metric 8:	Consistency of Exposure Administration	Low	The study provided few details on exposure administration
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were justified for a dose response by study authors
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
	Metric 15:	Number of Organisms and Replicates per Group	Low	4 embryos per well, the use of replicates was not reported
Domain 5: Outcome Assessment				

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Study Citation:	Behl, M., Hsieh, J. H., Shafer, T. J., Mundy, W. R., Rice, J. R., Boyd, W. A., Freedman, J. H., Hunter, E. S., Jarema, K. A., Padilla, S., Tice, R. R. (2015). Use of alternative assays to identify and prioritize organophosphorus flame retardants for potential developmental and neurotoxicity. Neurotoxicology and Teratology 52(Pt B):181-193.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; wild type; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3479540

Domain	Metric		Rating	Comments
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	Medium	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 (PODs)
	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, use of PODs was adequate

Additional Comments: abnormalities

Overall Quality Determination Medium

Study Citation:	Behl, M., Hsieh, J. H., Shafer, T. J., Mundy, W. R., Rice, J. R., Boyd, W. A., Freedman, J. H., Hunter, E. S., Jarema, K. A., Padilla, S., Tice, R. R. (2015). Use of alternative assays to identify and prioritize organophosphorus flame retardants for potential developmental and neurotoxicity. Neurotoxicology and Teratology 52(Pt B):181-193.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; wild type; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3479540

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent solvent control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided few details on exposure media preparation
	Metric 8:	Consistency of Exposure Administration	Low	The study provided few details on exposure administration
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were justified for a dose response by study authors
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
	Metric 15:	Number of Organisms and Replicates per Group	Low	4 embryos per well, the use of replicates was not reported
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest

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Study Citation:	Behl, M., Hsieh, J. H., Shafer, T. J., Mundy, W. R., Rice, J. R., Boyd, W. A., Freedman, J. H., Hunter, E. S., Jarema, K. A., Padilla, S., Tice, R. R. (2015). Use of alternative assays to identify and prioritize organophosphorus flame retardants for potential developmental and neurotoxicity. Neurotoxicology and Teratology 52(Pt B):181-193.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; wild type; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3479540

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	Medium	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 (PODs)
	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, use of PODs was adequate

Additional Comments: abnormalities

Overall Quality Determination Medium

Study Citation:	Fraser, K., T.W., Khezri, A., Jusdado, H., J.G., Lewandowska-Sabat, A. M., Henry, T., Ropstad, E. (2017). Toxicant induced behavioural aberrations in larval zebrafish are dependent on minor methodological alterations. Toxicology Letters 276:62-68.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wildtype; Embryo
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5549558

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The TBBPA was identified by name only.
	Metric 2:	Test Substance Source	Low	The source of the TBBPA was reported to be Sigma Aldrich. However, it was not reported to be analytically verified.
	Metric 3:	Test Substance Purity	High	The purity was reported to be 97%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of a concurrent negative control in which DMSO was used as a solvent at 0.01%.
	Metric 5:	Negative Control Response	High	The biological response of the negative control was reported in Fig. 1D, and was appropriate for the outcome of interest.
	Metric 6:	Randomized Allocation	Low	It was not reported how the embryos were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The test system was reported to be static in 96-well plates. One embryo was placed in each well with 200uL of test solution. The stock solution was reported to be prepared by mixing TBBPA in DMSO, but the preparation of the test concentrations was not reported. The concentration of the stock solution was not reported.
	Metric 8:	Consistency of Exposure Administration	Low	All exposures occurred in 96-well plates at 28C with a 14L:10D photoperiod with 200uL of test solution in each well with one embryo. However, the preparation of the test concentrations were not reported, so there are uncertainties regarding consistency.
	Metric 9:	Measurement of Test Substance Concentration	Low	It was not reported if the test concentrations were measured at any point in the study.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration was reported to be up to 100hpf starting at 6-7hpf. This appeared adequate for the outcome of interest.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	There were five exposure groups ranging from 150pM to 1.5uM. They are listed in Fig 1C and D. Spacing appeared adequate for the outcome of interest.
	Metric 12:	Testing at or Below Solubility Limit	High	Study authors reported the use of DMSO as a solvent control at 0.01% in the control and in the exposure groups. The solvent control response was adequate for the outcome of interest.
Domain 4: Test Organism				

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Study Citation:	Fraser, K., T.W., Khezri, A., Jusdado, H., J.G., Lewandowska-Sabat, A. M., Henry, T., Ropstad, E. (2017). Toxicant induced behavioural aberrations in larval zebrafish are dependent on minor methodological alterations. Toxicology Letters 276:62-68.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wildtype; Embryo
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5549558

Domain	Metric		Rating	Comments
	Metric 13:	Test Organism Characteristics	High	The zebrafish were reported to be cultured in house at the Section for Experimental Biomedicine at the Norwegian University of Life Sciences in Oslo, Norway. Embryos were collected 1hpf and placed in embryo media until the time of exposure.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the embryos were acclimated prior to testing.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	There was one embryo per well in the 96-well plate. There were 16 replicates per test concentration with a total of 96 embryos per group.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	The embryo tests were conducted at 28C with a 14L:10D photoperiod. The spawning adults that the embryos were obtained from were kept at 28C as well with a 14L:10D photoperiod. They were fed 3 x a day, 2 x with artemia and once with dry food.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest–changes in behavior due to TBBPA exposure.
	Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. Larval behavior was assessed at two times for TBBPA, 96 and 100hpf using ViewPoint Zebrabox and the accompanying video tracking software. Larval behavior monitored included the cumulative distance traveled and time spent active per minute during a light-dark-light cycle that lasted for 40 min.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. It was not reported if any acclimation occurred.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	All data were transferred to R version 2.15.0 for behavioral analysis. Data was checked form normality using visual examination, and linear mixed effect models were used for all analyses. More information is provided in section 2.6.
	Metric 22:	Reporting of Data	High	Control and exposure responses for the day/night (DN) cycle treatments were provided in Fig 1D and were adequate for the outcome of interest.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in Fig 1.

Additional Comments: This evaluation was on the effect of TBBPA on zebrafish larval behavior after 96hpf or 100hpf. 6-7hpf embryos were used at test initiation. The embryos used in this portion of the study had a photo-regime of 14L:10D. The outcome of interest was the behavioral outcome.

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Study Citation:	Fraser, K., T.W., Khezri, A., Jusdado, H., J.G., Lewandowska-Sabat, A. M., Henry, T., Ropstad, E. (2017). Toxicant induced behavioural aberrations in larval zebrafish are dependent on minor methodological alterations. Toxicology Letters 276:62-68.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wildtype; Embryo
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5549558

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

Study Citation:	Fraser, K., T.W., Khezri, A., Jusdado, H., J.G., Lewandowska-Sabat, A. M., Henry, T., Ropstad, E. (2017). Toxicant induced behavioural aberrations in larval zebrafish are dependent on minor methodological alterations. Toxicology Letters 276:62-68.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wildtype; Embryo
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5549558

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The TBBPA was identified by name only.
	Metric 2:	Test Substance Source	Low	The source of the TBBPA was reported to be Sigma Aldrich. However, it was not reported to be analytically verified.
	Metric 3:	Test Substance Purity	High	The purity was reported to be 97%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of a concurrent negative control in which DMSO was used as a solvent at 0.01%.
	Metric 5:	Negative Control Response	High	The biological response of the negative control was reported in Fig. 1C, and was appropriate for the outcome of interest.
	Metric 6:	Randomized Allocation	Low	It was not reported how the embryos were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The test system was reported to be static in 96-well plates. One embryo was placed in each well with 200uL of test solution. The stock solution was reported to be prepared by mixing TBBPA in DMSO, but the preparation of the test concentrations was not reported. The concentration of the stock solution was not reported.
	Metric 8:	Consistency of Exposure Administration	Low	All exposures occurred in 96-well plates at 28C with a 14L:10D photoperiod with 200uL of test solution in each well with one embryo. In the case of the study with complete darkness, the plates were placed in the incubation chamber with the previously mentioned photoperiod, but they were wrapped in aluminum foil to prevent exposure to light. However, the preparation of the test concentrations were not reported, so there are uncertainties regarding consistency.
	Metric 9:	Measurement of Test Substance Concentration	Low	It was not reported if the test concentrations were measured at any point in the study.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration was reported to be up to 100hpf starting at 6-7hpf. This appeared adequate for the outcome of interest.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	There were five exposure groups ranging from 150pM to 1.5uM. They are listed in Fig 1C and D. Spacing appeared adequate for the outcome of interest.
	Metric 12:	Testing at or Below Solubility Limit	High	Study authors reported the use of DMSO as a solvent control at 0.01% in the control and in the exposure groups. The solvent control response was adequate for the outcome of interest.
Domain 4: Test Organism				

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Study Citation:	Fraser, K., T.W., Khezri, A., Jusdado, H., J.G., Lewandowska-Sabat, A. M., Henry, T., Ropstad, E. (2017). Toxicant induced behavioural aberrations in larval zebrafish are dependent on minor methodological alterations. Toxicology Letters 276:62-68.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wildtype; Embryo
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5549558

Domain	Metric		Rating	Comments
	Metric 13:	Test Organism Characteristics	High	The zebrafish were reported to be cultured in house at the Section for Experimental Biomedicine at the Norwegian University of Life Sciences in Oslo, Norway. Embryos were collected 1hpf and placed in embryo media until the time of exposure.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the embryos were acclimated prior to testing.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	There was one embryo per well in the 96-well plate. There were 16 replicates per test concentration with a total of 96 embryos per group.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	The embryo tests were conducted at 28C within constant darkness. The spawning adults that the embryos were obtained from were kept at 28C as well with a 14L:10D photoperiod. They were fed 3 x a day, 2 x with artemia and once with dry food.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest–changes in behavior due to TBBPA exposure.
	Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. Larval behavior was assessed at two times for TBBPA, 96 and 100hpf using ViewPoint Zebrabox and the accompanying video tracking software. Larval behavior monitored included the cumulative distance traveled and time spent active per minute during a light-dark-light cycle that lasted for 40 min.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. It was not reported if any acclimation occurred.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	All data were transferred to R version 2.15.0 for behavioral analysis. Data was checked for normality using visual examination, and linear mixed effect models were used for all analyses. More information is provided in section 2.6.
	Metric 22:	Reporting of Data	High	Control and exposure responses for the all dark treatments were provided in Fig 1C and were adequate for the outcome of interest.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes. Variability was reported in Fig 1.

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Study Citation:	Fraser, K., T.W., Khezri, A., Jusdado, H., J.G., Lewandowska-Sabat, A. M., Henry, T., Ropstad, E. (2017). Toxicant induced behavioural aberrations in larval zebrafish are dependent on minor methodological alterations. Toxicology Letters 276:62-68.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wildtype; Embryo
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5549558

Domain	Metric	Rating	Comments
Additional Comments:	This evaluation was on the effect of TBBPA on zebrafish larval behavior after 96hpf or 100hpf. 6-7hpf embryos were used at test initiation. The embryos used in this portion of the study had a photo-regime of constant darkness (plates wrapped in aluminum foil). This would be compared to a photo-regime of 14L:10D. The outcome of interest was the behavioral outcome.		

Overall Quality Determination Medium

Study Citation:	Godfrey, A., Hooser, B., Abdelmoneim, A., Horzmann, K. A., Freemanc, J. L., Sepúlveda, M. S. (2017). Thyroid disrupting effects of halogenated and next generation chemicals on the swim bladder development of zebrafish. Aquatic Toxicology 193:228-235.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wildtype; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4167836

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The chemical was identified by name and CAS#.
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Chemical purity and grade were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	Medium	The biological response of the negative control group was reported and reasonable for reported outcomes, but survival or health outcomes were not reported.
	Metric 6:	Randomized Allocation	Medium	The study reported that embryos were randomly allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured or measurements were not reported.
	Metric 10:	Exposure Duration and Frequency	Medium	The 6d exposure may have needed to be longer because presence of maternal thyroid hormones could have dampened effects.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	There was only one exposure group per chemical.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The scientific name of the species was not given in the paper.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects. Each petri dish contained 20 embryos and each test consisted of a minimum ofthree replicates per dose with experiments repeated three times.
Domain 5: Outcome Assessment				

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Study Citation:	Godfrey, A., Hooser, B., Abdelmoneim, A., Horzmann, K. A., Freemanc, J. L., Sepúlveda, M. S. (2017). Thyroid disrupting effects of halogenated and next generation chemicals on the swim bladder development of zebrafish. Aquatic Toxicology 193:228-235.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wildtype; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4167836

Domain	Metric		Rating	Comments
	Metric 16:	Adequacy of Test Conditions	High	Organism housing, environmental conditions, food, water, and nutrients were conducive to maintenance of health, and biomass loading was appropriate.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest (section 2.3).
	Metric 18:	Consistency of Outcome Assessment	High	The outcome assessment was consistent across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described. BioRad CFX 2.1 software was used for real-time qPCR data acquisition andanalysis. Differences in gene expression were analyzed using the delta Ct method.
	Metric 22:	Reporting of Data	High	Gene expression data was shown in Figure 4F. It was reported that TBBPA caused no changes in gene expression.
	Metric 23:	Explanation of Unexpected Outcomes	High	Outcomes were satisfactorily explained. Measures of variability were shown in Figure 4F.

Additional Comments: None

Overall Quality Determination High

Study Citation:	Li, L., Bonneton, F., Tohme, M., Bernard, L., Chen, X. Y., Laudet, V. (2016). In Vivo Screening Using Transgenic Zebrafish Embryos Reveals New Effects of HDAC Inhibitors Trichostatin A and Valproic Acid on Organogenesis. PLoS ONE 11(2):e0149497.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; Insulin-GFP, Fili1-EGFP, Elastase A-EGFP,LFABP-EGFP, Dlx2b-EGFP, a1-tubulin-EGFP, Shh-GFP, Brn3c-mGFP; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5500466

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical identified by name and CASRN.
	Metric 2:	Test Substance Source	High	Chemical purchased from Sigma Aldrich.
	Metric 3:	Test Substance Purity	Low	Purity not stated.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Control made with solvent (DMSO) not exceeding the highest amount used in chemical treatment (0.01%).
	Metric 5:	Negative Control Response	Medium	Images of control embryos for zebrafish lines shown in Figures 1-6.
	Metric 6:	Randomized Allocation	Low	Random allocation not stated.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	TBBPA solubility low (~4 mg/L), minimal description of how chemicals in DMSO were prepared in E3 medium. Unclear if serial dilutions of chemical in E3 were utilized or whether all chemical treatments in E3 had the same percentage of DMSO.
	Metric 8:	Consistency of Exposure Administration	High	Chemical exposure appears to be consistent in beginning 5 hpf in each zebrafish line.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations not measured in 1-5 day static non-renewal system. Would have some concern regarding sorption to cell walls if plastic microtiter plate was utilized. Dilutions prepared in DMSO and further diluted in E3 medium.
	Metric 10:	Exposure Duration and Frequency	High	Exposure durations were dependent on the zebrafish line and typical of exposure times in zf transgenic lines for assessing impact on organogenesis.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	Number of exposure groups not stated. Unclear what doses were utilized for TBBPA dilution (authors mention various dilutions of 1E-3 uM to 10 uM chemical).
	Metric 12:	Testing at or Below Solubility Limit	Medium	Highest concentration (10 uM, ~5 mg/L) exceeds approximate water solubility of 4 mg/L. DMSO was utilized to increase solubility.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	Characterization references for transgenic zebrafish lines cited in Table 1, authors did not explicitly state that they obtained the zebrafish lines from the respective laboratory.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	Acclimatization not reported.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	30 embryos per well for a chemical concentration. No replicate wells reported within an experiment. Experiments were performed thrice.

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Study Citation:	Li, L., Bonneton, F., Tohme, M., Bernard, L., Chen, X. Y., Laudet, V. (2016). In Vivo Screening Using Transgenic Zebrafish Embryos Reveals New Effects of HDAC Inhibitors Trichostatin A and Valproic Acid on Organogenesis. PLoS ONE 11(2):e0149497.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; Insulin-GFP, Fili1-EGFP, Elastase A-EGFP,LFABP-EGFP, Dlx2b-EGFP, a1-tubulin-EGFP, Shh-GFP, Brn3c-mGFP; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5500466

Domain	Metric		Rating	Comments
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Biomass loading seems inappropriate in this study (30 embryos per well in 6 well plate with up to 5 day exposure). Dead embryos removed but not reported at what intervals and mortality not reported.
	Metric 17:	Outcome Assessment Methodology	High	GFP imaging of live embryos described well. Chorion removed at 24 hpf to increase visualization of GFP.
	Metric 18:	Consistency of Outcome Assessment	Low	Unclear how many time points or which time points were assessed for TBBPA in most of the zebrafish lines as Figures in paper describe treatments with other chemicals and negative findings not shown in Figures (in the case of three zebrafish lines for all chemical treatments).
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Medium	Fish imaged at various time points during exposure period. Unlikely to have substantial impact on results.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	N/A	Results were qualitative. Effects in embryos were assessed visually by authors and reported as positive if effects observed in >70% of the embryos.
	Metric 22:	Reporting of Data	Low	Negative effects (no effects, shown as minuses in Table 2) from TBBPA treatment were not shown in Figures nor described in text. Results are also reported in Fig S1.
	Metric 23:	Explanation of Unexpected Outcomes	Low	Insufficient information was provided to determine if excessive variability or unexpected outcomes occurred.

Additional Comments: This form applies to the transgenic zebrafish lines. No effects in organogenesis from TBBPA treatment was observed for most of the cell lines, however, details of exact time points at which effects were measured (i.e. when the images were taken) is lacking for these zebrafish lines (a range in provided in Table 1 and example time points are given for other chemicals included in this study). Additionally, its unclear the exact TBBPA concentrations and number of treatment groups utilized in the dilution series (a concentration range is provided by authors, however). Fig S1 provides effects of TBBPA at 2.5uM.

Overall Quality Determination — Low

Study Citation:	Mccormick, J. M., Paiva, M. S., Häggblom, M. M., Cooper, K. R., White, L. A. (2010). Embryonic exposure to tetrabromobisphenol A and its metabolites, bisphenol A and tetrabromobisphenol A dimethyl ether disrupts normal zebrafish (Danio rerio) development and matrix metalloproteinase expression. Aquatic Toxicology 100(3):255-262.
Duration:	Overall Duration: > 21 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB Strain; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1294687; Linked HERO ID(s): 5489020, 1294687

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 >98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent solvent control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was suitable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health

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Study Citation:	Mccormick, J. M., Paiva, M. S., Häggblom, M. M., Cooper, K. R., White, L. A. (2010). Embryonic exposure to tetrabromobisphenol A and its metabolites, bisphenol A and tetrabromobisphenol A dimethyl ether disrupts normal zebrafish (Danio rerio) development and matrix metalloproteinase expression. Aquatic Toxicology 100(3):255-262.
Duration:	Overall Duration: > 21 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB Strain; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1294687; Linked HERO ID(s): 5489020, 1294687

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	Medium	details of reported environmental conditions were not sufficient to evaluate if adequate
	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 clearly described
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: Edema, hemorrhage, malformation, hatch outcomes for the 28d study duration with 5d exposure.

Overall Quality Determination High

Study Citation:	Mccormick, J. M., Paiva, M. S., Häggblom, M. M., Cooper, K. R., White, L. A. (2010). Embryonic exposure to tetrabromobisphenol A and its metabolites, bisphenol A and tetrabromobisphenol A dimethyl ether disrupts normal zebrafish (Danio rerio) development and matrix metalloproteinase expression. Aquatic Toxicology 100(3):255-262.
Duration:	Overall Duration: > 21 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB Strain; Embryo
Health Outcome:	Cardiovascular
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1294687; Linked HERO ID(s): 5489020, 1294687

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 >98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent solvent control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was suitable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of 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:	Mccormick, J. M., Paiva, M. S., Häggblom, M. M., Cooper, K. R., White, L. A. (2010). Embryonic exposure to tetrabromobisphenol A and its metabolites, bisphenol A and tetrabromobisphenol A dimethyl ether disrupts normal zebrafish (Danio rerio) development and matrix metalloproteinase expression. Aquatic Toxicology 100(3):255-262.
Duration:	Overall Duration: > 21 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB Strain; Embryo
Health Outcome:	Cardiovascular
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1294687; Linked HERO ID(s): 5489020, 1294687

Domain		Metric	Rating	Comments
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Medium	details of reported environmental conditions were not sufficient to evaluate if adequate
	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 clearly described
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: Heart rate responses for 28d study with 5d exposure.

Overall Quality Determination **High**

Study Citation:	Mccormick, J. M., Paiva, M. S., Häggblom, M. M., Cooper, K. R., White, L. A. (2010). Embryonic exposure to tetrabromobisphenol A and its metabolites, bisphenol A and tetrabromobisphenol A dimethyl ether disrupts normal zebrafish (Danio rerio) development and matrix metalloproteinase expression. Aquatic Toxicology 100(3):255-262.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB Strain; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1294687; Linked HERO ID(s): 5489020, 1294687

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 >98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent solvent control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was suitable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of 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:	Mccormick, J. M., Paiva, M. S., Häggblom, M. M., Cooper, K. R., White, L. A. (2010). Embryonic exposure to tetrabromobisphenol A and its metabolites, bisphenol A and tetrabromobisphenol A dimethyl ether disrupts normal zebrafish (Danio rerio) development and matrix metalloproteinase expression. Aquatic Toxicology 100(3):255-262.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB Strain; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1294687; Linked HERO ID(s): 5489020, 1294687

Domain	Metric		Rating	Comments
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Medium	details of reported environmental conditions were not sufficient to evaluate if adequate
	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 clearly described
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: edema, hemorrhage, malformation, hatch

Overall Quality Determination High

Study Citation:	Mccormick, J. M., Paiva, M. S., Häggblom, M. M., Cooper, K. R., White, L. A. (2010). Embryonic exposure to tetrabromobisphenol A and its metabolites, bisphenol A and tetrabromobisphenol A dimethyl ether disrupts normal zebrafish (Danio rerio) development and matrix metalloproteinase expression. Aquatic Toxicology 100(3):255-262.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB Strain; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1294687; Linked HERO ID(s): 5489020, 1294687

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 >98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent solvent control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was suitable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of 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:	Mccormick, J. M., Paiva, M. S., Häggblom, M. M., Cooper, K. R., White, L. A. (2010). Embryonic exposure to tetrabromobisphenol A and its metabolites, bisphenol A and tetrabromobisphenol A dimethyl ether disrupts normal zebrafish (Danio rerio) development and matrix metalloproteinase expression. Aquatic Toxicology 100(3):255-262.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB Strain; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1294687; Linked HERO ID(s): 5489020, 1294687

Domain	Metric		Rating	Comments
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Medium	details of reported environmental conditions were not sufficient to evaluate if adequate
	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 clearly described
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: None

Overall Quality Determination High

Study Citation:	Mccormick, J. M., Paiva, M. S., Häggblom, M. M., Cooper, K. R., White, L. A. (2010). Embryonic exposure to tetrabromobisphenol A and its metabolites, bisphenol A and tetrabromobisphenol A dimethyl ether disrupts normal zebrafish (Danio rerio) development and matrix metalloproteinase expression. Aquatic Toxicology 100(3):255-262.
Duration:	Overall Duration: > 21 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB Strain; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1294687; Linked HERO ID(s): 5489020, 1294687

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 >98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent solvent control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was suitable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of 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:	Mccormick, J. M., Paiva, M. S., Häggblom, M. M., Cooper, K. R., White, L. A. (2010). Embryonic exposure to tetrabromobisphenol A and its metabolites, bisphenol A and tetrabromobisphenol A dimethyl ether disrupts normal zebrafish (Danio rerio) development and matrix metalloproteinase expression. Aquatic Toxicology 100(3):255-262.
Duration:	Overall Duration: > 21 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB Strain; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1294687; Linked HERO ID(s): 5489020, 1294687

Domain	Metric		Rating	Comments
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Medium	details of reported environmental conditions were not sufficient to evaluate if adequate
	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 clearly described
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: Mortality outcome for 28d study with 5d exposure.

Overall Quality Determination High

Study Citation:	Noyes, P. D., Haggard, D. E., Gonnerman, G. D., Tanguay, R. L. (2015). Advanced morphological - behavioral test platform reveals neurodevelopmental defects in embryonic zebrafish exposed to comprehensive suite of halogenated and organophosphate flame retardants. Toxicological Sciences 145(1):177-195.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; wild type (Tropical 5D); Embryo
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2953504

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The chemical is referred to by name, structure, and CASRN.
	Metric 2:	Test Substance Source	Low	Source was reported but the authors did not perform analytical verification.
	Metric 3:	Test Substance Purity	High	Purity is >98% and reported in Table 3.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	A solvent control (DMSO at 0.64%) was used.
	Metric 5:	Negative Control Response	High	All control responses are reported in the supplemental data PDF.
	Metric 6:	Randomized Allocation	Medium	Although not specifically reported as "random" the process of chorion removal and distribution into well plates was automated and is detailed in: "Mandrell, David, Lisa Truong, Caleb Jephson, Mushfiqur R. Sarker, Aaron Moore, Christopher Lang, Michael T. Simonich, and Robert L. Tanguay. "Automated zebrafish chorion removal and single embryo placement: optimizing throughput of zebrafish developmental toxicity screens." Journal of laboratory automation 17, no. 1 (2012): 66-74."
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The exposures were static non-renewal from 6 to 120 hpf. Renewal of compounds would have been preferred.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were conducted consistently among treatments and control.
	Metric 9:	Measurement of Test Substance Concentration	Low	No verification was performed, concentrations are reported as nominal.
	Metric 10:	Exposure Duration and Frequency	High	Exposure was initiated at 6 hpf and continued till 120 hpf. The assessments at 24 and 120 hpf are appropriate to capture embryo and larval periods.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Concentrations were: 64, 6.4, 0.64, 0.064, 0.0064, and 0 uM and represent a broad range of exposure concentrations.
	Metric 12:	Testing at or Below Solubility Limit	Medium	The highest concentration (64 uM) is above the solubility limit. The remaining four concentrations are below solubility. (solubility from final scopes for TBBPA, TPP, and TCEP are 4.1, 1.9 and 7.8 mg/L)
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The strain and source for broodfish was reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	Water quality parameters between broodfish and embryo/larvae were not reported.

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Study Citation:	Noyes, P. D., Haggard, D. E., Gonnerman, G. D., Tanguay, R. L. (2015). Advanced morphological - behavioral test platform reveals neurodevelopmental defects in embryonic zebrafish exposed to comprehensive suite of halogenated and organophosphate flame retardants. Toxicological Sciences 145(1):177-195.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; wild type (Tropical 5D); Embryo
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2953504

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Medium	All sample size numbers for treatment and control groups are listed in the first two pages of the supplemental data PDF for this work.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Photoperiod and temperature were reported, while other water quality conditions were not.
	Metric 17:	Outcome Assessment Methodology	Medium	Movement assays for embryos (24 hpf) and larvae (120 hpf) were described in detail on page 7 of 19.
	Metric 18:	Consistency of Outcome Assessment	High	Assessment was consistent among treatments and control.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	Nothing was reported to indicate differences in study groups based on environmental factors.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	No information in the study to indicate that differences were from animal attrition or health outcomes.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Embryo movement: ”Specifically, overall patterns of activity within each cycle interval (ie, baseline, excitation, refractory) were compared with those in vehicle controls by (1) estimating the 50% peak difference from controls in either direction and (2) performing a Kolmogorov–Smirnov test that compared the empirical cumulative distribution function between chemical treatments and controls. A Bonferroni-corrected p-value threshold of .01 (0.05/5 treatments¼.01) was used to determine statistical significance.”Larval Movement: ”As larval activity did not meet parametric assumptions of normality, Kruskal–Wallis analyses of variance and Dunn’s multiple comparison post tests were used to compare median locomotor activity per minute in treatment versus controls in each of the 5-min light/dark phases.”
	Metric 22:	Reporting of Data	High	All raw data across time for embryo and larval movement is reported in the supplemental notes (24 and 120 hpf).
	Metric 23:	Explanation of Unexpected Outcomes	High	No unexpected outcomes were reported by the authors.

Additional Comments: This form is for embryo movement assessment at 120 hpf for TBBPA.

Overall Quality Determination High

Study Citation:	Noyes, P. D., Haggard, D. E., Gonnerman, G. D., Tanguay, R. L. (2015). Advanced morphological - behavioral test platform reveals neurodevelopmental defects in embryonic zebrafish exposed to comprehensive suite of halogenated and organophosphate flame retardants. Toxicological Sciences 145(1):177-195.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; wild type (Tropical 5D); Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2953504

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The chemical is referred to by name, structure, and CASRN.
	Metric 2:	Test Substance Source	Low	Source was reported but the authors did not perform analytical verification.
	Metric 3:	Test Substance Purity	High	Purity is >98% and reported in Table 3.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	A solvent control (DMSO at 0.64%) was used.
	Metric 5:	Negative Control Response	High	All control responses are reported in the supplemental data PDF.
	Metric 6:	Randomized Allocation	Medium	Although not specifically reported as "random" the process of chorion removal and distribution into well plates was automated and is detailed in: "Mandrell, David, Lisa Truong, Caleb Jephson, Mushfiqur R. Sarker, Aaron Moore, Christopher Lang, Michael T. Simonich, and Robert L. Tanguay. "Automated zebrafish chorion removal and single embryo placement: optimizing throughput of zebrafish developmental toxicity screens." Journal of laboratory automation 17, no. 1 (2012): 66-74."
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The exposures were static non-renewal from 6 to 120 hpf. Renewal of compounds would have been preferred.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were conducted consistently among treatments and control.
	Metric 9:	Measurement of Test Substance Concentration	Low	No verification was performed, concentrations are reported as nominal.
	Metric 10:	Exposure Duration and Frequency	High	Exposure was initiated at 6 hpf and continued till 120 hpf. The assessments at 24 and 120 hpf are appropriate to capture embryo and larval periods. 24 hour developmental assessments included: delays in developmental progression, notochord deformities, and altered spontaneous movements. 120 hour developmental assessments included: 17 developmental malformations, including yolk sac edema (YSE) and pericardial edema (PE); body axis (AXIS), trunk length (TRUN), caudal fin (CFIN), pectoral fin (PFIN), pigmentation (PIG), and somite (SOMI) deformities; eye (EYE), snout (SNOU), jaw (JAW), and otolith (OTIC) malformations; gross brain development (BRAIN); notochord (NC) and circulatory (CIRC) deformities; swim bladder presence and inflation (SWIM); and touch responses (TR).
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Concentrations were: 64, 6.4, 0.64, 0.064, 0.0064, and 0 uM and represent a broad range of exposure concentrations.

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Study Citation:	Noyes, P. D., Haggard, D. E., Gonnerman, G. D., Tanguay, R. L. (2015). Advanced morphological - behavioral test platform reveals neurodevelopmental defects in embryonic zebrafish exposed to comprehensive suite of halogenated and organophosphate flame retardants. Toxicological Sciences 145(1):177-195.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; wild type (Tropical 5D); Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2953504

Domain	Metric		Rating	Comments
	Metric 12:	Testing at or Below Solubility Limit	Medium	The highest concentration (64 uM) is above the solubility limit. The remaining four concentrations are below solubility. (solubility from final scopes for TBBPA, TPP, and TCEP are 4.1, 1.9 and 7.8 mg/L)
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The strain and source for broodfish was reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	Water quality parameters between broodfish and embryo/larvae were not reported.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	All sample size numbers for treatment and control groups are listed in the first two pages of the supplemental data PDF for this work.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Photoperiod and temperature were reported, while other water quality conditions were not.
	Metric 17:	Outcome Assessment Methodology	Medium	Developmental assessment was described in the section titled ”Developmental malformation evaluations”. The specifc analysis is detailed in another publication that authors referenced as HERO ID 8591199
	Metric 18:	Consistency of Outcome Assessment	High	Assessment was consistent among treatments and control.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	Nothing was reported to indicate differences in study groups based on environmental factors.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	No information in the study to indicate that differences were from animal attrition or health outcomes.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	”Data collection was undertaken using a custom barcoding and tracking system (Zebrafish Acquisition and Analysis Program) to facilitate reliable management of the large amounts of data collected. Statistical analyses were performed using R code with testing methodologies used by Truong et al. (2014) to evaluate developmental toxicity of chemicals under the ToxCast program(RCoreTeam, 2014; Truong et al., 2014). Briefly, a binomial testwas performed that calculated lowest effect levels (LELs) foreach endpoint to identify incidences that exceeded a significantthreshold above controls. This test was preferable to a logisticregression as it accounted for the observed nonmonotonicity offlame retardant toxicity.”This reference is HERO ID 8591199

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Study Citation:	Noyes, P. D., Haggard, D. E., Gonnerman, G. D., Tanguay, R. L. (2015). Advanced morphological - behavioral test platform reveals neurodevelopmental defects in embryonic zebrafish exposed to comprehensive suite of halogenated and organophosphate flame retardants. Toxicological Sciences 145(1):177-195.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; wild type (Tropical 5D); Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2953504

Domain	Metric		Rating	Comments
	Metric 22:	Reporting of Data	High	All counts of data per treatment and control group are represented in the supplemental data for each compound and time point (24 and 120 hpf).
	Metric 23:	Explanation of Unexpected Outcomes	High	No unexpected outcomes were reported by the authors.

Additional Comments: This form is for developmental assessment at 120 hpf for TBBPA. The specific data and statistical significance for each compound are located within the supplemental PDF.

Overall Quality Determination High

Study Citation:	Noyes, P. D., Haggard, D. E., Gonnerman, G. D., Tanguay, R. L. (2015). Advanced morphological - behavioral test platform reveals neurodevelopmental defects in embryonic zebrafish exposed to comprehensive suite of halogenated and organophosphate flame retardants. Toxicological Sciences 145(1):177-195.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; wild type (Tropical 5D); Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2953504

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The chemical is referred to by name, structure, and CASRN.
	Metric 2:	Test Substance Source	Low	Source was reported but the authors did not perform analytical verification.
	Metric 3:	Test Substance Purity	High	Purity is >98% and reported in Table 3.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	A solvent control (DMSO at 0.64%) was used.
	Metric 5:	Negative Control Response	High	All control responses are reported in the supplemental data PDF.
	Metric 6:	Randomized Allocation	Medium	Although not specifically reported as "random" the process of chorion removal and distribution into well plates was automated and is detailed in: "Mandrell, David, Lisa Truong, Caleb Jephson, Mushfiqur R. Sarker, Aaron Moore, Christopher Lang, Michael T. Simonich, and Robert L. Tanguay. "Automated zebrafish chorion removal and single embryo placement: optimizing throughput of zebrafish developmental toxicity screens." Journal of laboratory automation 17, no. 1 (2012): 66-74."
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The exposures were static non-renewal from 6 to 120 hpf. Renewal of compounds would have been preferred.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were conducted consistently among treatments and control.
	Metric 9:	Measurement of Test Substance Concentration	Low	No verification was performed, concentrations are reported as nominal.
	Metric 10:	Exposure Duration and Frequency	High	Exposure was initiated at 6 hpf and continued till 120 hpf. The assessments at 24 and 120 hpf are appropriate to capture embryo and larval periods.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Concentrations were: 64, 6.4, 0.64, 0.064, 0.0064, and 0 uM and represent a broad range of exposure concentrations.
	Metric 12:	Testing at or Below Solubility Limit	Medium	The highest concentration (64 uM) is above the solubility limit. The remaining four concentrations are below solubility. (solubility from final scopes for TBBPA, TPP, and TCEP are 4.1, 1.9 and 7.8 mg/L)
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The strain and source for broodfish was reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	Water quality parameters between broodfish and embryo/larvae were not reported.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	All sample size numbers for treatment and control groups are listed in the first two pages of the supplemental data PDF for this work.

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Study Citation:	Noyes, P. D., Haggard, D. E., Gonnerman, G. D., Tanguay, R. L. (2015). Advanced morphological - behavioral test platform reveals neurodevelopmental defects in embryonic zebrafish exposed to comprehensive suite of halogenated and organophosphate flame retardants. Toxicological Sciences 145(1):177-195.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; wild type (Tropical 5D); Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2953504

Domain		Metric	Rating	Comments
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Photoperiod and temperature were reported, while other water quality conditions were not.
	Metric 17:	Outcome Assessment Methodology	Medium	Mortality assessment was described in the section titled ”Developmental malformation evaluations” but authors did not report specific criteria for death (ie, movement, heart-beat, color, etc.).
	Metric 18:	Consistency of Outcome Assessment	High	Assessment was consistent among treatments and control.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	Nothing was reported to indicate differences in study groups based on environmental factors.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	No information in the study to indicate that differences were from animal attrition or health outcomes.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	”Data collection was undertaken using a custom barcoding and tracking system (Zebrafish Acquisition and Analysis Program) to facilitate reliable management of the large amounts of data collected. Statistical analyses were performed using R code with testing methodologies used by Truong et al. (2014) to evaluate developmental toxicity of chemicals under the ToxCast program(RCoreTeam, 2014; Truong et al., 2014). Briefly, a binomial testwas performed that calculated lowest effect levels (LELs) foreach endpoint to identify incidences that exceeded a significantthreshold above controls. This test was preferable to a logisticregression as it accounted for the observed nonmonotonicity offlame retardant toxicity.”
	Metric 22:	Reporting of Data	High	All counts of data per treatment and control group are represented in the supplemental data for each compound and time point (24 and 120 hpf).
	Metric 23:	Explanation of Unexpected Outcomes	High	No unexpected outcomes were reported by the authors.

Additional Comments: This form is for Mortality assessment at 120 hpf for TBBPA. The specific data and statistical significance for each compound are located within the supplemental PDF.

Overall Quality Determination High

Study Citation:	Parsons, A., Lange, A., Hutchinson, T. H., Miyagawa, S., Iguchi, T., Kudoh, T., Tyler, C. R. (2019). Molecular mechanisms and tissue targets of brominated flame retardants, BDE-47 and TBBPA, in embryo-larval life stages of zebrafish (Danio rerio). Aquatic Toxicology 209:99-112.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; casper(mitfa;roy) mutant strain; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Receptor binding/ regulation of receptor activity
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5496817

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#.
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 97%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported via graph and was reasonable.
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided few details on exposure media preparation.
	Metric 8:	Consistency of Exposure Administration	Medium	No mention of irregularities in exposure administration.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	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	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reported environmental conditions were not sufficient to evaluate if adequate.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.

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Study Citation:	Parsons, A., Lange, A., Hutchinson, T. H., Miyagawa, S., Iguchi, T., Kudoh, T., Tyler, C. R. (2019). Molecular mechanisms and tissue targets of brominated flame retardants, BDE-47 and TBBPA, in embryo-larval life stages of zebrafish (Danio rerio). Aquatic Toxicology 209:99-112.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; casper(mitfa;roy) mutant strain; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Receptor binding/ regulation of receptor activity
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5496817

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There were no information presented 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: Several Genes were examined with qPCR and visualized with WISH from 0-48, 0-96, 0-120 hpf. The experimental design for these are presented in Figure 1 (page 4/14).

Overall Quality Determination High

Study Citation:	Song, M., Liang, D., Liang, Y., Chen, M., Wang, F., Wang, H., Jiang, G. (2014). Assessing developmental toxicity and estrogenic activity of halogenated bisphenol A on zebrafish (Danio rerio). Chemosphere 112:275-281.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2754370

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 >=98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control groups
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was 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 for making a stock solution
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The test organisms were adequately described unsure where they were obtained from
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest

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Study Citation:	Song, M., Liang, D., Liang, Y., Chen, M., Wang, F., Wang, H., Jiang, G. (2014). Assessing developmental toxicity and estrogenic activity of halogenated bisphenol A on zebrafish (Danio rerio). Chemosphere 112:275-281.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2754370

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
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis (LC50) was performed but not described adequately
	Metric 22:	Reporting of Data	Low	Data for mortality was not shown for control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: None

Overall Quality Determination High

Study Citation:	Song, M., Liang, D., Liang, Y., Chen, M., Wang, F., Wang, H., Jiang, G. (2014). Assessing developmental toxicity and estrogenic activity of halogenated bisphenol A on zebrafish (Danio rerio). Chemosphere 112:275-281.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2754370

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 >=98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control groups
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was 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 for making a stock solution
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The test organisms were adequately described unsure where they were obtained from
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups

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Study Citation:	Song, M., Liang, D., Liang, Y., Chen, M., Wang, F., Wang, H., Jiang, G. (2014). Assessing developmental toxicity and estrogenic activity of halogenated bisphenol A on zebrafish (Danio rerio). Chemosphere 112:275-281.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2754370

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
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Unclear if statistical analysis was performed b
	Metric 22:	Reporting of Data	Low	Data for hatch was not shown for control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: Hatch

Overall Quality Determination High

Study Citation:	Truong, L., Marvel, S., Reif, D. M., Thomas, D. G., Pande, P., Dasgupta, S., Simonich, M. T., Waters, K. M., Tanguay, R. L. (2020). The multi-dimensional embryonic zebrafish platform predicts flame retardant bioactivity. Reproductive toxicology (Elmsford, N.Y.) 96:359-369.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; Tropical 5D wild type; Embryo
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	13035613

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The TBBPA was identified by CASRN in Supplemental Table S1A.
	Metric 2:	Test Substance Source	High	The source of the TBBPA was reported to be Sigma in the supplemental material. The test substance was reported to have a certificate of analysis.
	Metric 3:	Test Substance Purity	High	The purity was reported to be >98%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of a concurrent negative control.
	Metric 5:	Negative Control Response	High	The negative control response is reported in the supplemental material.
	Metric 6:	Randomized Allocation	Low	It was not reported how the embryos were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The tests were conducted in 96-well plates with 100uL of embryo medium. The TBBPA was added to each well using the Hewlett-Packard D300 Digital Dispenser as a stock solution. The preparation of the stock solution was not reported in detail. The plates were sealed with Parafilm beneath the lid to reduce evaporation. They were also wrapped in aluminum foil to prevent photodegradation.
	Metric 8:	Consistency of Exposure Administration	High	The exposure were all conducted in 96-well plates with 100uL of embryo medium with one embryo per well.
	Metric 9:	Measurement of Test Substance Concentration	Low	It was not reported if the test substance was measured at any point in the study.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration was reported to be to 120hpf. Embryos were exposed when they were 6hpf. Mortality and behavioral outcomes were assessed at 24hpf and at 120hpf. This appeared adequate for the outcomes of interest.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	There were five exposure levels in addition to a control ranging from 0.1uM to 25uM. The spacing was adequate to observe the outcomes of interest.
	Metric 12:	Testing at or Below Solubility Limit	Medium	The highest test concentration was above the water solubility limit, but the rest of the test concentrations remained below the water solubility limit. It was reported the solvent DMSO was used in all wells to aid solubility with a final concentration of 0.64%.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	It was reported that tropical 5D wild type zebrafish were housed at the Oregon State University Sinnhuber Aquatic Laboratory. It is unclear if they were cultured in-house or from another source. 6 hour post fertilization embryos were used to initiate the study.

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Study Citation:	Truong, L., Marvel, S., Reif, D. M., Thomas, D. G., Pande, P., Dasgupta, S., Simonich, M. T., Waters, K. M., Tanguay, R. L. (2020). The multi-dimensional embryonic zebrafish platform predicts flame retardant bioactivity. Reproductive toxicology (Elmsford, N.Y.) 96:359-369.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; Tropical 5D wild type; Embryo
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	13035613

Domain	Metric		Rating	Comments
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the organisms were acclimated at all prior to the test.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	There was one embryo per well. It appears there were 32 embryos per test concentration in the supplemental data.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	The zebrafish were housed in 100-gallon tanks in a density of 1000 fish per tank. Each tank was maintained at 28C on a 14L:10D photoperiod. Feeding regimen and water characteristics were not reported. It is unclear if the test conditions were the same as the housing conditions.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcomes of interest-changes in embryonic tail flexions in response to visible light at 24hpf using the embryonic photomotor response assay (EPR) and changes in larval locomotor response using the larval photomotor response (LPR) assay at 120hpf.
	Metric 18:	Consistency of Outcome Assessment	High	Both the EPR and LPR were carried out consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. Little information was provided regarding testing conditions or possible acclimation.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were reported in section 2.6 and were appropriate for the outcomes of interest.
	Metric 22:	Reporting of Data	High	Both responses to the EPR and LPR are reported in the supplemental material for the treatment groups and the controls. LEL and BMD values are reported in supplemental tables S1A and S1B.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.

Additional Comments: This evaluation is for the embryonic and larval photomotor response assays. Behavior changes to light were assessed after exposure to five different TBBPA test concentrations at 24hpf and 120hpf.

Overall Quality Determination High

Study Citation:	Truong, L., Marvel, S., Reif, D. M., Thomas, D. G., Pande, P., Dasgupta, S., Simonich, M. T., Waters, K. M., Tanguay, R. L. (2020). The multi-dimensional embryonic zebrafish platform predicts flame retardant bioactivity. Reproductive toxicology (Elmsford, N.Y.) 96:359-369.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; Tropical 5D wild type; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	13035613

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The TBBPA was identified by CASRN in Supplemental Table S1A.
	Metric 2:	Test Substance Source	High	The source of the TBBPA was reported to be Sigma in the supplemental material. The test substance was reported to have a certificate of analysis.
	Metric 3:	Test Substance Purity	High	The purity was reported to be >98%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of a concurrent negative control.
	Metric 5:	Negative Control Response	High	The negative control response is reported in the supplemental material.
	Metric 6:	Randomized Allocation	Low	It was not reported how the embryos were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The tests were conducted in 96-well plates with 100uL of embryo medium. The TBBPA was added to each well using the Hewlett-Packard D300 Digital Dispenser as a stock solution. The preparation of the stock solution was not reported in detail. The plates were sealed with Parafilm beneath the lid to reduce evaporation. They were also wrapped in aluminum foil to prevent photodegradation.
	Metric 8:	Consistency of Exposure Administration	High	The exposure were all conducted in 96-well plates with 100uL of embryo medium with one embryo per well.
	Metric 9:	Measurement of Test Substance Concentration	Low	It was not reported if the test substance was measured at any point in the study.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration was reported to be to 120hpf. Embryos were exposed when they were 6hpf. Mortality and behavioral outcomes were assessed at 24hpf and at 120hpf. This appeared adequate for the outcomes of interest. Developmental malformations were assessed at 120hpf.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	There were five exposure levels in addition to a control ranging from 0.1uM to 25uM. The spacing was adequate to observe the outcomes of interest.
	Metric 12:	Testing at or Below Solubility Limit	Medium	The highest test concentration was above the water solubility limit, but the rest of the test concentrations remained below the water solubility limit. It was reported the solvent DMSO was used in all wells to aid solubility with a final concentration of 0.64%.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	It was reported that tropical 5D wild type zebrafish were housed at the Oregon State University Sinnhuber Aquatic Laboratory. It is unclear if they were cultured in-house or from another source. 6 hour post fertilization embryos were used to initiate the study.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the organisms were acclimated at all prior to the test.

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Study Citation:	Truong, L., Marvel, S., Reif, D. M., Thomas, D. G., Pande, P., Dasgupta, S., Simonich, M. T., Waters, K. M., Tanguay, R. L. (2020). The multi-dimensional embryonic zebrafish platform predicts flame retardant bioactivity. Reproductive toxicology (Elmsford, N.Y.) 96:359-369.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; Tropical 5D wild type; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	13035613

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Medium	There was one embryo per well. It appears there were 32 embryos per test concentration in the supplemental data.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	The zebrafish were housed in 100-gallon tanks in a density of 1000 fish per tank. Each tank was maintained at 28C on a 14L:10D photoperiod. Feeding regimen and water characteristics were not reported. It is unclear if the test conditions were the same as the housing conditions.
	Metric 17:	Outcome Assessment Methodology	Low	It was reported that 18 developmental malformations were assessed at 120hpf. The methods used for assessing these malformations were not described.
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the assessment methodology were limited. It is unclear what the methods were for assessing the 18 developmental malformations.
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. Little information was provided regarding testing conditions or possible acclimation.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were reported in section 2.6 and were appropriate for the outcomes of interest.
	Metric 22:	Reporting of Data	High	Malformations for each test concentration and control were reported in the Supplemental Material. LEL and BMD values are reported in supplemental tables S1A and S1B.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.

Additional Comments: This evaluation is for the 18 developmental malformation outcomes assessed after embryonic zebrafish were exposed to five different test concentrations of TBBPA. The methods for assessing these malformations was not reported in great detail. Data is reported in the supplemental material.

Overall Quality Determination Medium

Study Citation:	Truong, L., Marvel, S., Reif, D. M., Thomas, D. G., Pande, P., Dasgupta, S., Simonich, M. T., Waters, K. M., Tanguay, R. L. (2020). The multi-dimensional embryonic zebrafish platform predicts flame retardant bioactivity. Reproductive toxicology (Elmsford, N.Y.) 96:359-369.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; Tropical 5D wild type; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	13035613

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The TBBPA was identified by CASRN in Supplemental Table S1A.
	Metric 2:	Test Substance Source	High	The source of the TBBPA was reported to be Sigma in the supplemental material. The test substance was reported to have a certificate of analysis.
	Metric 3:	Test Substance Purity	High	The purity was reported to be >98%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of a concurrent negative control.
	Metric 5:	Negative Control Response	High	The negative control response is reported in the supplemental material.
	Metric 6:	Randomized Allocation	Low	It was not reported how the embryos were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The tests were conducted in 96-well plates with 100uL of embryo medium. The TBBPA was added to each well using the Hewlett-Packard D300 Digital Dispenser as a stock solution. The preparation of the stock solution was not reported in detail. The plates were sealed with Parafilm beneath the lid to reduce evaporation. They were also wrapped in aluminum foil to prevent photodegradation.
	Metric 8:	Consistency of Exposure Administration	High	The exposure were all conducted in 96-well plates with 100uL of embryo medium with one embryo per well.
	Metric 9:	Measurement of Test Substance Concentration	Low	It was not reported if the test substance was measured at any point in the study.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration was reported to be to 120hpf. Embryos were exposed when they were 6hpf. Mortality and behavioral outcomes were assessed at 24hpf and at 120hpf. This appeared adequate for the outcomes of interest.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	There were five exposure levels in addition to a control ranging from 0.1uM to 25uM. The spacing was adequate to observe the outcomes of interest.
	Metric 12:	Testing at or Below Solubility Limit	Medium	The highest test concentration was above the water solubility limit, but the rest of the test concentrations remained below the water solubility limit. It was reported the solvent DMSO was used in all wells to aid solubility with a final concentration of 0.64%.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	It was reported that tropical 5D wild type zebrafish were housed at the Oregon State University Sinnhuber Aquatic Laboratory. It is unclear if they were cultured in-house or from another source. 6 hour post fertilization embryos were used to initiate the study.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the organisms were acclimated at all prior to the test.

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Study Citation:	Truong, L., Marvel, S., Reif, D. M., Thomas, D. G., Pande, P., Dasgupta, S., Simonich, M. T., Waters, K. M., Tanguay, R. L. (2020). The multi-dimensional embryonic zebrafish platform predicts flame retardant bioactivity. Reproductive toxicology (Elmsford, N.Y.) 96:359-369.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; Tropical 5D wild type; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	13035613

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Medium	There was one embryo per well. It appears there were 32 embryos per test concentration in the supplemental data.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	The zebrafish were housed in 100-gallon tanks in a density of 1000 fish per tank. Each tank was maintained at 28C on a 14L:10D photoperiod. Feeding regimen and water characteristics were not reported. It is unclear if the test conditions were the same as the housing conditions.
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology addressed or reported the intended outcomes of interest-mortality at 24hpf and 120hpf. The outcome assessment methodology for determining mortality was not reported. It was unclear how embryos or larvae were determined to be dead.
	Metric 18:	Consistency of Outcome Assessment	Low	Mortality was assessed at 24hpf and 120hpf, but the methods for determining mortality in embryos and larvae 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. Little information was provided regarding testing conditions or possible acclimation.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were reported in section 2.6 and were appropriate for the outcomes of interest.
	Metric 22:	Reporting of Data	High	Exposure and control responses for mortality were reported in the supplemental material. LEL and BMD values are reported in supplemental tables S1A and S1B.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.

Additional Comments: This evaluation is for the mortality outcomes assessed at 24hpf and 120hpf after exposure to five different test concentrations of TBBPA. Data for the mortality responses is reported in the supplemental material.

Overall Quality Determination Medium

Study Citation:	Usenko, C. Y., Abel, E. L., Hopkins, A., Martinez, G., Tijerina, J., Kudela, M., Norris, N., Joudeh, L., Bruce, E. D. (2016). Evaluation of Common Use Brominated Flame Retardant (BFR) Toxicity Using a Zebrafish Embryo Model.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; Embryo
Health Outcome:	Mechanistic-Cell signaling/function-Oxidative stress (including redox biology)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5554410

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	Medium	The biological responses of the negative control group(s) were reported but only via graph
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Medium	Reporting omissions are unlikely to have a substantial impact on results
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	Medium	The duration of exposure was reported and reasonably appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test animals was not reported
	Metric 14:	Acclimatization and Pretreatment Conditions	Medium	The study did not report whether pretreatment conditions were the same for control and exposed groups,
	Metric 15:	Number of Organisms and Replicates per Group	Medium	high number of replicates used
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest

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Study Citation:	Usenko, C. Y., Abel, E. L., Hopkins, A., Martinez, G., Tijerina, J., Kudela, M., Norris, N., Joudeh, L., Bruce, E. D. (2016). Evaluation of Common Use Brominated Flame Retardant (BFR) Toxicity Using a Zebrafish Embryo Model.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; Embryo
Health Outcome:	Mechanistic-Cell signaling/function-Oxidative stress (including redox biology)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5554410

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	Medium	outcomes were assessed consistently across study groups but few details were provided
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	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 via graph
	Metric 23:	Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained

Additional Comments: AChE (Figure 4) and GST response (figure 5) at 120 hpf

Overall Quality Determination Medium

Study Citation:	Usenko, C. Y., Abel, E. L., Hopkins, A., Martinez, G., Tijerina, J., Kudela, M., Norris, N., Joudeh, L., Bruce, E. D. (2016). Evaluation of Common Use Brominated Flame Retardant (BFR) Toxicity Using a Zebrafish Embryo Model.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5554410

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	Medium	The biological responses of the negative control group(s) were reported but only via graph
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Medium	Reporting omissions are unlikely to have a substantial impact on results
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	Medium	The duration of exposure was reported and reasonably appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test animals was not reported
	Metric 14:	Acclimatization and Pretreatment Conditions	Medium	The study did not report whether pretreatment conditions were the same for control and exposed groups,
	Metric 15:	Number of Organisms and Replicates per Group	Medium	A high number of replicates were used.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Environmental conditions were not sufficiently reported to evaluate if adequate.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	Medium	Outcomes were assessed consistently across study groups but few details were provided

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Study Citation:	Usenko, C. Y., Abel, E. L., Hopkins, A., Martinez, G., Tijerina, J., Kudela, M., Norris, N., Joudeh, L., Bruce, E. D. (2016). Evaluation of Common Use Brominated Flame Retardant (BFR) Toxicity Using a Zebrafish Embryo Model.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5554410

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 via graph
	Metric 23:	Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained

Additional Comments: Developmental malformations are presented in Figure 2, Page 6.

Overall Quality Determination Medium

Study Citation:	Usenko, C. Y., Abel, E. L., Hopkins, A., Martinez, G., Tijerina, J., Kudela, M., Norris, N., Joudeh, L., Bruce, E. D. (2016). Evaluation of Common Use Brominated Flame Retardant (BFR) Toxicity Using a Zebrafish Embryo Model.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; Embryo
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5554410

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	Medium	The biological responses of the negative control group(s) were reported but only via graph
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Medium	Reporting omissions are unlikely to have a substantial impact on results
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	Medium	The duration of exposure was reported and reasonably 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	Exposure concentrations above 5 mg/L are above the solubility limit for TBBPA.
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	Medium	The study did not report whether pretreatment conditions were the same for control and exposed groups,
	Metric 15:	Number of Organisms and Replicates per Group	Medium	high number of replicates used
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	Medium	outcomes were assessed consistently across study groups but few details were provided

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Study Citation:	Usenko, C. Y., Abel, E. L., Hopkins, A., Martinez, G., Tijerina, J., Kudela, M., Norris, N., Joudeh, L., Bruce, E. D. (2016). Evaluation of Common Use Brominated Flame Retardant (BFR) Toxicity Using a Zebrafish Embryo Model.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; Embryo
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5554410

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 via graph
	Metric 23:	Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained

Additional Comments: Tail movement is detailed on page 7 and in figure 3.

Overall Quality Determination Medium

Study Citation:	Usenko, C. Y., Abel, E. L., Hopkins, A., Martinez, G., Tijerina, J., Kudela, M., Norris, N., Joudeh, L., Bruce, E. D. (2016). Evaluation of Common Use Brominated Flame Retardant (BFR) Toxicity Using a Zebrafish Embryo Model.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5554410

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	Medium	The biological responses of the negative control group(s) were reported but only via graph
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Medium	Reporting omissions are unlikely to have a substantial impact on results
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	Medium	The duration of exposure was reported and reasonably appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test animals was not reported
	Metric 14:	Acclimatization and Pretreatment Conditions	Medium	The study did not report whether pretreatment conditions were the same for control and exposed groups.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	High number of replicates used.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18:	Consistency of Outcome Assessment	Medium	Outcomes were assessed consistently across study groups but few details were provided.

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Study Citation:	Usenko, C. Y., Abel, E. L., Hopkins, A., Martinez, G., Tijerina, J., Kudela, M., Norris, N., Joudeh, L., Bruce, E. D. (2016). Evaluation of Common Use Brominated Flame Retardant (BFR) Toxicity Using a Zebrafish Embryo Model.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5554410

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 via graph.
	Metric 23:	Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.

Additional Comments: Mortality results are presented on table 2 and Figure 1 on page 5.

Overall Quality Determination Medium

Study Citation:	Wu, S., Ji, G., Liu, J., Zhang, S., Gong, Y., Shi, L. (2016). TBBPA induces developmental toxicity, oxidative stress, and apoptosis in embryos and zebrafish larvae (Danio rerio). Environmental Toxicology 31(10):1241-1249.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wild type; Embryo
Health Outcome:	Cardiovascular
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4563499

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 >98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was 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	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and 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	The solvent concentration was appropriate
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The 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:	Wu, S., Ji, G., Liu, J., Zhang, S., Gong, Y., Shi, L. (2016). TBBPA induces developmental toxicity, oxidative stress, and apoptosis in embryos and zebrafish larvae (Danio rerio). Environmental Toxicology 31(10):1241-1249.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wild type; Embryo
Health Outcome:	Cardiovascular
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4563499

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
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There were no differences among groups that could have impacted 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 (Figure 6).
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This evaluation is for the histological analysis of cardiac tissue.

Overall Quality Determination High

Study Citation:	Wu, S., Ji, G., Liu, J., Zhang, S., Gong, Y., Shi, L. (2016). TBBPA induces developmental toxicity, oxidative stress, and apoptosis in embryos and zebrafish larvae (Danio rerio). Environmental Toxicology 31(10):1241-1249.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wild type; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4563499

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 >98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was 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	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and 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	The solvent concentration was appropriate
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The 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:	Wu, S., Ji, G., Liu, J., Zhang, S., Gong, Y., Shi, L. (2016). TBBPA induces developmental toxicity, oxidative stress, and apoptosis in embryos and zebrafish larvae (Danio rerio). Environmental Toxicology 31(10):1241-1249.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wild type; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4563499

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
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: survival and hatch

Overall Quality Determination High

Study Citation:	Wu, S., Ji, G., Liu, J., Zhang, S., Gong, Y., Shi, L. (2016). TBBPA induces developmental toxicity, oxidative stress, and apoptosis in embryos and zebrafish larvae (Danio rerio). Environmental Toxicology 31(10):1241-1249.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wild type; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4563499

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 >98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was 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	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and 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	The solvent concentration was appropriate
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The 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:	Wu, S., Ji, G., Liu, J., Zhang, S., Gong, Y., Shi, L. (2016). TBBPA induces developmental toxicity, oxidative stress, and apoptosis in embryos and zebrafish larvae (Danio rerio). Environmental Toxicology 31(10):1241-1249.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wild type; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4563499

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
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: malformations

Overall Quality Determination High

Study Citation:	Wu, S., Ji, G., Liu, J., Zhang, S., Gong, Y., Shi, L. (2016). TBBPA induces developmental toxicity, oxidative stress, and apoptosis in embryos and zebrafish larvae (Danio rerio). Environmental Toxicology 31(10):1241-1249.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wild type; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cytotoxicity-Oxidative stress (including redox biology)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4563499

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 >98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was 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	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and 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	The solvent concentration was appropriate
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The 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:	Wu, S., Ji, G., Liu, J., Zhang, S., Gong, Y., Shi, L. (2016). TBBPA induces developmental toxicity, oxidative stress, and apoptosis in embryos and zebrafish larvae (Danio rerio). Environmental Toxicology 31(10):1241-1249.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wild type; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cytotoxicity-Oxidative stress (including redox biology)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4563499

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
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: enzymes, apoptosis

Overall Quality Determination High

Study Citation:	Zhu, B., Han, J., Lei, L., Hua, J., Zuo, Y., Zhou, B. (2021). Effects of SiO2 nanoparticles on the uptake of tetrabromobisphenol A and its impact on the thyroid endocrine system in zebrafish larvae. Ecotoxicology and Environmental Safety 209:111845.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB strain; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10116509

Domain		Metric	Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The TBBPA was identified by CASRN.
	Metric 2:	Test Substance Source	Low	The TBBPA was reported to be from Sigma, but it was not reported to be analytically verified.
	Metric 3:	Test Substance Purity	High	The purity was reported to be >97%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of an appropriate concurrent negative control.
	Metric 5:	Negative Control Response	High	The negative control responses for developmental outcomes were reported in Table 1.
	Metric 6:	Randomized Allocation	Medium	It was reported that the zebrafish eggs were randomly selected and exposed to TBBPA.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	It was reported that the TBBPA was prepared in DMSO and stored at 4C. However, no other information regarding the test chemical preparation, including test concentration preparation, was reported. The test system was a static system with daily renewals.
	Metric 8:	Consistency of Exposure Administration	Low	Exposures occurred in 600mL of exposure media starting at 2h post fertilization until 120hpf. It is unclear how many embryos were placed in each test chamber. The size of the test chambers were not reported. Exposure solutions were renewed daily. Little else was described regarding the exposure system.
	Metric 9:	Measurement of Test Substance Concentration	High	Water samples were analyzed using GC-MS/MS. TBBPA content in the water is reported in Figure 1A.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration was reported to be from 2hpf to 120hpf. This was adequate to observe the outcomes of interest.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	There were three exposure levels of TBBPA, which is a little lower than is typical, but the single chemical exposure of TBBPA was also compared to several concentrations of TBBPA combined with silicon dioxide nanoparticles. The three exposure levels of TBBPA alone were adequate to observe dose responses.
	Metric 12:	Testing at or Below Solubility Limit	High	The test concentrations were under the water solubility limit. The TBBPA was also prepared in DMSO as a solvent.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	It was unclear if the adult zebrafish that provided the embryos were cultured in the performing laboratory or obtained from elsewhere. The embryos were 2hpf at the start of the study.

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Study Citation:	Zhu, B., Han, J., Lei, L., Hua, J., Zuo, Y., Zhou, B. (2021). Effects of SiO2 nanoparticles on the uptake of tetrabromobisphenol A and its impact on the thyroid endocrine system in zebrafish larvae. Ecotoxicology and Environmental Safety 209:111845.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB strain; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10116509

Domain	Metric		Rating	Comments
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It is unclear if any acclimation occurred. Adults were reported to be held at 28C while embryos were obtained, but the test temperature was not reported.
	Metric 15:	Number of Organisms and Replicates per Group	Low	It was reported 600 embryos were randomly selected for exposure to the test concentrations. The number of embryos per test chamber was not reported. There were three replicates per test concentration.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Little information was reported on the environmental conditions. Adults were kept at 28C with a 14L:10D photoperiod, but water characteristics were not described, feeding regimen was not reported, and loading rate was not reported.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome(s) of interest-changes in developmental parameters due to TBBPA exposure, including malformations (axial spinal curvature, pericardial edema, and yolk sac edema), body length, and body weight.
	Metric 18:	Consistency of Outcome Assessment	Medium	After 120hpf, body length and body weight were measured. It was reported that developmental parameters were such as axial spinal curvature, pericardial edema, and yolk sac edema were reported to be recorded daily, but the details of how this was determined 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 in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Differences between the control group and each exposure gorup were evaluated by ANOVA followed by Dunnett's multiple comparison using SPSS v16.0.
	Metric 22:	Reporting of Data	High	Results for developmental outcomes for exposure groups and control groups were reported in Table 1.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.

Additional Comments: This evaluation is for the effect of TBBPA on zebrafish embryo development. Malformations including axial spinal curvature, pericardial edema, and yolk sac edema were reported under "malformations (%)." Body length and body weight were also assessed.

Overall Quality Determination Medium

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Study Citation:	Zhu, B., Han, J., Lei, L., Hua, J., Zuo, Y., Zhou, B. (2021). Effects of SiO2 nanoparticles on the uptake of tetrabromobisphenol A and its impact on the thyroid endocrine system in zebrafish larvae. Ecotoxicology and Environmental Safety 209:111845.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB strain; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10116509

Domain	Metric	Rating	Comments

Study Citation:	Zhu, B., Han, J., Lei, L., Hua, J., Zuo, Y., Zhou, B. (2021). Effects of SiO2 nanoparticles on the uptake of tetrabromobisphenol A and its impact on the thyroid endocrine system in zebrafish larvae. Ecotoxicology and Environmental Safety 209:111845.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB strain; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10116509

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The TBBPA was identified by CASRN.
	Metric 2:	Test Substance Source	Low	The TBBPA was reported to be from Sigma, but it was not reported to be analytically verified.
	Metric 3:	Test Substance Purity	High	The purity was reported to be >97%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of an appropriate concurrent negative control.
	Metric 5:	Negative Control Response	High	The negative control responses for mortality outcomes (survival and hatching %) were reported in Table 1.
	Metric 6:	Randomized Allocation	Medium	It was reported that the zebrafish eggs were randomly selected and exposed to TBBPA.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	It was reported that the TBBPA was prepared in DMSO and stored at 4C. However, no other information regarding the test chemical preparation, including test concentration preparation, was reported. The test system was a static system with daily renewals.
	Metric 8:	Consistency of Exposure Administration	Low	Exposures occurred in 600mL of exposure media starting at 2h post fertilization until 120hpf. It is unclear how many embryos were placed in each test chamber. The size of the test chambers were not reported. Exposure solutions were renewed daily. Little else was described regarding the exposure system.
	Metric 9:	Measurement of Test Substance Concentration	High	Water samples were analyzed using GC-MS/MS. TBBPA content in the water is reported in Figure 1A.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration was reported to be from 2hpf to 120hpf. This was adequate to observe the outcomes of interest.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	There were three exposure levels of TBBPA, which is a little lower than is typical, but the single chemical exposure of TBBPA was also compared to several concentrations of TBBPA combined with silicon dioxide nanoparticles. The three exposure levels of TBBPA alone were adequate to observe dose responses.
	Metric 12:	Testing at or Below Solubility Limit	High	The test concentrations were under the water solubility limit. The TBBPA was also prepared in DMSO as a solvent.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	It was unclear if the adult zebrafish that provided the embryos were cultured in the performing laboratory or obtained from elsewhere. The embryos were 2hpf at the start of the study.

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Study Citation:	Zhu, B., Han, J., Lei, L., Hua, J., Zuo, Y., Zhou, B. (2021). Effects of SiO2 nanoparticles on the uptake of tetrabromobisphenol A and its impact on the thyroid endocrine system in zebrafish larvae. Ecotoxicology and Environmental Safety 209:111845.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB strain; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10116509

Domain	Metric		Rating	Comments
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It is unclear if any acclimation occurred. Adults were reported to be held at 28C while embryos were obtained, but the test temperature was not reported.
	Metric 15:	Number of Organisms and Replicates per Group	Low	It was reported 600 embryos were randomly selected for exposure to the test concentrations. The number of embryos per test chamber was not reported. There were three replicates per test concentration.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Little information was reported on the environmental conditions. Adults were kept at 28C with a 14L:10D photoperiod, but water characteristics were not described, feeding regimen was not reported, and loading rate was not reported.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome(s) of interest-mortality in terms of survival and percent hatch.
	Metric 18:	Consistency of Outcome Assessment	Medium	Embryo survival and hatched larvae were checked daily, though the exact methods for this were unclear.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Differences between the control group and each exposure gorup were evaluated by ANOVA followed by Dunnett's multiple comparison using SPSS v16.0.
	Metric 22:	Reporting of Data	High	Results for mortality outcomes for exposure groups and control groups were reported in Table 1.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.

Additional Comments: This evaluation is for the effect of TBBPA on zebrafish embryo survival and larval hatch reported in Table 1.

Overall Quality Determination Medium

Study Citation:	Zhu, B., Han, J., Lei, L., Hua, J., Zuo, Y., Zhou, B. (2021). Effects of SiO2 nanoparticles on the uptake of tetrabromobisphenol A and its impact on the thyroid endocrine system in zebrafish larvae. Ecotoxicology and Environmental Safety 209:111845.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB strain; Embryo
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10116509

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The TBBPA was identified by CASRN.
	Metric 2:	Test Substance Source	Low	The TBBPA was reported to be from Sigma, but it was not reported to be analytically verified.
	Metric 3:	Test Substance Purity	High	The purity was reported to be >97%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of an appropriate concurrent negative control.
	Metric 5:	Negative Control Response	High	The negative control response for TBBPA content in embryos is reported in Figure 1B. No TBBPA was found in the control embryos.
	Metric 6:	Randomized Allocation	Medium	It was reported that the zebrafish eggs were randomly selected and exposed to TBBPA.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	It was reported that the TBBPA was prepared in DMSO and stored at 4C. However, no other information regarding the test chemical preparation, including test concentration preparation, was reported. The test system was a static system with daily renewals.
	Metric 8:	Consistency of Exposure Administration	Low	Exposures occurred in 600mL of exposure media starting at 2h post fertilization until 120hpf. It is unclear how many embryos were placed in each test chamber. The size of the test chambers were not reported. Exposure solutions were renewed daily. Little else was described regarding the exposure system.
	Metric 9:	Measurement of Test Substance Concentration	High	Water samples were analyzed using GC-MS/MS. TBBPA content in the water is reported in Figure 1A.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration was reported to be from 2hpf to 120hpf. This was adequate to observe the outcomes of interest.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	There were three exposure levels of TBBPA, which is a little lower than is typical, but the single chemical exposure of TBBPA was also compared to several concentrations of TBBPA combined with silicon dioxide nanoparticles. The three exposure levels of TBBPA alone were adequate to observe dose responses.
	Metric 12:	Testing at or Below Solubility Limit	High	The test concentrations were under the water solubility limit. The TBBPA was also prepared in DMSO as a solvent.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	It was unclear if the adult zebrafish that provided the embryos were cultured in the performing laboratory or obtained from elsewhere. The embryos were 2hpf at the start of the study.

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Study Citation:	Zhu, B., Han, J., Lei, L., Hua, J., Zuo, Y., Zhou, B. (2021). Effects of SiO2 nanoparticles on the uptake of tetrabromobisphenol A and its impact on the thyroid endocrine system in zebrafish larvae. Ecotoxicology and Environmental Safety 209:111845.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB strain; Embryo
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10116509

Domain	Metric		Rating	Comments
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It is unclear if any acclimation occurred. Adults were reported to be held at 28C while embryos were obtained, but the test temperature was not reported.
	Metric 15:	Number of Organisms and Replicates per Group	Low	It was reported 600 embryos were randomly selected for exposure to the test concentrations. The number of embryos per test chamber was not reported. There were three replicates per test concentration.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Little information was reported on the environmental conditions. Adults were kept at 28C with a 14L:10D photoperiod, but water characteristics were not described, feeding regimen was not reported, and loading rate was not reported.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome(s) of interest-TBBPA concentrations in zebrafish larvae post exposure.
	Metric 18:	Consistency of Outcome Assessment	Medium	It was reported that larvae were randomly collected from each beaker for analysis. GC-MS/MS was used to determine tissue concentrations. However, it is unclear when the larvae were collected for analysis. Likely this was at 120hpf, but it was never explicitly stated.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Differences between the control group and each exposure gorup were evaluated by ANOVA followed by Dunnett's multiple comparison using SPSS v16.0.
	Metric 22:	Reporting of Data	High	Results for TBBPA concentrations in larvae are reported in Figure 1B. Both control and exposure results are reported here.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.

Additional Comments: This evaluation is for the the accumulation of TBBPA in zebrafish larvae during a 118h exposure. Embryos were exposed to TBBPA from 2hpf to 120hpf. TBBPA content in the embryos is reported in Fig. 1.

Overall Quality Determination Medium

Study Citation:	Zhu, B., Han, J., Lei, L., Hua, J., Zuo, Y., Zhou, B. (2021). Effects of SiO2 nanoparticles on the uptake of tetrabromobisphenol A and its impact on the thyroid endocrine system in zebrafish larvae. Ecotoxicology and Environmental Safety 209:111845.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB strain; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Endocrine toxicity
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10116509

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The TBBPA was identified by CASRN.
	Metric 2:	Test Substance Source	Low	The TBBPA was reported to be from Sigma, but it was not reported to be analytically verified.
	Metric 3:	Test Substance Purity	High	The purity was reported to be >97%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of an appropriate concurrent negative control.
	Metric 5:	Negative Control Response	High	Negative control responses for mechanistic outcomes are reported in Figures 2-4. Figure 2 reports thyroid hormone responses, Figure 3 reports gene transcription profiles, and Figure 4 reports Western blot analyses of TTR protein levels. All control responses were adequate.
	Metric 6:	Randomized Allocation	Medium	It was reported that the zebrafish eggs were randomly selected and exposed to TBBPA.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	It was reported that the TBBPA was prepared in DMSO and stored at 4C. However, no other information regarding the test chemical preparation, including test concentration preparation, was reported. The test system was a static system with daily renewals.
	Metric 8:	Consistency of Exposure Administration	Low	Exposures occurred in 600mL of exposure media starting at 2h post fertilization until 120hpf. It is unclear how many embryos were placed in each test chamber. The size of the test chambers were not reported. Exposure solutions were renewed daily. Little else was described regarding the exposure system.
	Metric 9:	Measurement of Test Substance Concentration	High	Water samples were analyzed using GC-MS/MS. TBBPA content in the water is reported in Figure 1A.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration was reported to be from 2hpf to 120hpf. This was adequate to observe the outcomes of interest.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	There were three exposure levels of TBBPA, which is a little lower than is typical, but the single chemical exposure of TBBPA was also compared to several concentrations of TBBPA combined with silicon dioxide nanoparticles. The three exposure levels of TBBPA alone were adequate to observe dose responses.
	Metric 12:	Testing at or Below Solubility Limit	High	The test concentrations were under the water solubility limit. The TBBPA was also prepared in DMSO as a solvent.
Domain 4: Test Organism				

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Study Citation:	Zhu, B., Han, J., Lei, L., Hua, J., Zuo, Y., Zhou, B. (2021). Effects of SiO2 nanoparticles on the uptake of tetrabromobisphenol A and its impact on the thyroid endocrine system in zebrafish larvae. Ecotoxicology and Environmental Safety 209:111845.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB strain; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Endocrine toxicity
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10116509

Domain	Metric		Rating	Comments
	Metric 13:	Test Organism Characteristics	Low	It was unclear if the adult zebrafish that provided the embryos were cultured in the performing laboratory or obtained from elsewhere. The embryos were 2hpf at the start of the study.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It is unclear if any acclimation occurred. Adults were reported to be held at 28C while embryos were obtained, but the test temperature was not reported.
	Metric 15:	Number of Organisms and Replicates per Group	Low	It was reported 600 embryos were randomly selected for exposure to the test concentrations. The number of embryos per test chamber was not reported. There were three replicates per test concentration.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Little information was reported on the environmental conditions. Adults were kept at 28C with a 14L:10D photoperiod, but water characteristics were not described, feeding regimen was not reported, and loading rate was not reported.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome(s) of interest-T3 and T4 hormone levels, gene transcription profiles, and Western blot analyses of TTR protein levels after exposure to TBBPA.
	Metric 18:	Consistency of Outcome Assessment	High	The outcome assessments for the mechanistic outcomes were reported clearly and were conducted consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Differences between the control group and each exposure gorup were evaluated by ANOVA followed by Dunnett's multiple comparison using SPSS v16.0.
	Metric 22:	Reporting of Data	High	Control and exposure responses for each concentrations were reported. Figure 2 reports thyroid hormone responses, Figure 3 reports gene transcription profiles, and Figure 4 reports Western blot analyses of TTR protein levels.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.

Additional Comments: This evaluation is for the the effect of TBBPA on mechanistic outcomes in a zebrafish embryo exposure. Study authors assessed T3 and T4 levels of thyroid hormones. They also assessed gene transcription profiles (primer sequences for the q-RT-PCR are reported in the supplemental material), and TTR levels via Wester blot analysis. These results can be found in Figures 2-4.

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Study Citation:	Zhu, B., Han, J., Lei, L., Hua, J., Zuo, Y., Zhou, B. (2021). Effects of SiO2 nanoparticles on the uptake of tetrabromobisphenol A and its impact on the thyroid endocrine system in zebrafish larvae. Ecotoxicology and Environmental Safety 209:111845.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB strain; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Endocrine toxicity
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10116509

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

Study Citation:	Zhu, B., Zhao, G., Yang, L., Zhou, B. (2018). Tetrabromobisphenol A caused neurodevelopmental toxicity via disrupting thyroid hormones in zebrafish larvae. Chemosphere 197:353-361.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wild strain; Embryo
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5489136

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The source was reported but the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >97%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported for behavior assay (Fig 5).
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The experimental system and test media preparation methods were reported. Stock solution was prepared in DMSO but no details were given regarding dilution water used. Experiments were conducted in glass beakers and 600 ml of exposure media were used. The final concentration of DMSO in test media was 0.01% (vol/vol) The test media was renewed by 50% everyday. Exposure concentrations were not verified."The exposure protocol and parameters determined were the same as part I" experiments.
	Metric 8:	Consistency of Exposure Administration	Medium	Details of exposure administration were reported. But it was not clearly stated that exposure volume and number of embryos were the same for controls and treatment groups. Embryos were " randomly distributed into glass beakers (approximately 500 embryos each) containing 600mL exposure media." The exact number of embryos used per concentration was not provided.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	Only one treatment group.
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.

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Study Citation:	Zhu, B., Zhao, G., Yang, L., Zhou, B. (2018). Tetrabromobisphenol A caused neurodevelopmental toxicity via disrupting thyroid hormones in zebrafish larvae. Chemosphere 197:353-361.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wild strain; Embryo
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5489136

Domain	Metric		Rating	Comments
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized and/or whether pre-treatment conditions were the same for control and exposed groups
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects. "Briefly, zebrafish larvae were placed into 24-well plates (1 larva per well), and each plate contained all replicates for all control and treated groups." There were12 plates for part 2 of the experiment.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reported environmental conditions were not sufficient to evaluate if adequate and whether differences occurred between control and exposed populations.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest. Locomotor behavior assay was described well.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
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 Figure 5.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This form pertains to behavioral outcome (average swimming speed) of zebrafish embryos in part 2 of the experiment, where zebrafish embryos were exposed to TBBPA and T3 alone and in mixture. There was only one exposure group of TBBPA alone in this part of the experiment.

Overall Quality Determination High

Study Citation:	Zhu, B., Zhao, G., Yang, L., Zhou, B. (2018). Tetrabromobisphenol A caused neurodevelopmental toxicity via disrupting thyroid hormones in zebrafish larvae. Chemosphere 197:353-361.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wild strain; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5489136

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The source was reported but the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >97%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and adequate. Control responses for thyroid hormone concentration, acetylcholinesterase activity and mRNA expression levels were reported in figures 1, 2, 3 & 4.
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The experimental system and test media preparation methods were reported. Stock solution was prepared in DMSO but no details were given regarding dilution water used. Experiments were conducted in glass beakers and 600 ml of exposure media were used. The final concentration of DMSO in test media was 0.01% (vol/vol) The test media was renewed by 50% everyday. Exposure concentrations were not verified.
	Metric 8:	Consistency of Exposure Administration	Medium	Details of exposure administration were reported. But it was not clearly stated that exposure volume and number of embryos were the same for controls and treatment groups. Embryos were " randomly distributed into glass beakers (approximately 500 embryos each) containing 600mL exposure media." The exact number of embryos used per concentration was not provided.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response. Four exposure groups (50, 100, 200 and 400 mg/L, nominal concentrations) were reported.
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized and/or whether pretreatment conditions were the same for control and exposed groups.

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Study Citation:	Zhu, B., Zhao, G., Yang, L., Zhou, B. (2018). Tetrabromobisphenol A caused neurodevelopmental toxicity via disrupting thyroid hormones in zebrafish larvae. Chemosphere 197:353-361.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wild strain; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5489136

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects. Approximately 500 embryos were exposed in 600 ml of exposure media. This number seems very high compared to the recommendations in OECD method 212 . 4 replicates were used per concentration.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reported environmental conditions were not sufficient to evaluate if adequate and whether differences occurred between control and exposed populations.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest. Methods for RNA isolation, quantitative PCR, Thyroid hormone concentration, acetylcholinesterase activity were reported.
	Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups.
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	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 Figures 1,2 3 and 4.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This form pertains to mechanistic outcome (gene expression, thyroid hormone concentration and acetylcholinesterase activity) of zebrafish embryos in the single exposure experiment.

Overall Quality Determination High

Study Citation:	Zhu, B., Zhao, G., Yang, L., Zhou, B. (2018). Tetrabromobisphenol A caused neurodevelopmental toxicity via disrupting thyroid hormones in zebrafish larvae. Chemosphere 197:353-361.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wild strain; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5489136

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#.
	Metric 2:	Test Substance Source	Low	The source was reported but the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >97%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	Low	The biological response of the negative control group was not reported. The part 2 of the experiment was conducted based on the results of part 1, which meant that these were 2 separate experiments. However, control response was not shown separately in Table S4.
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The experimental system and test media preparation methods were reported. Stock solution was prepared in DMSO but no details were given regarding dilution water used. Experiments were conducted in glass beakers and 600 ml of exposure media were used. The final concentration of DMSO in test media was 0.01% (vol/vol) The test media was renewed by 50% everyday. Exposure concentrations were not verified.
	Metric 8:	Consistency of Exposure Administration	Medium	Details of exposure administration were reported. But it was not clearly stated that exposure volume and number of embryos were the same for controls and treatment groups. Embryos were " randomly distributed into glass beakers (approximately 500 embryos each) containing 600mL exposure media." The exact number of embryos used per concentration was not provided.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	Only one exposure group tested.
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized and/or whether pretreatment conditions were the same for control and exposed groups.

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Study Citation:	Zhu, B., Zhao, G., Yang, L., Zhou, B. (2018). Tetrabromobisphenol A caused neurodevelopmental toxicity via disrupting thyroid hormones in zebrafish larvae. Chemosphere 197:353-361.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wild strain; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5489136

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects. Approximately 500 embryos were exposed in 600 ml of exposure media. This number seems very high compared to the OECD method 212. 4 replicates were used per concentration.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reported environmental conditions were not sufficient to evaluate if adequate and whether differences occurred between control and exposed populations.
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology (malformations and weigh measurements) reported the intended outcome of interest. Malformation examples were provided but details regarding assessment methods were lacking. For weight measurements, larvae were composited but detailed methods were lacking.
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited. " The survival, malformation (eg., axialspinal curvature, pericardial edema and yolk-sac edema) andhatched larvae were recorded during the exposure period." The timing of outcome assessment for recording malformations was not provided.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described.
	Metric 22:	Reporting of Data	Medium	Data for exposure-related findings were presented for each treatment group (Table S4).
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This form pertains to growth outcome (malformations and weight) of zebrafish embryos in the part 2 of the experiment, where zebrafish embryos were exposed to TBBPA and T3 alone and in mixture.

Overall Quality Determination Medium

Study Citation:	Zhu, B., Zhao, G., Yang, L., Zhou, B. (2018). Tetrabromobisphenol A caused neurodevelopmental toxicity via disrupting thyroid hormones in zebrafish larvae. Chemosphere 197:353-361.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wild strain; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5489136

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The source was reported but the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >97%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	Medium	The biological response of the negative control group was reported and adequate (86.8% survival and 92.2% for hatching). Control responses for weight and malformations were given in Table S4.
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The experimental system and test media preparation methods were reported. Stock solution was prepared in DMSO but no details were given regarding dilution water used. Experiments were conducted in glass beakers and 600 ml of exposure media were used. The final concentration of DMSO in test media was 0.01% (vol/vol) The test media was renewed by 50% everyday. Exposure concentrations were not verified.
	Metric 8:	Consistency of Exposure Administration	Medium	Details of exposure administration were reported. But it was not clearly stated that exposure volume and number of embryos were the same for controls and treatment groups. Embryos were " randomly distributed into glass beakers (approximately 500 embryos each) containing 600mL exposure media." The exact number of embryos used per concentration was not provided.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response. Four exposure groups (50, 100, 200 and 400 mg/L, nominal concentrations) were reported.
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized and/or whether pretreatment conditions were the same for control and exposed groups.

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Study Citation:	Zhu, B., Zhao, G., Yang, L., Zhou, B. (2018). Tetrabromobisphenol A caused neurodevelopmental toxicity via disrupting thyroid hormones in zebrafish larvae. Chemosphere 197:353-361.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wild strain; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5489136

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects. Approximately 500 embryos were exposed in 600 ml of exposure media. This number seems very high compared to the recommendations in OECD method 212 . 4 replicates were used per concentration.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reported environmental conditions were not sufficient to evaluate if adequate and whether differences occurred between control and exposed populations.
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology reported the intended outcome of interest. Malformation examples were provided but details regarding assessment methods were lacking. For weight measurements, larvae were composited but detailed methods were lacking.
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited. ” The survival, malformation (eg., axialspinal curvature, pericardial edema and yolk-sac edema) andhatched larvae were recorded during the exposure period.” The timing of outcome assessment for recording malformations was not provided.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described
	Metric 22:	Reporting of Data	High	Data (percent malformations and weight) for exposure-related findings were presented for each treatment and control group in Table S4.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This form pertains to growth outcome (malformations and weight) of zebrafish embryos in the single exposure experiment.

Overall Quality Determination Medium

Study Citation:	Zhu, B., Zhao, G., Yang, L., Zhou, B. (2018). Tetrabromobisphenol A caused neurodevelopmental toxicity via disrupting thyroid hormones in zebrafish larvae. Chemosphere 197:353-361.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wild strain; Embryo
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5489136

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The source was reported but the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >97%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group (section 2.6).
	Metric 5:	Negative Control Response	High	Control responses for locomotor activity measurements were given in Fig 5.
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The experimental system and test media preparation methods were reported. Stock solution was prepared in DMSO but no details were given regarding dilution water used. Experiments were conducted in glass beakers and 600 ml of exposure media were used. The final concentration of DMSO in test media was 0.01% (vol/vol) The test media was renewed by 50% everyday. Exposure concentrations were not verified.
	Metric 8:	Consistency of Exposure Administration	Medium	Details of exposure administration were reported. But it was not clearly stated that exposure volume and number of embryos were the same for controls and treatment groups. Embryos were " randomly distributed into glass beakers (approximately 500 embryos each) containing 600mL exposure media." The exact number of embryos used per concentration was not provided.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response. Four exposure groups (50, 100, 200 and 400 mg/L, nominal concentrations) were reported.
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized and/or whether pretreatment conditions were the same for control and exposed groups,

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Study Citation:	Zhu, B., Zhao, G., Yang, L., Zhou, B. (2018). Tetrabromobisphenol A caused neurodevelopmental toxicity via disrupting thyroid hormones in zebrafish larvae. Chemosphere 197:353-361.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wild strain; Embryo
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5489136

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects. ”Briefly, zebrafish larvae wereplaced into 24-well plates (1 larva per well), and each plate containedall replicates for all control and treated groups.” There were12 plates for single exposure experiment.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reported environmental conditions were not sufficient to evaluate if adequate and whether differences occurred between control and exposed populations.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest. Locomotor behavior assay was described well.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups. Locomotor behavior assay was done at 144 hpf.
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	Statistical methods were adequately described.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group (Fig 5).
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.
Additional Comments:	This form pertains to behavior outcome (average swimming speed) measured in zebrafish embryos in the single exposure experiment.			

Overall Quality Determination High

Study Citation:	Zhu, B., Zhao, G., Yang, L., Zhou, B. (2018). Tetrabromobisphenol A caused neurodevelopmental toxicity via disrupting thyroid hormones in zebrafish larvae. Chemosphere 197:353-361.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wild strain; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5489136

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The source was reported but the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >97%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	Medium	The biological response of the negative control group was reported and adequate (86.8% survival and 92.2% for hatching).
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The experimental system and test media preparation methods were reported. Stock solution was prepared in DMSO but no details were given regarding dilution water used. Experiments were conducted in glass beakers and 600 ml of exposure media were used. The final concentration of DMSO in test media was 0.01% (vol/vol). The test media was renewed by 50% everyday. Exposure concentrations were not verified.
	Metric 8:	Consistency of Exposure Administration	Medium	Details of exposure administration were reported. But it was not clearly stated that exposure volume and number of embryos were the same for controls and treatment groups. Embryos were " randomly distributed into glass beakers (approximately 500 embryos each) containing 600mL exposure media." The exact number of embryos used per concentration was not provided.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and 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. Four exposure groups (50, 100, 200 and 400 mg/L, nominal concentrations) were reported.
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized and/or whether pretreatment conditions were the same for control and exposed groups.

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Study Citation:	Zhu, B., Zhao, G., Yang, L., Zhou, B. (2018). Tetrabromobisphenol A caused neurodevelopmental toxicity via disrupting thyroid hormones in zebrafish larvae. Chemosphere 197:353-361.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wild strain; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5489136

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects. Approximately 500 embryos were exposed in 600 ml of exposure media. This number seems very high compared to the OECD method 212. 4 replicates were used per concentration.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reported environmental conditions were not sufficient to evaluate if adequate and whether differences occurred between control and exposed populations.
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology reported the intended outcome of interest. But the exact number of embryos used per concentration were not provided, without which percent survival and hatching cannot be accurately determined. Criteria for assessing death were not provided.
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited. " The survival, malformation (eg., axialspinal curvature, pericardial edema and yolk-sac edema) andhatched larvae were recorded during the exposure period." The timing of outcome assessment was not provided.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group (Table S4).
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This form pertains to mortality outcome (percent survival and percent hatch) of zebrafish embryos in the single exposure experiment.

Overall Quality Determination Medium

Study Citation:	Zhu, B., Zhao, G., Yang, L., Zhou, B. (2018). Tetrabromobisphenol A caused neurodevelopmental toxicity via disrupting thyroid hormones in zebrafish larvae. Chemosphere 197:353-361.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wild strain; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5489136

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The source was reported but the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >97%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	Low	The biological response of the negative control group was not reported. The part 2 of the experiment was conducted based on the results of part 1, which meant that these were 2 separate experiments. However, control response was not shown separately in Table S4.
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The experimental system and test media preparation methods were reported. Stock solution was prepared in DMSO but no details were given regarding dilution water used. Experiments were conducted in glass beakers and 600 ml of exposure media were used. The final concentration of DMSO in test media was 0.01% (vol/vol) The test media was renewed by 50% everyday. Exposure concentrations were not verified.
	Metric 8:	Consistency of Exposure Administration	Medium	Details of exposure administration were reported. But it was not clearly stated that exposure volume and number of embryos were the same for controls and treatment groups. Embryos were " randomly distributed into glass beakers (approximately 500 embryos each) containing 600mL exposure media." The exact number of embryos used per concentration was not provided.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	Only one exposure group tested.
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized and/or whether pretreatment conditions were the same for control and exposed groups.

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Study Citation:	Zhu, B., Zhao, G., Yang, L., Zhou, B. (2018). Tetrabromobisphenol A caused neurodevelopmental toxicity via disrupting thyroid hormones in zebrafish larvae. Chemosphere 197:353-361.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wild strain; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5489136

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects. Approximately 500 embryos were exposed in 600 ml of exposure media. This number seems very high compared to the OECD method 212. 4 replicates were used per concentration.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reported environmental conditions were not sufficient to evaluate if adequate and whether differences occurred between control and exposed populations.
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology reported the intended outcome of interest. But the exact number of embryos used per concentration were not provided, without which percent survival and hatching cannot be accurately determined. Criteria for assessing death were not provided
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited. " The survival, malformation (eg., axialspinal curvature, pericardial edema and yolk-sac edema) andhatched larvae were recorded during the exposure period." The timing of outcome assessment was not provided.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described
	Metric 22:	Reporting of Data	Medium	Data for exposure-related findings were presented for each treatment group (Table S4).
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This form pertains to mortality outcome (percent survival and percent hatch) of zebrafish embryos in part 2 of the experiment, where zebrafish embryos were exposed to TBBPA and T3 alone and in mixture. Control response for part 2 of the experiment was not shown separately in Table S4.

Overall Quality Determination Medium

Study Citation:	Zhu, B., Zhao, G., Yang, L., Zhou, B. (2018). Tetrabromobisphenol A caused neurodevelopmental toxicity via disrupting thyroid hormones in zebrafish larvae. Chemosphere 197:353-361.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wild strain; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5489136

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The source was reported but the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >97%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and adequate. Control responses for thyroid hormone concentration, acetylcholinesterase activity and mRNA expression levels were reported in figures 1, 2, 3 & 4.
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The experimental system and test media preparation methods were reported. Stock solution was prepared in DMSO but no details were given regarding dilution water used. Experiments were conducted in glass beakers and 600 ml of exposure media were used. The final concentration of DMSO in test media was 0.01% (vol/vol). The test media was renewed by 50% everyday. Exposure concentrations were not verified.
	Metric 8:	Consistency of Exposure Administration	Medium	Details of exposure administration were reported. But it was not clearly stated that exposure volume and number of embryos were the same for controls and treatment groups. Embryos were " randomly distributed into glass beakers (approximately 500 embryos each) containing 600mL exposure media." The exact number of embryos used per concentration was not provided.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	Only one exposure group used.
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized and/or whether pretreatment conditions were the same for control and exposed groups.

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Study Citation:	Zhu, B., Zhao, G., Yang, L., Zhou, B. (2018). Tetrabromobisphenol A caused neurodevelopmental toxicity via disrupting thyroid hormones in zebrafish larvae. Chemosphere 197:353-361.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Danio rerio*; AB wild strain; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5489136

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects. Approximately 500 embryos were exposed in 600 ml of exposure media. This number seems very high compared to the recommendations in OECD method 212 . 4 replicates were used per concentration.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reported environmental conditions were not sufficient to evaluate if adequate and whether differences occurred between control and exposed populations.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest. Methods for RNA isolation, quantitative PCR, Thyroid hormone concentration, acetylcholinesterase activity were reported.
	Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups.
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	Statistical methods were adequately described
	Metric 22:	Reporting of Data	High	Mechanistic data for exposure-related findings were presented for each treatment and control group in Figures 1,2 3 and 4.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This form pertains to mechanistic outcome (gene expression, thyroid hormone concentration and acetylcholinesterase activity) of zebrafish embryos in part 2 of the experiment, where zebrafish embryos were exposed to TBBPA and T3 alone and in mixture.

Overall Quality Determination High

Study Citation:	EAG Laboratories, (2018). Tetrabromobisphenol A: Zebrafish short-term reproduction assay (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; *Danio rerio*; Adult
Health Outcome:	Hepatic/Liver
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064187

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	High	The source was reported and the test substance identity was analytically verified by a commercial 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 for assessed outcomes.
	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 was described in adequate detail (page 15/270), and methods for preparation of test media were suitable for this chemical and given in detail (page 17/270).
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods. Measured concentrations were similar to nominal concentrations.
	Metric 10:	Exposure Duration and Frequency	High	An approved standard test duration of 21 days was used for the short-term reproduction assay.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were justified and suitable for a dose response. Four exposure groups (9.2, 29, 94, and 300 μg active ingredient (a.i.)/L) were used and these concentrations "were based on the results of an exploratory range-finding study and reported LC50 values for the test substance."
	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(pages 13-14/270).
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Pretreatment conditions very similar to test conditions, and were the same for control and exposed groups.

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Study Citation:	EAG Laboratories, (2018). Tetrabromobisphenol A: Zebrafish short-term reproduction assay (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; *Danio rerio*; Adult
Health Outcome:	Hepatic/Liver
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064187

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects. "Four replicate test chambers were maintained in each treatment and control group. Each replicate contained a reproductive group of five males and five females, for a total of 40 fish per treatment or control group."
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Organism housing, environmental conditions, food and water were conducive to maintenance of health and biomass loading was appropriate.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest. Details are in Appendix 9.
	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 (Table 5).
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described.
	Metric 22:	Reporting of Data	High	Results were described in Appendix 9 (Histopathology Report). Liver data was given in Tables 9.3 and 9.4 and Appendix 9.5.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This evaluation is for the histology assessment of liver tissue.

Overall Quality Determination High

Study Citation:	EAG Laboratories, (2018). Tetrabromobisphenol A: Zebrafish short-term reproduction assay (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; *Danio rerio*; Adult
Health Outcome:	Endocrine
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064187

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	High	The source was reported and the test substance identity was analytically verified by a commercial 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 for assessed outcomes.
	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 was described in adequate detail (page 15/270), and methods for preparation of test media were suitable for this chemical and given in detail (page 17/270).
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods. Measured concentrations were similar to nominal concentrations.
	Metric 10:	Exposure Duration and Frequency	High	An approved standard test duration of 21 days was used for this study.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were justified and suitable for a dose response. 4 exposure groups (9.2, 29, 94, and 300 μg active ingredient (a.i.)/L) were used and these concentrations "were based on the results of an exploratory range-finding study and reported LC50 values for the test substance."
	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(pages 13-14/270).
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Pretreatment conditions very similar to test conditions, and were the same for control and exposed groups.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects. "Four replicate test chambers were maintained in each treatment and control group. Each replicate contained a reproductive group of five males and five females, for a total of 40 fish per treatment or control group."

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Study Citation:	EAG Laboratories, (2018). Tetrabromobisphenol A: Zebrafish short-term reproduction assay (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; *Danio rerio*; Adult
Health Outcome:	Endocrine
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064187

Domain	Metric		Rating	Comments
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Organism housing, environmental conditions, food and water were conducive to maintenance of health and biomass loading was appropriate.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest. Details of the methods were given in Appendix 9.
	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 (Table 5).
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group in tables 9.5-9.6 and Appendix 9.15.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This evaluation is for the histology assessment of thyroid tissue.

Overall Quality Determination High

Study Citation:	EAG Laboratories, (2018). Tetrabromobisphenol A: Zebrafish short-term reproduction assay (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; *Danio rerio*; Adult
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064187

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	High	The source was reported and the test substance identity was analytically verified by a commercial 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 for assessed outcomes.
	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 was described in adequate detail (page 15/270), and methods for preparation of test media were suitable for this chemical and given in detail (page 17/270).
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods. Measured concentrations were similar to nominal concentrations.
	Metric 10:	Exposure Duration and Frequency	High	An approved standard test duration of 21 days was used for the short-term reproduction assay.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were justified and suitable for a dose response. 4 exposure groups (9.2, 29, 94, and 300 μg active ingredient (a.i.)/L) were used and these concentrations "were based on the results of an exploratory range-finding study and reported LC50 values for the test substance."
	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(pages 13-14/270).
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Pretreatment conditions very similar to test conditions, and were the same for control and exposed groups.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects. "Four replicate test chambers were maintained in each treatment and control group. Each replicate contained a reproductive group of five males and five females, for a total of 40 fish per treatment or control group."

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Study Citation:	EAG Laboratories, (2018). Tetrabromobisphenol A: Zebrafish short-term reproduction assay (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; *Danio rerio*; Adult
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064187

Domain	Metric		Rating	Comments
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Organism housing, environmental conditions, food and water were conducive to maintenance of health and biomass loading was appropriate.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest. "The number of eggs spawned and the number of fertile eggs were determined for each spawning group daily during the 21-dayexposure. Fecundity was expressed on the basis of number of eggs per surviving female per reproductive (exposure) day per replicate and as cumulative eggs produced over the exposure period." Gonad weight measurements were taken before gonadal histological analysis.
	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 (Table 5).
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group. Gonad weight and GSI were given in Table 8 and appendices 24 & 25. Data for reproductive end points were given in table 6 , appendices 22& 23.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: A well-conducted and well-written study on a 21 day reproduction assay in zebra fish. This evaluation form is relevant to the reproductive outcomes (GSI, gonad weight, eggs per female, % fertilization) reported in the study

Overall Quality Determination High

Study Citation:	EAG Laboratories, (2018). Tetrabromobisphenol A: Zebrafish short-term reproduction assay (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; *Danio rerio*; Adult
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064187

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	High	The source was reported and the test substance identity was analytically verified by a commercial 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 for assessed outcomes.
	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 was described in adequate detail (page 15/270), and methods for preparation of test media were suitable for this chemical and given in detail (page 17/270).
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods. Measured concentrations were similar to nominal concentrations.
	Metric 10:	Exposure Duration and Frequency	High	An approved standard test duration of 21 days was used for the short-term reproduction assay.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were justified and suitable for a dose response. 4 exposure groups (9.2, 29, 94, and 300 μg active ingredient (a.i.)/L) were used and these concentrations "were based on the results of an exploratory range-finding study and reported LC50 values for the test substance."
	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 (pages 13-14/270).
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Pretreatment conditions very similar to test conditions, and were the same for control and exposed groups.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects. "Four replicate test chambers were maintained in each treatment and control group. Each replicate contained a reproductive group of five males and five females, for a total of 40 fish per treatment or control group."

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Study Citation:	EAG Laboratories, (2018). Tetrabromobisphenol A: Zebrafish short-term reproduction assay (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; *Danio rerio*; Adult
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064187

Domain	Metric		Rating	Comments
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Organism housing, environmental conditions, food and water were conducive to maintenance of health and biomass loading was appropriate.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest. Methods to determine plasma vitellogenin concentration and hepatic vitellogenin were given in detail.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions (Table 5).
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group (Table 8).
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: A well-conducted and well-written study on a 21 day reproduction assay in zebra fish. This evaluation form is relevant to the mechanistic outcome (plasma and hepatic vitellogenin).

Overall Quality Determination High

Study Citation:	EAG Laboratories, (2018). Tetrabromobisphenol A: Zebrafish short-term reproduction assay (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; *Danio rerio*; Adult
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064187

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	High	The source was reported and the test substance identity was analytically verified by a commercial 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 for assessed outcomes.
	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 was described in adequate detail (page 15/270), and methods for preparation of test media were suitable for this chemical and given in detail (page 17/270).
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods. Measured concentrations were similar to nominal concentrations.
	Metric 10:	Exposure Duration and Frequency	High	An approved standard test duration of 21 days was used for the short-term reproduction assay.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were justified and suitable for a dose response. 4 exposure groups (9.2, 29, 94, and 300 μg active ingredient (a.i.)/L) were used and these concentrations "were based on the results of an exploratory range-finding study and reported LC50 values for the test substance."
	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 (pages 13-14/270).
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Pretreatment conditions very similar to test conditions, and were the same for control and exposed groups.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects. "Four replicate test chambers were maintained in each treatment and control group. Each replicate contained a reproductive group of five males and five females, for a total of 40 fish per treatment or control group."

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Study Citation:	EAG Laboratories, (2018). Tetrabromobisphenol A: Zebrafish short-term reproduction assay (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; *Danio rerio*; Adult
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064187

Domain	Metric		Rating	Comments
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Organism housing, environmental conditions, food and water were conducive to maintenance of health and biomass loading was appropriate.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest. "The wet weight and standard length of each fish were recorded at test termination. Fish were blotted dry and weighed to the nearest 0.1 mg. Standard length was measured to the nearest millimeter".
	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 (Table 5).
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group (Table 7).
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: A well-conducted and well-written study on a 21 day reproduction assay in zebra fish. This evaluation form is relevant to the growth outcome (body length and weight measurements).

Overall Quality Determination High

Study Citation:	EAG Laboratories, (2018). Tetrabromobisphenol A: Zebrafish short-term reproduction assay (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; *Danio rerio*; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064187

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	High	The source was reported and test substance identity was analytically verified by a commercial 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 adequate for assessed outcomes.
	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 was described in adequate detail (page 15/270), and methods for preparation of test media were suitable for this chemical and given in detail (page 17/270).
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods. Measured concentrations were similar to nominal concentrations.
	Metric 10:	Exposure Duration and Frequency	High	An approved standard test duration of 21 days was used for the short-term reproduction assay.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were justified and suitable for a dose response. 4 exposure groups (9.2, 29, 94, and 300 μg active ingredient (a.i.)/L) were used and these concentrations "were based on the results of an exploratory range-finding study and reported LC50 values for the test substance."
	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 (pages 13-14/270).
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Pretreatment conditions very similar to test conditions, and were the same for control and exposed groups.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects. "Four replicate test chambers were maintained in each treatment and control group. Each replicate contained a reproductive group of five males and five females, for a total of 40 fish per treatment or control group."

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Study Citation:	EAG Laboratories, (2018). Tetrabromobisphenol A: Zebrafish short-term reproduction assay (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; *Danio rerio*; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064187

Domain	Metric		Rating	Comments
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Organism housing, environmental conditions, food and water were conducive to maintenance of health and biomass loading was appropriate.
	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. Mortality was assessed daily and dead fish were removed from test chambers as soon as possible.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described in Appendix 10.
	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: A well-conducted and well-written study on a 21 day reproduction assay in zebra fish. This evaluation form is relevant to the mortality outcome.

Overall Quality Determination High

Study Citation:	Song, M., Liang, D., Liang, Y., Chen, M., Wang, F., Wang, H., Jiang, G. (2014). Assessing developmental toxicity and estrogenic activity of halogenated bisphenol A on zebrafish (Danio rerio). Chemosphere 112:275-281.
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; *Danio rerio*; male; Adult
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2754370

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 >=98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control groups
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was 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 for making a stock solution
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The test organisms were adequately described unsure where they were obtained from
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest

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Study Citation:	Song, M., Liang, D., Liang, Y., Chen, M., Wang, F., Wang, H., Jiang, G. (2014). Assessing developmental toxicity and estrogenic activity of halogenated bisphenol A on zebrafish (Danio rerio). Chemosphere 112:275-281.
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; *Danio rerio*; male; Adult
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2754370

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
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: estrogenic activity

Overall Quality Determination High

Study Citation:	Song, M., Liang, D., Liang, Y., Chen, M., Wang, F., Wang, H., Jiang, G. (2014). Assessing developmental toxicity and estrogenic activity of halogenated bisphenol A on zebrafish (Danio rerio). Chemosphere 112:275-281.
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; *Danio rerio*; male; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2754370

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 >=98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control groups
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was 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 for making a stock solution
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The test organisms were adequately described unsure where they were obtained from
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups

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Study Citation:	Song, M., Liang, D., Liang, Y., Chen, M., Wang, F., Wang, H., Jiang, G. (2014). Assessing developmental toxicity and estrogenic activity of halogenated bisphenol A on zebrafish (Danio rerio). Chemosphere 112:275-281.
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; *Danio rerio*; male; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2754370

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
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: None

Overall Quality Determination High

Study Citation:	Chen, J., Tanguay, R. L., Simonich, M., Nie, S., Zhao, Y., Li, L., Bai, C., Dong, Q., Huang, C., Lin, K. (2016). TBBPA chronic exposure produces sex-specific neurobehavioral and social interaction changes in adult zebrafish. Neurotoxicology and Teratology 56:9-15.
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; *Danio rerio*; AB; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5519142

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >97%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	Low	Mortality was not reported for the control group.
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Only two concentrations were tested
	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 the source of the stock 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	replicates weren't used for initial exposure
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest

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Study Citation:	Chen, J., Tanguay, R. L., Simonich, M., Nie, S., Zhao, Y., Li, L., Bai, C., Dong, Q., Huang, C., Lin, K. (2016). TBBPA chronic exposure produces sex-specific neurobehavioral and social interaction changes in adult zebrafish. Neurotoxicology and Teratology 56:9-15.
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; *Danio rerio*; AB; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5519142

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal health outcomes unrelated to exposure
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described
	Metric 22:	Reporting of Data	Low	Mortality data was not shown. Authors just mentioned that no mortality was produced by the TBBPA exposures.
	Metric 23:	Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained.

Additional Comments: None

Overall Quality Determination Medium

Study Citation:	Chen, J., Tanguay, R. L., Simonich, M., Nie, S., Zhao, Y., Li, L., Bai, C., Dong, Q., Huang, C., Lin, K. (2016). TBBPA chronic exposure produces sex-specific neurobehavioral and social interaction changes in adult zebrafish. Neurotoxicology and Teratology 56:9-15.
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; *Danio rerio*; AB; Embryo
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5519142

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >97%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Only two concentrations were tested
	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 the source of the stock 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	replicates weren't used for initial exposure
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 sex ratio assessment was not reported in the methods section.
	Metric 18:	Consistency of Outcome Assessment	Low	Unclear if outcome was assessed consistently.

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Study Citation:	Chen, J., Tanguay, R. L., Simonich, M., Nie, S., Zhao, Y., Li, L., Bai, C., Dong, Q., Huang, C., Lin, K. (2016). TBBPA chronic exposure produces sex-specific neurobehavioral and social interaction changes in adult zebrafish. Neurotoxicology and Teratology 56:9-15.
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; *Danio rerio*; AB; Embryo
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5519142

Domain	Metric		Rating	Comments
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal health outcomes unrelated to exposure
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described
	Metric 22:	Reporting of Data	Medium	Sex ration results were given in the text in section 3.1, but no data was shown.
	Metric 23:	Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained.

Additional Comments: None

Overall Quality Determination Medium

Study Citation:	Chen, J., Tanguay, R. L., Simonich, M., Nie, S., Zhao, Y., Li, L., Bai, C., Dong, Q., Huang, C., Lin, K. (2016). TBBPA chronic exposure produces sex-specific neurobehavioral and social interaction changes in adult zebrafish. Neurotoxicology and Teratology 56:9-15.
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; *Danio rerio*; AB; Embryo
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5519142

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >97%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Only two concentrations were tested
	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 the source of the stock 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	replicates weren't used for initial exposure
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups

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Study Citation:	Chen, J., Tanguay, R. L., Simonich, M., Nie, S., Zhao, Y., Li, L., Bai, C., Dong, Q., Huang, C., Lin, K. (2016). TBBPA chronic exposure produces sex-specific neurobehavioral and social interaction changes in adult zebrafish. Neurotoxicology and Teratology 56:9-15.
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; *Danio rerio*; AB; Embryo
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5519142

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

Additional Comments: None

Overall Quality Determination High

Study Citation:	Chen, J., Tanguay, R. L., Simonich, M., Nie, S., Zhao, Y., Li, L., Bai, C., Dong, Q., Huang, C., Lin, K. (2016). TBBPA chronic exposure produces sex-specific neurobehavioral and social interaction changes in adult zebrafish. Neurotoxicology and Teratology 56:9-15.
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; *Danio rerio*; AB; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5519142

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >97%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Only two concentrations were tested
	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 the source of the stock 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	replicates weren't used for initial exposure
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups

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Study Citation:	Chen, J., Tanguay, R. L., Simonich, M., Nie, S., Zhao, Y., Li, L., Bai, C., Dong, Q., Huang, C., Lin, K. (2016). TBBPA chronic exposure produces sex-specific neurobehavioral and social interaction changes in adult zebrafish. Neurotoxicology and Teratology 56:9-15.
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; *Danio rerio*; AB; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5519142

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

Additional Comments: None

Overall Quality Determination High

Study Citation:	Kuiper, R. V., Brandhof, v.d., E. J., Leonards, P. E., Ven, v.d., L. T., Wester, P. W., Vos, J. G. (2007). Toxicity of tetrabromobisphenol A (TBBPA) in zebrafish (Danio rerio) in a partial life-cycle test. Archives of Toxicology 81(1):1-9.
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; *Danio rerio*; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1414702

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	High	The test substance identity was analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.17%
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 reasonable
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods
	Metric 10:	Exposure Duration and Frequency	High	multigeneration exposure was more ambitious than standard durations
	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	Stock concentrations exceeded solubility but solvents at an appropriate level aided in dissolution
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health

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Study Citation:	Kuiper, R. V., Brandhof, v.d., E. J., Leonards, P. E., Ven, v.d., L. T., Wester, P. W., Vos, J. G. (2007). Toxicity of tetrabromobisphenol A (TBBPA) in zebrafish (Danio rerio) in a partial life-cycle test. Archives of Toxicology 81(1):1-9.
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; *Danio rerio*; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1414702

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
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There were no reported differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical analysis was conducted and adequate.
	Metric 22:	Reporting of Data	High	Survival data was shown in Table 2.
	Metric 23:	Explanation of Unexpected Outcomes	Low	No measures of variability were given with the data.

Additional Comments: This evaluation form is for the assessment of survival in exposed embryos.

Overall Quality Determination High

Study Citation:	Kuiper, R. V., Brandhof, v.d., E. J., Leonards, P. E., Ven, v.d., L. T., Wester, P. W., Vos, J. G. (2007). Toxicity of tetrabromobisphenol A (TBBPA) in zebrafish (Danio rerio) in a partial life-cycle test. Archives of Toxicology 81(1):1-9.
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; *Danio rerio*; Adult
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1414702

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	High	The test substance identity was analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.17%
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 reasonable
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods
	Metric 10:	Exposure Duration and Frequency	High	multigeneration exposure was more ambitious than standard durations
	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	Stock concentrations exceeded solubility but solvents at an appropriate level aided in dissolution
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups

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Study Citation:	Kuiper, R. V., Brandhof, v.d., E. J., Leonards, P. E., Ven, v.d., L. T., Wester, P. W., Vos, J. G. (2007). Toxicity of tetrabromobisphenol A (TBBPA) in zebrafish (Danio rerio) in a partial life-cycle test. Archives of Toxicology 81(1):1-9.
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; *Danio rerio*; Adult
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1414702

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
	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 clearly described
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: None

Overall Quality Determination High

Study Citation:	Kuiper, R. V., Brandhof, v.d., E. J., Leonards, P. E., Ven, v.d., L. T., Wester, P. W., Vos, J. G. (2007). Toxicity of tetrabromobisphenol A (TBBPA) in zebrafish (Danio rerio) in a partial life-cycle test. Archives of Toxicology 81(1):1-9.
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; *Danio rerio*; Adult
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1414702

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	High	The test substance identity was analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.17%
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 reasonable
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods
	Metric 10:	Exposure Duration and Frequency	High	multigeneration exposure was more ambitious than standard durations
	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	Stock concentrations exceeded solubility but solvents at an appropriate level aided in dissolution
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Low	Only one female and one male from each group were analyzed
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:	Kuiper, R. V., Brandhof, v.d., E. J., Leonards, P. E., Ven, v.d., L. T., Wester, P. W., Vos, J. G. (2007). Toxicity of tetrabromobisphenol A (TBBPA) in zebrafish (Danio rerio) in a partial life-cycle test. Archives of Toxicology 81(1):1-9.
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; *Danio rerio*; Adult
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1414702

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
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Uninformative	Statistical analysis was not conducted
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group that was analyzed
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: body burden

Overall Quality Determination — Uninformative

Study Citation:	Kuiper, R. V., Brandhof, v.d., E. J., Leonards, P. E., Ven, v.d., L. T., Wester, P. W., Vos, J. G. (2007). Toxicity of tetrabromobisphenol A (TBBPA) in zebrafish (Danio rerio) in a partial life-cycle test. Archives of Toxicology 81(1):1-9.
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; *Danio rerio*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1414702

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	High	The test substance identity was analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.17%
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 responses of the control group were reported and adequate.
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods
	Metric 10:	Exposure Duration and Frequency	High	multigeneration exposure was more ambitious than standard durations
	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	Stock concentrations exceeded solubility but solvents at an appropriate level aided in dissolution
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups

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Study Citation:	Kuiper, R. V., Brandhof, v.d., E. J., Leonards, P. E., Ven, v.d., L. T., Wester, P. W., Vos, J. G. (2007). Toxicity of tetrabromobisphenol A (TBBPA) in zebrafish (Danio rerio) in a partial life-cycle test. Archives of Toxicology 81(1):1-9.
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; *Danio rerio*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1414702

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
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There were no reported differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical analysis was conducted and adequate for the outcomes.
	Metric 22:	Reporting of Data	High	Hatching % data was shown in Table 2. Development/growth observations were reported in the text.
	Metric 23:	Explanation of Unexpected Outcomes	Low	Measures of variability were not given in Table 2.

Additional Comments: This evaluation is for the assessment of development/growth for exposed embryos.

Overall Quality Determination High

Study Citation:	Alzualde, A., Behl, M., Sipes, N. S., Hsieh, J. H., Alday, A., Tice, R. R., Paules, R. S., Muriana, A., Quevedo, C. (2018). Toxicity profiling of flame retardants in zebrafish embryos using a battery of assays for developmental toxicity, neurotoxicity, cardiotoxicity and hepatotoxicity toward human relevance. Neurotoxicology and Teratology 70:40-50.
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*; Wild Type AB; Embryo
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5164137

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical referred to by Name, CASRN, and Structure in Supplemental file (word file).
	Metric 2:	Test Substance Source	Low	Source and Lot number were reported but the test substance identity was NOT analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Purity was reported from manufacturer (Albermarle) as 99%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Solvent controls were used and consisted of 0.5% DMSO.
	Metric 5:	Negative Control Response	Low	It is not clear that the authors assessed any concentrations or control other than the LEL (lowest effect level).
	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	DMSO was used as the solvent and the same stock solution for used to prepare all exposure solutions. Experiments were conducted in well plates. Treatment solutions were replaced at day two of the 4 day exposure.
	Metric 8:	Consistency of Exposure Administration	High	Exposure administration appear to be consistent across treatment groups and controls.
	Metric 9:	Measurement of Test Substance Concentration	Medium	Exposure concentration in the test media was not measured. The concentration of TBBPA was assessed from a pooled sample of fish that were at the concentration that produced the LEL (lowest effect level), which was reported as nominal and internal concentration. This was reported for both experiment 1 (Fig 4) and experiment 2 (supplement table 6).
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was for 2 and 4 days post-fertilization and was appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	The study only assessed internal concentration at one level although the study had several ranges: Two experiments with similar concentrations were conducted. ranges for experiment 1 are: 1, 1.5 ,2, 3, 5, 7.5, 10, 20 uM, and Experiment 2 were 0.5, 1, 1.5, 2, 3, 5, 7.5, 10,uM. Internal concentration determined only for the larvae exposed to the lowest effect level of the test substance.

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Study Citation:	Alzualde, A., Behl, M., Sipes, N. S., Hsieh, J. H., Alday, A., Tice, R. R., Paules, R. S., Muriana, A., Quevedo, C. (2018). Toxicity profiling of flame retardants in zebrafish embryos using a battery of assays for developmental toxicity, neurotoxicity, cardiotoxicity and hepatotoxicity toward human relevance. Neurotoxicology and Teratology 70:40-50.
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*; Wild Type AB; Embryo
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5164137

Domain	Metric		Rating	Comments
	Metric 12:	Testing at or Below Solubility Limit	High	The LEL concentrations (nominal at 1.5 uM) chosen for internal concentration analysis are all under solubility. All treatment concentrations >7.5 uM were over solubility listed in the final scope at 4.1 mg/L. A majority of the treatment concentrations were under solubility.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Zebrafish husbandry, egg production, collection and housing of embryos were described well.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized and/or whether pre-treatment conditions were the same for control and exposed groups. Overall, water quality parameters were not well detailed for this study.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The authors reported that there were 16 individuals per concentration and the experiment was conducted at each concentration twice.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Environmental conditions during exposure were not reported.
	Metric 17:	Outcome Assessment Methodology	High	Section 2.3.2 reports the outcome assessment for internal concentration of TBBPA.
	Metric 18:	Consistency of Outcome Assessment	High	Outcome assessment was consistent across treatment groups and controls.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information on outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Authors just reported the internal concentration from the LEL (1.5 uM). No statistics applied.
	Metric 22:	Reporting of Data	Low	Data on concentrations were only presented as a mean and reported in table 1 and supplemental table 6. Also summarized in Figure 4.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This form is for ADME. Data on concentrations were only presented as a mean and reported in table 1 and supplemental table 6. Also summarized in Figure 4.

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Study Citation:	Alzualde, A., Behl, M., Sipes, N. S., Hsieh, J. H., Alday, A., Tice, R. R., Paules, R. S., Muriana, A., Quevedo, C. (2018). Toxicity profiling of flame retardants in zebrafish embryos using a battery of assays for developmental toxicity, neurotoxicity, cardiotoxicity and hepatotoxicity toward human relevance. Neurotoxicology and Teratology 70:40-50.
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*; Wild Type AB; Embryo
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5164137

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

Study Citation:	Alzualde, A., Behl, M., Sipes, N. S., Hsieh, J. H., Alday, A., Tice, R. R., Paules, R. S., Muriana, A., Quevedo, C. (2018). Toxicity profiling of flame retardants in zebrafish embryos using a battery of assays for developmental toxicity, neurotoxicity, cardiotoxicity and hepatotoxicity toward human relevance. Neurotoxicology and Teratology 70:40-50.
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*; Wild Type AB; Embryo
Health Outcome:	Hepatic/Liver
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5164137

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical referred to by Name, CASRN, and Structure in Supplemental file (word file).
	Metric 2:	Test Substance Source	Low	Source and Lot number were reported but the test substance was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Purity was reported from manufacturer (Albermarle) as 99%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Solvent controls were used and consisted of 0.5% DMSO.
	Metric 5:	Negative Control Response	High	Control response was given for the chemical for which hepatoxicity was observed (Fig 2D).
	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	DMSO was used as the solvent and the same stock solution for used to prepare all exposure solutions. Experiments were conducted in well plates. Treatment solutions were replaced at day two of the 4 day exposure.
	Metric 8:	Consistency of Exposure Administration	High	Exposure administration appear to be consistent across treatment groups and controls.
	Metric 9:	Measurement of Test Substance Concentration	Medium	Exposure concentrations were not measured and are reported as nominal.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was for 2 and 4 days post-fertilization and was appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Two experiments with similar concentrations were conducted. ranges for experiment 1 are: 1 1.5 2 3 5 7.5 10 20 um, and Experiment 2 were 0.5 1 1.5 2 3 5 7.5 10 uM
	Metric 12:	Testing at or Below Solubility Limit	Low	Treatment concentration >7.5 uM were above solubility. A majority of treatments were below solubility. Control response for hepatotoxicity assay for TPP was not reported.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Zebrafish husbandry, egg production, collection and housing of embryos were described well.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized and/or whether pretreatment conditions were the same for control and exposed groups. Overall, water quality parameters were not well detailed for this study.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Authors report 16 embryos per concentration and the experiment was conducted twice.

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Study Citation:	Alzualde, A., Behl, M., Sipes, N. S., Hsieh, J. H., Alday, A., Tice, R. R., Paules, R. S., Muriana, A., Quevedo, C. (2018). Toxicity profiling of flame retardants in zebrafish embryos using a battery of assays for developmental toxicity, neurotoxicity, cardiotoxicity and hepatotoxicity toward human relevance. Neurotoxicology and Teratology 70:40-50.
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*; Wild Type AB; Embryo
Health Outcome:	Hepatic/Liver
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5164137

Domain	Metric		Rating	Comments
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Environmental conditions during exposure were not reported.
	Metric 17:	Outcome Assessment Methodology	High	Section 2.3.4 reports the outcome assessment for hepatotoxicity.
	Metric 18:	Consistency of Outcome Assessment	High	Outcome assessment was consistent across treatment groups and controls.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information on outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	N/A	Visual inspection of liver yielded negative results. Therefore optical density measurements of liver were not taken for TBBPA samples.
	Metric 22:	Reporting of Data	Low	Specific results on hepatoxicity are not reported. The authors summarize the results in table 3 and supplemental table 8.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This form is for the hepatoxicity. Data are presented in table 3 and supplement table 8. No differences were observed.

Overall Quality Determination High

Study Citation:	Alzualde, A., Behl, M., Sipes, N. S., Hsieh, J. H., Alday, A., Tice, R. R., Paules, R. S., Muriana, A., Quevedo, C. (2018). Toxicity profiling of flame retardants in zebrafish embryos using a battery of assays for developmental toxicity, neurotoxicity, cardiotoxicity and hepatotoxicity toward human relevance. Neurotoxicology and Teratology 70:40-50.
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*; cmlc2:CopGFP; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5164137

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical referred to by Name, CASRN, and Structure in Supplemental file (word file).
	Metric 2:	Test Substance Source	Low	Source and Lot number were reported. The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Purity was reported from manufacturer (Albermarle) as 99%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Solvent controls were used and consisted of 0.5% DMSO.
	Metric 5:	Negative Control Response	High	Control responses are reported for mortality graphically in Figure 1 (page 5/11).
	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	DMSO was used as the solvent and the same stock solution for used to prepare all exposure solutions. Experiments were conducted in well plates. Treatment solutions were replaced at day two of the 4 day exposure.
	Metric 8:	Consistency of Exposure Administration	High	Exposure administration appear to be consistent across treatment groups and controls.
	Metric 9:	Measurement of Test Substance Concentration	Medium	Exposure concentrations were not measured and are reported as nominal. Treatment solutions were replaced at day two of the 4 day exposure.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was for 2 and 4 days post-fertilization and was appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	8 exposure groups were used based on the dose-range finding study.
	Metric 12:	Testing at or Below Solubility Limit	Medium	The two highest concentrations (10 and 20 uM) were above the solubility limit for this compound as listed in the final scope (4.1 mg/L). All hazard values listed for these experiments are under the solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Zebrafish husbandry, egg production, collection and housing of embryos were described well.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized and/or whether pretreatment conditions were the same for control and exposed groups. Overall, water quality parameters were not well detailed for this study.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	5 embryos per well and 15 embryos per concentration were used for the main experiment. This experiment was conducted twice.

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Study Citation:	Alzualde, A., Behl, M., Sipes, N. S., Hsieh, J. H., Alday, A., Tice, R. R., Paules, R. S., Muriana, A., Quevedo, C. (2018). Toxicity profiling of flame retardants in zebrafish embryos using a battery of assays for developmental toxicity, neurotoxicity, cardiotoxicity and hepatotoxicity toward human relevance. Neurotoxicology and Teratology 70:40-50.
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*; cmlc2:CopGFP; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5164137

Domain	Metric		Rating	Comments
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Environmental conditions during exposure were not reported.
	Metric 17:	Outcome Assessment Methodology	Medium	Mortality assessment method was not reported. In the OECD guidelines for fish embryo test (Test No. 236), indicators of lethality are coagulation of embryos, lack of somite formation, non-detachment of the tail and lack of heartbeat. These are not discussed in the paper.
	Metric 18:	Consistency of Outcome Assessment	High	Outcome assessment was consistent across treatment groups and controls.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information on outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	”Percentage of altered and dead embryos was used for Effective Concentration 50% (EC50) and Lethal Concentration 50% (LC50) calculations applying a nonlinear regression test (sigmoidal dose-response curve) using the GraphPad Prism (GraphPad Software). Ateratogenic Index (TI) was estimated as the ratio between LC50 and EC50.”
	Metric 22:	Reporting of Data	Medium	Results per treatment concentration were not reported at the level of the replicate, however, the paper has sufficient data presented in the narrative, supplemental materials, and figures/tables.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This form is for embryo mortality within the study. The data are presented within Figure 1 (page 5/11), Table 1 (page 6/11), and Supplemental Table 6 (6/8).

Overall Quality Determination High

Study Citation:	Alzualde, A., Behl, M., Sipes, N. S., Hsieh, J. H., Alday, A., Tice, R. R., Paules, R. S., Muriana, A., Quevedo, C. (2018). Toxicity profiling of flame retardants in zebrafish embryos using a battery of assays for developmental toxicity, neurotoxicity, cardiotoxicity and hepatotoxicity toward human relevance. Neurotoxicology and Teratology 70:40-50.
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*; cmlc2:CopGFP; Embryo
Health Outcome:	Cardiovascular
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5164137

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical referred to by Name, CASRN, and Structure in Supplemental file (word file).
	Metric 2:	Test Substance Source	Low	Source and Lot number were reported. But the test substance identity was NOT analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Purity was reported from manufacturer (Albermarle) as 99%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Solvent controls were used and consisted of 0.5% DMSO.
	Metric 5:	Negative Control Response	High	Control responses for cardiotoxicity are reported in Figure 3B (page 8/11).
	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	DMSO was used as the solvent and the same stock solution for used to prepare all exposure solutions. Experiments were conducted in well plates. Treatment solutions were replaced at day two of the 4 day exposure.
	Metric 8:	Consistency of Exposure Administration	High	Exposure administration appear to be consistent across treatment groups and controls.
	Metric 9:	Measurement of Test Substance Concentration	Medium	Exposure concentrations were not measured and are reported as nominal. Treatment solutions were replaced at day two of the 4 day exposure.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was for 2 and 4 days post-fertilization and was appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	5 exposure groups were used. Exposure concentrations were both under and over solubility at 1,3, 10, 30, 100 uM. This represented a broad range of treatment concentrations for testing effects.
	Metric 12:	Testing at or Below Solubility Limit	Medium	Concentrations >10 uM are over solubility listed in the final scope at 4.1 mg/L.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Zebrafish husbandry, egg production, collection and housing of embryos were described well.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized and/or whether pretreatment conditions were the same for control and exposed groups. Overall, water quality parameters were not well detailed for this study.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	5 embryos per well and 15 embryos per concentration were used for the main experiment. This experiment was conducted twice.

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Study Citation:	Alzualde, A., Behl, M., Sipes, N. S., Hsieh, J. H., Alday, A., Tice, R. R., Paules, R. S., Muriana, A., Quevedo, C. (2018). Toxicity profiling of flame retardants in zebrafish embryos using a battery of assays for developmental toxicity, neurotoxicity, cardiotoxicity and hepatotoxicity toward human relevance. Neurotoxicology and Teratology 70:40-50.
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*; cmlc2:CopGFP; Embryo
Health Outcome:	Cardiovascular
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5164137

Domain	Metric		Rating	Comments
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Environmental conditions during exposure were not reported.
	Metric 17:	Outcome Assessment Methodology	High	Section 2.3.5 reports the outcome assessment for heart rate and malformations.
	Metric 18:	Consistency of Outcome Assessment	High	Outcome assessment was consistent across treatment groups and controls.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information on outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	”As heartbeat followed a non-Gaussian distribution (skewness value -0.9, $p < 0{,}001$), a Mann-Whitney U test was applied to compare treated versus control groups.”
	Metric 22:	Reporting of Data	Medium	Specific results on cardiotoxicity at the individual and treatment level are not provided but table 3 and figure 4 report results at Lowest Effect Levels.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This form is for Cardiotoxicity and results are presented in Figure 3 (page 8/11) and Table 3 (page 8/11) and Supplemental Table 8 (8/8).

Overall Quality Determination High

Study Citation:	Alzualde, A., Behl, M., Sipes, N. S., Hsieh, J. H., Alday, A., Tice, R. R., Paules, R. S., Muriana, A., Quevedo, C. (2018). Toxicity profiling of flame retardants in zebrafish embryos using a battery of assays for developmental toxicity, neurotoxicity, cardiotoxicity and hepatotoxicity toward human relevance. Neurotoxicology and Teratology 70:40-50.
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*; cmlc2:CopGFP; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5164137

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical referred to by Name, CASRN, and Structure in Supplemental file (word file).
	Metric 2:	Test Substance Source	Low	Source and Lot number were reported but the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Purity was reported from manufacturer (Albermarle) as 99%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Solvent controls were used and consisted of 0.5% DMSO.
	Metric 5:	Negative Control Response	High	Control responses are reported for developmental toxicity graphically in Figure 1 (page 5/11).
	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	DMSO was used as the solvent and the same stock solution for used to prepare all exposure solutions. Experiments were conducted in well plates. Treatment solutions were replaced at day two of the 4 day exposure.
	Metric 8:	Consistency of Exposure Administration	High	Exposure administration appear to be consistent across treatment groups and controls.
	Metric 9:	Measurement of Test Substance Concentration	Medium	Exposure concentrations were not measured and are reported as nominal. Treatment solutions were replaced at day two of the 4 day exposure.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was for 2 and 4 days post-fertilization and was appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	8 exposure groups were used based on the dose-range finding study.
	Metric 12:	Testing at or Below Solubility Limit	Medium	The two highest concentrations (10 and 20 uM) were above the solubility limit for this compound as listed in the final scope (4.1 mg/L). All hazard values listed for these experiments are under the solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Zebrafish husbandry, egg production, collection and housing of embryos were described well.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized and/or whether pretreatment conditions were the same for control and exposed groups. Overall, water quality parameters were not well detailed for this study.

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Study Citation:	Alzualde, A., Behl, M., Sipes, N. S., Hsieh, J. H., Alday, A., Tice, R. R., Paules, R. S., Muriana, A., Quevedo, C. (2018). Toxicity profiling of flame retardants in zebrafish embryos using a battery of assays for developmental toxicity, neurotoxicity, cardiotoxicity and hepatotoxicity toward human relevance. Neurotoxicology and Teratology 70:40-50.
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*; cmlc2:CopGFP; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5164137

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Medium	5 embryos per well and 15 embryos per concentration were used for the main experiment. This experiment was conducted twice.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Environmental conditions during exposure were not reported.
	Metric 17:	Outcome Assessment Methodology	High	Section 2.3.1 lists the criteria for morphological analysis of embryo developmental toxicity.
	Metric 18:	Consistency of Outcome Assessment	High	Outcome assessment was consistent across treatment groups and controls.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information on outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	EC50 were estimated by non linear regression in graphpad prism
	Metric 22:	Reporting of Data	Medium	Specific results on developmental toxicity per treatment concentration were not reported at the level of the replicate, however, the paper has sufficient data presented in the narrative, supplemental materials, and figures/tables.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This form is for developmental toxicity. The EC50 values and variance components for these studies are presented in Table 1 (page 6/11), Supplemental Table 6 (page 6/8), and Figure 1 (page 5/11).

Overall Quality Determination High

Study Citation:	Alzualde, A., Behl, M., Sipes, N. S., Hsieh, J. H., Alday, A., Tice, R. R., Paules, R. S., Muriana, A., Quevedo, C. (2018). Toxicity profiling of flame retardants in zebrafish embryos using a battery of assays for developmental toxicity, neurotoxicity, cardiotoxicity and hepatotoxicity toward human relevance. Neurotoxicology and Teratology 70:40-50.
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*; Wild Type AB; Embryo
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5164137

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical referred to by Name, CASRN, and Structure in Supplemental file (word file).
	Metric 2:	Test Substance Source	Low	Source and Lot number were reported but the test substance identity was NOT analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Purity was reported from manufacturer (Albermarle) as 99%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Solvent controls were used and consisted of 0.5% DMSO.
	Metric 5:	Negative Control Response	High	Control responses for behavior are presented across timepoints of observation in Supplemental table 7 (page 7/8), Supplemental figure 2 (page 2/3).
	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	DMSO was used as the solvent and the same stock solution for used to prepare all exposure solutions. Experiments were conducted in well plates. Treatment solutions were replaced at day two of the 4 day exposure.
	Metric 8:	Consistency of Exposure Administration	High	Exposure administration appear to be consistent across treatment groups and controls.
	Metric 9:	Measurement of Test Substance Concentration	Medium	Exposure concentrations were not measured and are reported as nominal. Treatment solutions were replaced at day two of the 4 day exposure.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was for 2 and 4 days post-fertilization and was appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Two experiments with similar concentrations were conducted. Ranges for experiment 1 are: 0.1, 0.3, 0.6, 1.5 and 3 uM, and Experiment 2 were 0.15, 0.3, 0.6, 1 and 1.5 uM.
	Metric 12:	Testing at or Below Solubility Limit	High	All treatment concentrations were under solubility listed in the final scope at 4.1 mg/L.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Zebrafish husbandry, egg production, collection and housing of embryos were described well.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized and/or whether pretreatment conditions were the same for control and exposed groups. Overall, water quality parameters were not well detailed for this study.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	No TGs for sample size on behavior studies but 16 embryos per concentration is similar to other studies on movement metrics.

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Study Citation:	Alzualde, A., Behl, M., Sipes, N. S., Hsieh, J. H., Alday, A., Tice, R. R., Paules, R. S., Muriana, A., Quevedo, C. (2018). Toxicity profiling of flame retardants in zebrafish embryos using a battery of assays for developmental toxicity, neurotoxicity, cardiotoxicity and hepatotoxicity toward human relevance. Neurotoxicology and Teratology 70:40-50.
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*; Wild Type AB; Embryo
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5164137

Domain	Metric		Rating	Comments
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Environmental conditions during exposure were not reported.
	Metric 17:	Outcome Assessment Methodology	High	Section 2.3.3 reports the outcome assessment for movement metrics.
	Metric 18:	Consistency of Outcome Assessment	High	Outcome assessment was consistent across treatment groups and controls.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information on outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	”The mean of the total distance moved by embryos in each group was measured in two-minute time bins and treated versus control groups were compared using unpaired Student’s t-test.”
	Metric 22:	Reporting of Data	Medium	Specific results on cardiotoxicity at the individual and treatment level are not provided but table 3 and figure 4 report results at Lowest Effect Levels.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This form is for the behavior/movement data presented in Table 2 (page 7/11), Supplemental table 7 (page 7/8), Supplemental figure 2 (page 2/3). TBBPA did not influence light or dark movement metrics in these fish at any concentrations.

Overall Quality Determination High

Study Citation:	Huang, G. Y., Ying, G. G., Liang, Y. Q., Zhao, J. L., Yang, B., Liu, S., Liu, Y. S. (2013). Hormonal effects of tetrabromobisphenol A using a combination of in vitro and in vivo assays. Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 157(4):344-351.
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; *Gambusia affinis*; Juvenile
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547473

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical name was provided (tetrabromobisphenol A (TBBPA)), but no CASRN, structure, or other chemical descriptors were reported.
	Metric 2:	Test Substance Source	Low	Source was reported (Sigma-Aldrich, USA), but identity was not analytically verified.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of TBBPA were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	A solvent control group (0.002% DMSO) was used.
	Metric 5:	Negative Control Response	High	Gene expression (mRNA) was reported and adequate (Figure 4).
	Metric 6:	Randomized Allocation	Medium	Authors reported that healthy adult male mosquitofish werechosen at random and transferred to experimental aquaria (section 2.4).
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The study lacked some details in the preparation of exposure solutions. Authors reported using 0.002% DMSO and that experiment was performed under the static condition and water replaced with freshly prepared treatment water every other day. However, they did not describe how the stock solution or treatment solutions were prepared.
	Metric 8:	Consistency of Exposure Administration	High	Evidence showed that exposure administration was consistent across groups.
	Metric 9:	Measurement of Test Substance Concentration	Medium	Exposure concentrations were measured by HPLC-DAD (Table 1).
	Metric 10:	Exposure Duration and Frequency	High	Exposure duration (60 d) was reported and appropriate for the study type and endpoint (evaluation of TBBPA effects on gene expression).
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	The goal was not to determine a dose-dependent effect. The number of exposure groups (juveniles: 0 and 100 nM) was adequate.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were below the solubility limit (highest concentration was 100 nM = 0.054 mg/L TBBPA and the water solubility of TBBPA is 4.15 mg/L at 25 C).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The organisms (mosquitofish) are well described and appropriate for the outcome. Juvenile mosquitofish (approximately 5 days old) were obtained by an artificial fertilization method.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	For the adult experiments, the authors explicitly reported that fish were acclimated for at least three weeks. However, the authors did not explicitly indicate if the juvenile fish were acclimated.

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Study Citation:	Huang, G. Y., Ying, G. G., Liang, Y. Q., Zhao, J. L., Yang, B., Liu, S., Liu, Y. S. (2013). Hormonal effects of tetrabromobisphenol A using a combination of in vitro and in vivo assays. Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 157(4):344-351.
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; *Gambusia affinis*; Juvenile
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547473

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The number of fish and replicates exposed to the chemical were reported and sufficient (20 juvenile fish and three replicates per treatment group); 8 juvenile fish were collected per group for RNA extraction and gene expression analysis.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Housing and maintenance conditions were reported and appropriate (aerated freshwater at 23–25 °C, natural photoperiod, fed with flake red worms twice daily, residual bait and feces removed daily).
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology (real-time PCR) was reported and appropriate for the intended outcome of interest (TBBPA effects on gene expression of steroid hormones).
	Metric 18:	Consistency of Outcome Assessment	High	As reported by authors, outcome was assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among study groups in exposure conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There is no information to suggest differences among groups of juvenile fish.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical differences between experimental groups for each gene expression were determined by one-way analysis of variance (ANOVA), followed by Duncan's multiple comparison tests.
	Metric 22:	Reporting of Data	High	Results were presented for both treatments groups (Figure 4, section 3.3).
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This form represents mRNA expression in juveniles and is located in Figure 2 and 4 and within the results section 3.2 and 3.3 on pages 3-4/8. The goal of the study was to assess hormonal activities of TBBPA using [1] in vitro recombinant yeast estrogen screen (YES) and yeast androgen screen (YAS) assays and [2] an in vivo model to investigate expression patterns of Vtg, ERα, ERβ, ARα and ARβ genes in adult male and juvenile mosquitofish, as we all the effects of TBBPA on histological changes of the liver and testis of adult male mosquitofish. As per our PECO, yeast studies, except for assessing genotoxicity or mutagenicity, are EXCLUDED; so the in vitro yeast assays of the study were not evaluated. This form was used to evaluate the effects of TBBPA on expression patterns of Vtg, ERα, ERβ, ARα and ARβ genes in juvenile (sexually immature) mosquitofish.

Overall Quality Determination High

Study Citation:	Huang, G. Y., Ying, G. G., Liang, Y. Q., Zhao, J. L., Yang, B., Liu, S., Liu, Y. S. (2013). Hormonal effects of tetrabromobisphenol A using a combination of in vitro and in vivo assays. Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 157(4):344-351.
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; *Gambusia affinis*; Adult
Health Outcome:	Hepatic/Liver
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547473

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical name was provided (tetrabromobisphenol A (TBBPA)), but no CASRN, structure, or other chemical descriptors were reported.
	Metric 2:	Test Substance Source	Low	Source was reported (Sigma-Aldrich, USA), but identity was not analytically verified.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of TBBPA were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	A solvent control group (0.002% DMSO) was used.
	Metric 5:	Negative Control Response	Medium	Histological examination of the morphology of the liver was reported and adequate. A representative control was shown in Figure 5.
	Metric 6:	Randomized Allocation	Medium	Authors reported that healthy adult male mosquitofish werechosen at random and transferred to experimental aquaria (section 2.4).
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The study lacked some details in the preparation of exposure solutions. Authors reported using 0.002% DMSO and that experiment was performed under the static condition and water replaced with freshly prepared treatment water every other day. However, they did not describe how the stock solution or treatment solutions were prepared.
	Metric 8:	Consistency of Exposure Administration	High	Evidence showed that exposure administration was consistent across groups.
	Metric 9:	Measurement of Test Substance Concentration	Medium	Exposure concentrations were measured by HPLC-DAD (Table 1). These data show one initial and 48 hour sampling. Samples were not conducted throughout the exposure period.
	Metric 10:	Exposure Duration and Frequency	High	Exposure duration (60 d) was reported and appropriate for the study type and endpoint (evaluation of TBBPA effects on liver tissue morphology).
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing (adults: 0, 50, 250 and 500 nM) were adequate.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were below the solubility limit (highest concentration was 500 nM = 0.272 mg/L TBBPA and the water solubility of TBBPA is 4.15 mg/L at 25 C).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The organisms (mosquitofish) are well described and appropriate for the outcome. However, the fish were captured from a river (the Liuxi River, reported as an area protected from pollution), and thus while the fish were acclimated for at least three weeks prior to exposure, there are uncertainties about the fitness/underlying conditions of field fish.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Fish were acclimated for at least three weeks prior to exposure.

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Study Citation:	Huang, G. Y., Ying, G. G., Liang, Y. Q., Zhao, J. L., Yang, B., Liu, S., Liu, Y. S. (2013). Hormonal effects of tetrabromobisphenol A using a combination of in vitro and in vivo assays. Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 157(4):344-351.
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; *Gambusia affinis*; Adult
Health Outcome:	Hepatic/Liver
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547473

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The number of fish and replicates exposed to the chemical were reported and sufficient (12 adult males or 20 juveniles and three replicates per treatment group). For the histological analysis of the liver and testis, two adult male fish per replicate tank were used.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Housing and maintenance conditions were reported and appropriate (aerated freshwater at 23–25 °C, natural photoperiod, fed with flake red worms twice daily, residual bait and feces removed daily).
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology (histological analysis using light microscopy) was reported and appropriate for the intended outcome of interest (TBBPA effects on liver morphology).
	Metric 18:	Consistency of Outcome Assessment	High	As reported by authors, outcome was assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among study groups in environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	Fish were captured in a river and while they were acclimated for at least three weeks prior to exposure, there are uncertainties about the unknown underlaying health status that could contribute to variation in responses from chemical exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	N/A	Statistical analysis of such photographic observations are often not performed for histology. Results consisted of histopathological imaging.
	Metric 22:	Reporting of Data	Low	Adult fish were exposed to 0, 50, 250 and 500 nM TBBPA and tissues collected (section 2.4), but in the text (section 3.4) and Figure 5 authors only presented results and images of the 0 and 500 nM TBBPA groups.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This form represents liver histology and is located on Figure 5 (page 6/8) and section 3.4 on page 4/8. The goal of the study was to assess hormonal activities of TBBPA using [1] in vitro recombinant yeast estrogen screen (YES) and yeast androgen screen (YAS) assays and [2] an in vivo model to investigate expression patterns of Vtg, ERα, ERβ, ARα and ARβ genes in adult male and juvenile mosquitofish, as we all the effects of TBBPA on histological changes of the liver and testis of adult male mosquitofish. As per our PECO, yeast studies, except for assessing genotoxicity or mutagenicity, are EXCLUDED; so the in vitro yeast assays of the study were not evaluated. This form was used to evaluate the effects of TBBPA on histological changes of the liver of adult male mosquitofish.

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Study Citation:	Huang, G. Y., Ying, G. G., Liang, Y. Q., Zhao, J. L., Yang, B., Liu, S., Liu, Y. S. (2013). Hormonal effects of tetrabromobisphenol A using a combination of in vitro and in vivo assays. Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 157(4):344-351.
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; *Gambusia affinis*; Adult
Health Outcome:	Hepatic/Liver
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547473

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

Study Citation:	Huang, G. Y., Ying, G. G., Liang, Y. Q., Zhao, J. L., Yang, B., Liu, S., Liu, Y. S. (2013). Hormonal effects of tetrabromobisphenol A using a combination of in vitro and in vivo assays. Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 157(4):344-351.
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; *Gambusia affinis*; Adult
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547473

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical name was provided (tetrabromobisphenol A (TBBPA)), but no CASRN, structure, or other chemical descriptors were reported.
	Metric 2:	Test Substance Source	Low	Source was reported (Sigma-Aldrich, USA), but identity was not analytically verified.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of TBBPA were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	A solvent control group (0.002% DMSO) was used.
	Metric 5:	Negative Control Response	High	Gene expression (mRNA) was reported and adequate (Figure 2, 3).
	Metric 6:	Randomized Allocation	Medium	Authors reported that healthy adult male mosquitofish werechosen at random and transferred to experimental aquaria (section 2.4).
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The study lacked some details in the preparation of exposure solutions. Authors reported using 0.002% DMSO and that experiment was performed under the static condition and water replaced with freshly prepared treatment water every other day. However, they did not describe how the stock solution or treatment solutions were prepared.
	Metric 8:	Consistency of Exposure Administration	High	Evidence showed that exposure administration was consistent across groups.
	Metric 9:	Measurement of Test Substance Concentration	Medium	Exposure concentrations were measured by HPLC-DAD (Table 1).
	Metric 10:	Exposure Duration and Frequency	High	Exposure duration (60 d) was reported and appropriate for the study type and endpoint (evaluation of TBBPA effects on gene expression).
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing (adults: 0, 50, 250 and 500 nM) were adequate.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were below the solubility limit (highest concentration was 500 nM = 0.272 mg/L TBBPA and the water solubility of TBBPA is 4.15 mg/L at 25 C).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The organisms (mosquitofish) are well described and appropriate for the outcome. However, the fish were captured from a river (the Liuxi River, reported as an area protected from pollution), and thus while the fish were acclimated for at least three weeks prior to exposure, there are uncertainties about the fitness/underlying conditions of field fish.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Fish were acclimated for at least three weeks prior to exposure.

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Study Citation:	Huang, G. Y., Ying, G. G., Liang, Y. Q., Zhao, J. L., Yang, B., Liu, S., Liu, Y. S. (2013). Hormonal effects of tetrabromobisphenol A using a combination of in vitro and in vivo assays. Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 157(4):344-351.
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; *Gambusia affinis*; Adult
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547473

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The number of fish and replicates exposed to the chemical were reported and sufficient (12 adult males and three replicates per treatment group). Livers and testes from 5 male individuals per group were dissected for RNA extraction and gene expression analysis.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Housing and maintenance conditions were reported and appropriate (aerated freshwater at 23–25 °C, natural photoperiod, fed with flake red worms twice daily, residual bait and feces removed daily).
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology (real-time PCR) was reported and appropriate for the intended outcome of interest (TBBPA effects on gene expression of steroid hormone receptors).
	Metric 18:	Consistency of Outcome Assessment	High	As reported by authors, outcome was assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among study groups in exposure conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	Fish were captured in a river and while they were acclimated for at least three weeks prior to exposure, there are uncertainties about the unknown underlaying health status that could contribute to variation in exposure responses.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical differences between experimental groups for each gene expression were determined by one-way analysis of variance (ANOVA), followed by Duncan's multiple comparison tests.
	Metric 22:	Reporting of Data	High	Results were presented for all treatments groups (Figure 3, section 3.3).
	Metric 23:	Explanation of Unexpected Outcomes	Medium	There were no unexpected outcomes.

Additional Comments: This form represents mechanistic data of mRNA expression in adults. These data are presented in figure 2 and figure 3 and sections 3.2 and 3.3 on pages 3-4/8. The goal of the study was to assess hormonal activities of TBBPA using [1] in vitro recombinant yeast estrogen screen (YES) and yeast androgen screen (YAS) assays and [2] an in vivo model to investigate expression patterns of Vtg, ERα, ERβ, ARα and ARβ genes in adult male and juvenile mosquitofish, as we all the effects of TBBPA on histological changes of the liver and testis of adult male mosquitofish. As per our PECO, yeast studies, except for assessing genotoxicity or mutagenicity, are EXCLUDED; so the in vitro yeast assays of the study were not evaluated. This form was used to evaluate the effects of TBBPA on expression patterns of Vtg, ERα, ERβ, ARα and ARβ genes in adult male mosquitofish.

Overall Quality Determination High

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Study Citation:	Huang, G. Y., Ying, G. G., Liang, Y. Q., Zhao, J. L., Yang, B., Liu, S., Liu, Y. S. (2013). Hormonal effects of tetrabromobisphenol A using a combination of in vitro and in vivo assays. Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 157(4):344-351.
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; *Gambusia affinis*; Adult
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547473

Domain	Metric	Rating	Comments

Study Citation:	Huang, G. Y., Ying, G. G., Liang, Y. Q., Zhao, J. L., Yang, B., Liu, S., Liu, Y. S. (2013). Hormonal effects of tetrabromobisphenol A using a combination of in vitro and in vivo assays. Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 157(4):344-351.
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; *Gambusia affinis*; Adult
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547473

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical name was provided (tetrabromobisphenol A (TBBPA)), but no CASRN, structure, or other chemical descriptors were reported.
	Metric 2:	Test Substance Source	Low	Source was reported (Sigma-Aldrich, USA), but identity was not analytically verified.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of TBBPA were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	A solvent control group (0.002% DMSO) was used.
	Metric 5:	Negative Control Response	Medium	Histological examination of the morphology of the testis was reported and adequate. A representative control was shown in Figure 5.
	Metric 6:	Randomized Allocation	Medium	Authors reported that healthy adult male mosquitofish werechosen at random and transferred to experimental aquaria (section 2.4).
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The study lacked some details in the preparation of exposure solutions. Authors reported using 0.002% DMSO and that experiment was performed under the static condition and water replaced with freshly prepared treatment water every other day. However, they did not describe how the stock solution or treatment solutions were prepared.
	Metric 8:	Consistency of Exposure Administration	High	Evidence showed that exposure administration was consistent across groups.
	Metric 9:	Measurement of Test Substance Concentration	Medium	Exposure concentrations were measured by HPLC-DAD (Table 1).
	Metric 10:	Exposure Duration and Frequency	High	Exposure duration (60 d) was reported and appropriate for the study type and endpoint (evaluation of TBBPA effects on testis tissue morphology).
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing (adults: 0, 50, 250 and 500 nM) were adequate.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were below the solubility limit (highest concentration was 500 nM = 0.272 mg/L TBBPA and the water solubility of TBBPA is 4.15 mg/L at 25 C).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The organisms (mosquitofish) are well described and appropriate for the outcome. However, the fish were captured from a river (the Liuxi River, reported as an area protected from pollution), and thus while the fish were acclimated for at least three weeks prior to exposure, there are uncertainties about the fitness/underlying conditions of field fish.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Fish were acclimated for at least three weeks prior to exposure.

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Study Citation:	Huang, G. Y., Ying, G. G., Liang, Y. Q., Zhao, J. L., Yang, B., Liu, S., Liu, Y. S. (2013). Hormonal effects of tetrabromobisphenol A using a combination of in vitro and in vivo assays. Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 157(4):344-351.
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; *Gambusia affinis*; Adult
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547473

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The number of fish and replicates exposed to the chemical were reported and sufficient (12 adult males and three replicates per treatment group). For the histological analysis of the liver and testis, two adult male fish per replicate tank were used.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Housing and maintenance conditions were reported and appropriate (aerated freshwater at 23–25 °C, natural photoperiod, fed with flake red worms twice daily, residual bait and feces removed daily).
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology (histological analysis using light microscopy) was reported and appropriate for the intended outcome of interest (TBBPA effects on testis tissue morphology).
	Metric 18:	Consistency of Outcome Assessment	High	As reported by authors, outcome was assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among study groups in exposure conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	Fish were captured in a river and while they were acclimated for at least three weeks prior to exposure, there are uncertainties about the unknown underlaying health status that could contribute to variation in exposure responses.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	N/A	Statistical analysis of such photographic observations are often not performed for histology. Results consisted of histopathological imaging.
	Metric 22:	Reporting of Data	Low	Adult fish were exposed to 0, 50, 250 and 500 nM TBBPA and tissues collected (section 2.4), but in the text (section 3.4) and Figure 5 authors only presented results and images of the 0 and 500 nM TBBPA groups.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This form represents testis histology from treatment exposures and is in the paper in figure 5 on page 6/8 and in the results section 3.4 on page 4/8. The goal of the study was to assess hormonal activities of TBBPA using [1] in vitro recombinant yeast estrogen screen (YES) and yeast androgen screen (YAS) assays and [2] an in vivo model to investigate expression patterns of Vtg, ERα, ERβ, ARα and ARβ genes in adult male and juvenile mosquitofish, as we all the effects of TBBPA on histological changes of the liver and testis of adult male mosquitofish. As per our PECO, yeast studies, except for assessing genotoxicity or mutagenicity, are EXCLUDED; so the in vitro yeast assays of the study were not evaluated. This form was used to evaluate the effects of TBBPA on histological changes of the testis of adult male mosquitofish.

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Study Citation:	Huang, G. Y., Ying, G. G., Liang, Y. Q., Zhao, J. L., Yang, B., Liu, S., Liu, Y. S. (2013). Hormonal effects of tetrabromobisphenol A using a combination of in vitro and in vivo assays. Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 157(4):344-351.
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; *Gambusia affinis*; Adult
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547473

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

Study Citation:	Union Carbide Corporation Environmental Services, (1978). Acute toxicity of FMBP4A (tetrabromobisphenol A) to the bluegill sunfish, Lepomis macrochirus rafinesque.
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*; Rafinesque; Juvenile
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5549811

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The chemical name and CASRN were provided. Lot No. and form of the test substance (white powder) were also mentioned.
	Metric 2:	Test Substance Source	Low	The source was not reported and the test substance was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/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 groups, both water and solvent control.
	Metric 5:	Negative Control Response	High	The biological responses of the negative control groups were 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	Medium	The experimental system and test media preparation methods were adequately reported but did not completely account for physical-chemical properties. Tests were conducted in 5 gallon glass jars with 15 liters of water. Acetone was used as the solvent. Concentrations were not measured during the study.
	Metric 8:	Consistency of Exposure Administration	High	Exposure administration appears to be consistent across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	Acute fish toxicity study of 96-hr duration.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	5 exposure groups were used and were adequate to address the purpose of the study.
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate ((i.e., no effects on biological responses were observed in the solvent control and no interactions were expected between the solvent and test substance).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were obtained from a commercial hatchery in Nebraska. The test organisms were 6 months old and mean length and weight were reported
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to the test water for 24 hours prior to testing.

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Study Citation:	Union Carbide Corporation Environmental Services, (1978). Acute toxicity of FMBP4A (tetrabromobisphenol A) to the bluegill sunfish, Lepomis macrochirus rafinesque.
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*; Rafinesque; Juvenile
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5549811

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Low	10 organisms were used per test concentration but replicates were not included in the experiment.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Organism housing and biological loading (0.39g/l; below OECD max loading of 0.8g wet wt. fish/L) was appropriate. Measured dissolved oxygen concentration in few exposure groups were low compared to controls (Page 8/10).
	Metric 17:	Outcome Assessment Methodology	Uninformative	Behavior metrics are observational and not scored by any described metrics.
	Metric 18:	Consistency of Outcome Assessment	High	Behavior outcome was assessed consistently across study groups at 24 hour intervals.
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. Dissolved oxygen (DO) results were very low in few exposure groups at 48 and 96 hrs but since replicates were not included in the experiment, statistical tests could not be conducted to test whether DO played a role in observed toxicity.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure (e.g., infection) that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Uninformative	Statistical analysis was not necessary for behavior outcome- qualitative results.
	Metric 22:	Reporting of Data	Low	Qualitative results were described in the text.
	Metric 23:	Explanation of Unexpected Outcomes	Low	Insufficient information was provided to determine if excessive variability or unexpected outcomes occurred.

Additional Comments: No replicates were included in the experiment. Dissolved oxygen levels were not maintained in the treatment groups compared to controls. Qualitative results for behavior outcome was described in the text. The 96 hr no effect level was 0.18 mg/l.

Overall Quality Determination Uninformative

Study Citation:	Union Carbide Corporation Environmental Services, (1978). Acute toxicity of FMBP4A (tetrabromobisphenol A) to the bluegill sunfish, Lepomis macrochirus rafinesque.
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*; Rafinesque; Juvenile
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5549811

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The chemical name and CASRN were provided. Lot No. and form of the test substance (white powder) were also mentioned.
	Metric 2:	Test Substance Source	Low	The source was not reported and the test substance was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/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 groups, both water and solvent control.
	Metric 5:	Negative Control Response	High	The biological responses of the negative control groups were 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	Medium	The experimental system and test media preparation methods were adequately reported but did not completely account for physical-chemical properties. Tests were conducted in 5 gallon glass jars with 15 liters of water. Acetone was used as the solvent. Concentrations were not measured during the study.
	Metric 8:	Consistency of Exposure Administration	High	Exposure administration appears to be consistent across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	Acute fish toxicity study of 96-hr duration.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	5 exposure groups were used and were adequate to address the purpose of the study.
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate ((i.e., no effects on biological responses were observed in the solvent control and no interactions were expected between the solvent and test substance).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were obtained from a commercial hatchery in Nebraska. The test organisms were 6 months old and mean length and weight were reported
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to the test water for 24 hours prior to testing.
	Metric 15:	Number of Organisms and Replicates per Group	Low	10 organisms were used per test concentration but replicates were not included in the experiment.

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Study Citation:	Union Carbide Corporation Environmental Services, (1978). Acute toxicity of FMBP4A (tetrabromobisphenol A) to the bluegill sunfish, Lepomis macrochirus rafinesque.
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*; Rafinesque; Juvenile
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5549811

Domain	Metric		Rating	Comments
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Organism housing and biological loading (0.39g/l; below OECD max loading of 0.8g wet wt. fish/L) was appropriate. Measured dissolved oxygen concentration in few exposure groups were low compared to controls (Page 8/10).
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome of interest.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups at 24, 48 and 96 hrs.
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. Dissolved oxygen (DO) results were very low in few exposure groups at 48 and 96 hrs but since replicates were not included in the experiment, statistical tests could not be conducted to test whether DO played a role in observed toxicity.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure (e.g., infection) that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately. LC50 value and confidence intervals were derived but statistical methods used were not 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	Insufficient information was provided to determine if excessive variability or unexpected outcomes occurred.

Additional Comments: No replicates were included in the experiment. Dissolved oxygen levels were not maintained in the treatment groups. LC 50 value and confidence interval were reported for 24 48 and 96 hours.

Overall Quality Determination Medium

Study Citation:	Stoner Labs, (1978). Bioaccumulation of tetrabromobisphenol, in the bluegill sunfish.
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; *Lepomis macrochirus*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5546499

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity of test substance was not reported but specific activity was reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	A different solvent was used in the control than in the treatment group (page 8/12).
	Metric 5:	Negative Control Response	Medium	The mortality of the negative control group was reported and higher than treated fish. The control group experienced 9.2% mortality over the study while the TBBPA group reported 2.5% cumulative mortality.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups per treatment tanks. Total number of animals were reported on page 5/12.
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 did not completely account for physical-chemical properties, ie slime build up in tanks.
	Metric 8:	Consistency of Exposure Administration	Low	Details of exposure administration were reported, but inconsistencies in administration such as changing supply tubing may impact results.
	Metric 9:	Measurement of Test Substance Concentration	Medium	Exposure concentrations were measured but controls were not reported.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	only one concentration tested, not looking for a dose response.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations (0.0098 mg/L) were at or below the water solubility limit (4.1 mg/L).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	There are minor reservations or uncertainties about the size range of test organisms (0.5 - 2.0 g) and no age was reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.
	Metric 15:	Number of Organisms and Replicates per Group	Low	Large number of fish exposed but no replicates were used. Water quality parameters throughout this exposure period were not reported.

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Study Citation:	Stoner Labs, (1978). Bioaccumulation of tetrabromobisphenol, in the bluegill sunfish.
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; *Lepomis macrochirus*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5546499

Domain	Metric		Rating	Comments
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate, feeding rates or type of food not reported.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed the intended outcomes.
	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	Low	Reported information indicated that one or more study groups experienced disproportionate test organism attrition.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	N/A	Only one concentration with bioaccumulation being the priority for reported data.
	Metric 22:	Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group.
	Metric 23:	Explanation of Unexpected Outcomes	Low	Unclear how bacterial accumulation on surfaces affected control or treated fish

Additional Comments: None

Overall Quality Determination Low

Study Citation:	Stoner Labs, (1978). Bioaccumulation of tetrabromobisphenol, in the bluegill sunfish.
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; *Lepomis macrochirus*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5546499

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity of test substance was not reported but specific activity was reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	A different solvent was used in the control than in the treatment group (page 8/12).
	Metric 5:	Negative Control Response	Medium	The mortality of the negative control group was reported and higher than treated fish. The control group experienced 9.2% mortality over the study while the TBBPA group reported 2.5% cumulative mortality.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups per treatment tanks. Total number of animals were reported on page 5/12.
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 did not completely account for physical-chemical properties, ie slime build up in tanks.
	Metric 8:	Consistency of Exposure Administration	Low	Details of exposure administration were reported, but inconsistencies in administration such as changing supply tubing may impact results.
	Metric 9:	Measurement of Test Substance Concentration	Medium	Exposure concentrations were measured but controls were not reported.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	Only one concentration was tested, not looking for a dose response.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations (0.0098 mg/L) were at or below the water solubility limit (4.1 mg/L).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	There are minor reservations or uncertainties about the size range of test organisms (0.5 - 2.0 g) and no age was reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.
	Metric 15:	Number of Organisms and Replicates per Group	Low	Large number of fish exposed but no replicates were used. Water quality parameters throughout this exposure period were not reported.
Domain 5: Outcome Assessment				

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Study Citation:	Stoner Labs, (1978). Bioaccumulation of tetrabromobisphenol, in the bluegill sunfish.
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; *Lepomis macrochirus*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5546499

Domain	Metric		Rating	Comments
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate, feeding rates or type of food not reported.
	Metric 17:	Outcome Assessment Methodology	Low	Mortality assessment was not mentioned in the methods.
	Metric 18:	Consistency of Outcome Assessment	Low	Since methods were not reported, consistency is unclear.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Low	Reported information indicated that one or more study groups experienced disproportionate test organism attrition.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	N/A	Only one concentration with bioaccumulation being the priority for reported data.
	Metric 22:	Reporting of Data	High	Mortality data was shown in Table II.
	Metric 23:	Explanation of Unexpected Outcomes	Low	Unclear how bacterial accumulation on surfaces affected control or treated fish.

Additional Comments: None

Overall Quality Determination Low

Study Citation:	Debenest, T., Gagné, F., Petit, A. N., André, C., Kohli, M., Blaise, C. (2010). Ecotoxicity of a brominated flame retardant (tetrabromobisphenol A) and its derivatives to aquatic organisms. Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 152(4):407-412.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oncorhynchus mykiss*; in vitro hepatocyte test; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Cytotoxicity-Photosynthesis
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1294788

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%
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Calculation implies that a negative control was used
	Metric 5:	Negative Control Response	Low	The biological response of the negative control groups was not reported.
	Metric 6:	Randomized Allocation	Low	No random allocation reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
	Metric 8:	Consistency of Exposure Administration	Low	details of exposure administration were not provided
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured or measurements were not reported
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
	Metric 12:	Testing at or Below Solubility Limit	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test animals was not reported
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized
	Metric 15:	Number of Organisms and Replicates per Group	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Reported environmental conditions were not sufficient to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed

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Study Citation:	Debenest, T., Gagné, F., Petit, A. N., André, C., Kohli, M., Blaise, C. (2010). Ecotoxicity of a brominated flame retardant (tetrabromobisphenol A) and its derivatives to aquatic organisms. Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 152(4):407-412.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oncorhynchus mykiss*; in vitro hepatocyte test; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Cytotoxicity-Photosynthesis
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1294788

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22:	Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	Low	unexpected outcomes were not reported

Additional Comments: This evaluation form is for the in vitro test conducted using rainbow trout hepatocyte to assess for cytotoxicity after 48h.

Overall Quality Determination Low

Study Citation:	Ltd,, W.I. (2003). Tetrabromobisphenol A: A 96-hour flow-through acute toxicity test with the rainbow trout (Oncorhynchus mykiss). Final report.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oncorhynchus mykiss*; Juvenile
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	7019954

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	High	The source of the test substance was reported, The batch/lot number was identified and the chemical identity was verified by analytical methods.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.17%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using appropriate concurrent negative control groups.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes (Table 4, page 20).
	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 media preparation were described in adequate detail.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods (HPLC/MS, appendix 3).
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type, 96 hours.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	only two treatment concentrations were used.
	Metric 12:	Testing at or Below Solubility Limit	Medium	The solvent concentration was appropriate but some precipitate was reported.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions and all pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Low	Only two replicates were used (Table 4, page 20/57).
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:	Ltd,, W.I. (2003). Tetrabromobisphenol A: A 96-hour flow-through acute toxicity test with the rainbow trout (Oncorhynchus mykiss). Final report.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oncorhynchus mykiss*; Juvenile
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	7019954

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	No information was presented to indicate differences among groups due to animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described on page 14/57.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: Mortality results are presented on page 15/57 and in Table 4 on page 20/57.

Overall Quality Determination High

Study Citation:	EAG Laboratories, (2018). Tetrabromobisphenol A: Japanese medaka short-term reproduction assay (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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064189

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 a commercial 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 for assessed outcomes
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods
	Metric 10:	Exposure Duration and Frequency	High	An approved standard duration was used
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for assessing the desired endpoints
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were above the water solubility limit. DMF was used was used to solubilize test substance.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	four replicates with adequate number of organisms per treatment
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:	EAG Laboratories, (2018). Tetrabromobisphenol A: Japanese medaka short-term reproduction assay (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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064189

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 were no differences reported 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
	Metric 23:	Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained, only a couple of mortalities. $\leq$10% mortality in the controls.

Additional Comments: GSI, fecundity, fertility, eggs per female, papillary count

Overall Quality Determination High

Study Citation:	EAG Laboratories, (2018). Tetrabromobisphenol A: Japanese medaka short-term reproduction assay (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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064189

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 a commercial 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 for assessed outcomes
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods
	Metric 10:	Exposure Duration and Frequency	High	An approved standard duration was used
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for assessing the desired endpoints
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were above the water solubility limit. DMF was used was used to solubilize test substance.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	four replicates with adequate number of organisms per treatment
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). Tetrabromobisphenol A: Japanese medaka short-term reproduction assay (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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064189

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 were no differences reported 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
	Metric 23:	Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained, only a couple of mortalities. $\leq$10% mortality in the controls.

Additional Comments: None

Overall Quality Determination — High

Study Citation:	EAG Laboratories, (2018). Tetrabromobisphenol A: Japanese medaka short-term reproduction assay (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:	Endocrine
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064189

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 a commercial 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 for assessed outcomes.
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods.
	Metric 10:	Exposure Duration and Frequency	High	An approved standard duration was used.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for assessing the desired endpoints.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were above the water solubility limit. DMF was used to solubilize the test substance.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	There were four replicates with adequate number of organisms per treatment.
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 with details in Appendix 7.

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Study Citation:	EAG Laboratories, (2018). Tetrabromobisphenol A: Japanese medaka short-term reproduction assay (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:	Endocrine
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064189

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There were no differences reported among groups that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described.
	Metric 22:	Reporting of Data	Medium	Authors reported, "there were no TBBPA-related or background lesions identified in examined sections of thyroid." No data or figures were shown.
	Metric 23:	Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.

Additional Comments: This evaluation is for the histology assessment of thyroid tissue.

Overall Quality Determination High

Study Citation:	EAG Laboratories, (2018). Tetrabromobisphenol A: Japanese medaka short-term reproduction assay (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:	Hepatic/Liver
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064189

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 a commercial 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 the preparation of test media were described in adequate detail.
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods.
	Metric 10:	Exposure Duration and Frequency	High	An approved standard duration was used.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for assessing the desired endpoints.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were above the water solubility limit. DMF was used to solubilize the test substance.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	There were four replicates with adequate number of organisms per treatment..
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 in detail in Appendix 7.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.

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Study Citation:	EAG Laboratories, (2018). Tetrabromobisphenol A: Japanese medaka short-term reproduction assay (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:	Hepatic/Liver
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064189

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 were no differences reported 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 in Table 4 and Figures 7, 8, and 9.
	Metric 23:	Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.

Additional Comments: This evaluation is for the histology assessment of liver tissue.

Overall Quality Determination High

Study Citation:	EAG Laboratories, (2018). Tetrabromobisphenol A: Japanese medaka short-term reproduction assay (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:	Mechanistic-Biomarkers (exposure and effect)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064189

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 a commercial 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 for assessed outcomes
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods
	Metric 10:	Exposure Duration and Frequency	High	An approved standard duration was used
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for assessing the desired endpoints
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were above the water solubility limit. DMF was used was used to solubilize test substance.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	four replicates with adequate number of organisms per treatment
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). Tetrabromobisphenol A: Japanese medaka short-term reproduction assay (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:	Mechanistic-Biomarkers (exposure and effect)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064189

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 were no differences reported 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
	Metric 23:	Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained, only a couple of mortalities. $\leq$10% mortality in the controls.

Additional Comments: vitellogenin mRNA

Overall Quality Determination High

Study Citation:	EAG Laboratories, (2018). Tetrabromobisphenol A: Japanese medaka short-term reproduction assay (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:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064189

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 a commercial 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 for assessed outcomes
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods
	Metric 10:	Exposure Duration and Frequency	High	An approved standard duration was used
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for assessing the desired endpoints
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were above the water solubility limit. DMF was used was used to solubilize test substance.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	four replicates with adequate number of organisms per treatment
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). Tetrabromobisphenol A: Japanese medaka short-term reproduction assay (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:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064189

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 were no differences reported 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
	Metric 23:	Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained, only a couple of mortalities. $\leq$10% mortality in the controls.

Additional Comments: length, weight

Overall Quality Determination High

Study Citation:	Ye, G., Chen, Y., Wang, H. O., Ye, T., Lin, Y., Huang, Q., Chi, Y., Dong, S. (2016). Metabolomics approach reveals metabolic disorders and potential biomarkers associated with the developmental toxicity of tetrabromobisphenol A and tetrachlorobisphenol A. Scientific Reports 6:35257.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555779

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only, and no CASRN or other structural descriptors were reported.
	Metric 2:	Test Substance Source	Low	The source of the test substance was reported (Tokyo Chemical Industry, Tokyo, Japan), but the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity was reported as 97%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Hatchability as surrogate for mortality: Authors reported using a concurrent control (Figure 1Sd), but details of the use of a control group were not offered in the exposure methodology section for this outcome (last paragraph of page 8). In addition, because details were not offered, the authors did not explicitly indicate if the control was a solvent control (DMSO) or if the control was in reality a negative control.
	Metric 5:	Negative Control Response	High	Hatchability as surrogate for mortality: The biological response (percent hatching) of the solvent control group was reported and appropriate (87% hatchability, Figure 1Sd). For example, OECD Test 236 Fish Embryo Acute Toxicity (FET) Test which is a 96 hour test recommends >80% hatchability in the control group.
	Metric 6:	Randomized Allocation	Medium	Authors reported that embryos were randomly selected andcultured in 90-mm Petri dishes with 20-mL artificial seawater (page 8).
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	Hatchability as surrogate for mortality: Authors reported preparing TBBPA in DMSO to prepare the stock solution and that the final DMSO concentration in artificial seawater was 0.2%. They also reported that 20-mL artificial seawater was added to 90-mm Petri dishes for embryos or 150-mL artificial seawater to 300-mL glass containers for larvae and that culture liquid was refreshed daily. However, the authors did not offer a rationale for the use of 0.2% DMSO. For example, OECD Test 236 Fish Embryo Acute Toxicity (FET) Test recommends that the final concentration of a solvent should not exceed 100 µl/L or 0.01%.
	Metric 8:	Consistency of Exposure Administration	Low	The was no evidence to suggest that the treatment concentrations were consistently administered across groups. But it was unclear if the control group received DMSO alone.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.

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Study Citation:	Ye, G., Chen, Y., Wang, H. O., Ye, T., Lin, Y., Huang, Q., Chi, Y., Dong, S. (2016). Metabolomics approach reveals metabolic disorders and potential biomarkers associated with the developmental toxicity of tetrabromobisphenol A and tetrachlorobisphenol A. Scientific Reports 6:35257.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555779

Domain	Metric		Rating	Comments
	Metric 10:	Exposure Duration and Frequency	High	Hatchability as surrogate for mortality: The duration of exposure (from 2 to 10 dpf = 8 day exposure) was reported in the methods (p. 8) and appropriate for assessing hatchability and developmental endpoints
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Hatchability as surrogate for mortality: The number of exposure groups and spacing was reported for a dose response (0, 50, 200 and 800 μ g/L TBBPA). However, the spacing factor was 4 and exceeded recommended spacing factors of test guidelines for the embryonic or aquatic toxicity studies (e.g., OECD Test 236 or OCSPP 850.1000).
	Metric 12:	Testing at or Below Solubility Limit	High	The exposure concentrations were 0, 50, 200 and 800 μ g/L TBBPA; below the water solubility limit. In addition, authors used a solvent (0.2% DMSO), and the exposure was conducted in artificial seawater (3%), so, under these conditions solubility of TBBPA was expected to be higher than 4.15 mg/L at 25 C, pH 7.56.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The test organisms (marine medaka, Oryzias melastigma, embryos) were adequately described, but it was unclear if embryos were from an established laboratory culture or from another source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Hatchability as surrogate for mortality: Authors reported that synchronized fertilized embryos were obtained within 2 hours afterthe initiation of photophase, and fertilization was confirmed using a dissecting microscope. Embryos were exposed to TBBPA from 2 to 10 dpf (days post fertilization). There is no evidence to indicate that embryos were not treated under the same pretreatment conditions.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Hatchability as surrogate for mortality: The numbers of test organisms (35 embryos) and replicates (three) were reported and sufficient.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Hatchability as surrogate for mortality: 35 embryos were cultured in 90-mm Petri dishes with 20-mL artificial seawater and newly hatched larvae were transferred to a 300-mL glass break added with 150-mL artificial seawater. The culture liquid was refreshed daily. Marine medaka embryos hatch around 10 dpf, so exogenous feeding had not started and external feeding was unnecessary. However, it was unclear if the embryos and larvae were kept at the same temperature and salinity conditions as the adults (cultured in 28 $\pm$ 1 °C, 3% salinity).
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology (count of percent hatched embryos) addressed the intended outcome (hatchability as surrogate for mortality).
	Metric 18:	Consistency of Outcome Assessment	High	Hatchability as surrogate for mortality: There was not evidence to indicate that the outcome assessment methodology was not employed consistently across groups.
Domain 6: Confounding / Variable Control				

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Study Citation:	Ye, G., Chen, Y., Wang, H. O., Ye, T., Lin, Y., Huang, Q., Chi, Y., Dong, S. (2016). Metabolomics approach reveals metabolic disorders and potential biomarkers associated with the developmental toxicity of tetrabromobisphenol A and tetrachlorobisphenol A. Scientific Reports 6:35257.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555779

Domain	Metric		Rating	Comments
	Metric 19:	Confounding Variables in Test Design and Procedures	Medium	Hatchability as surrogate for mortality: There were no reported differences among the study groups in environmental conditions (e.g., number of organisms per experimental unit), however, authors did not clarify other factors (e.g., temperature) that could influence the outcome assessment.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Hatchability as surrogate for mortality: Statistical methods were described. A non-parametric test (two-tailed Mann-Whitney U test) was performed using MATLAB 7.11 (MathWorks, USA).
	Metric 22:	Reporting of Data	High	Hatchability as surrogate for mortality: Data were presented for each treatment and control group in Supplementary Information (Figure S 1).
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: The goal of the study was to investigate the effects of TBBPA on hatching, development (morphology), and global metabolism and nervous system assessed by a metabolomics analysis in marine medaka (Oryzias melastigma) embryos. This forms was used to evaluate hatch and hatchability which is in this case was a surrogate for mortality (i.e., exposure was to embryos and authors reported number/percent of hatched embryos inferring that those embryos that did not hatch died).

Overall Quality Determination Medium

Study Citation:	Ye, G., Chen, Y., Wang, H. O., Ye, T., Lin, Y., Huang, Q., Chi, Y., Dong, S. (2016). Metabolomics approach reveals metabolic disorders and potential biomarkers associated with the developmental toxicity of tetrabromobisphenol A and tetrachlorobisphenol A. Scientific Reports 6:35257.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Embryo
Health Outcome:	Cardiovascular
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555779

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only, and no CASRN or other structural descriptors were reported.
	Metric 2:	Test Substance Source	Low	The source of the test substance was reported (Tokyo Chemical Industry, Tokyo, Japan), but the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity was reported as 97%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Heart rate: Authors reported using a concurrent control (Figure 4b), but details of the use of a control group were not offered in the exposure methodology section for this outcome (last paragraph of page 8). In addition, because details were not offered, the authors did not explicitly indicate if the control was a solvent control (DMSO) or if the control was in reality a negative control.
	Metric 5:	Negative Control Response	High	Heart rate: The biological response of the negative control group was reported and adequate (Figure 4b).
	Metric 6:	Randomized Allocation	Low	Heart rate: Authors reported that embryos were randomly selected and cultured in 90-mm Petri dishes with 20-mL artificial seawater (page 8).
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	Heart rate: Authors reported preparing TBBPA in DMSO to prepare the stock solution and that the final DMSO concentration in artificial seawater was 0.2%. They also reported that 20-mL artificial seawater was added to 90-mm Petri dishes for embryos or 150-mL artificial seawater to 300-mL glass containers for larvae and that culture liquid was refreshed daily. However, the authors did not offer a rationale for the use of 0.2% DMSO. For example, OECD Test 236 Fish Embryo Acute Toxicity (FET) Test or OCSPP 850.1075 recommends that the final concentration of a solvent should not exceed 100 µl/L or 0.01%.
	Metric 8:	Consistency of Exposure Administration	Low	The was no evidence to suggest that the treatment concentrations were consistently administered across groups. But it was unclear if the control group received DMSO alone.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	Heart rate: The duration of exposure (from 2 to 10 dpf = 8 day exposure) was reported in the methods (p. 8) and appropriate for assessing heart rate impacts.

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Study Citation:	Ye, G., Chen, Y., Wang, H. O., Ye, T., Lin, Y., Huang, Q., Chi, Y., Dong, S. (2016). Metabolomics approach reveals metabolic disorders and potential biomarkers associated with the developmental toxicity of tetrabromobisphenol A and tetrachlorobisphenol A. Scientific Reports 6:35257.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Embryo
Health Outcome:	Cardiovascular
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555779

Domain	Metric		Rating	Comments
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Heart rate: The number of exposure groups and spacing was reported for a dose response (0, 50, 200 and 800 μ g/L TBBPA). However, the spacing factor was 4 and exceeded recommended spacing factors of test guidelines for the embryonic or aquatic toxicity studies (e.g., OECD Test 236 or OCSPP 850.1000).
	Metric 12:	Testing at or Below Solubility Limit	High	The exposure concentrations were 0, 50, 200 and 800 μ g/L TBBPA; below the water solubility limit. In addition, authors used a solvent (0.2% DMSO), and the exposure was conducted in artificial seawater (3%), so, under these conditions solubility of TBBPA was expected to be higher than 4.15 mg/L at 25 C, pH 7.56.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The test organisms (marine medaka, Oryzias melastigma, embryos) were adequately described, but it was unclear if embryos were from an established laboratory culture or from another source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Heart rate: Authors reported that synchronized fertilized embryos were obtained within 2 hours after the initiation of photophase, and fertilization was confirmed using a dissecting microscope. Embryos were exposed to TBBPA from 2 to 10 dpf (days post fertilization). There is no evidence to indicate that embryos were not treated under the same pretreatment conditions.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Heart rate: In the methods, the authors reported that the numbers of test organisms was 35 embryos and that the exposure was three replicates. However, Figure 4b indicated that number of embryos per group was 67–80.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Heart rate: 35 embryos were cultured in 90-mm Petri dishes with 20-mL artificial seawater and newly hatched larvae were transferred to a 300-mL glass break added with 150-mL artificial seawater. The culture liquid was refreshed daily. Marine medaka embryos hatch around 10 dpf, so exogenous feeding had not started and external feeding was unnecessary. However, it was unclear if the embryos and larvae were kept at the same temperature and salinity conditions as the adults (cultured in 28 $\pm$ 1 °C, 3% salinity).
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology (heart beat counts of embryos for 60 s) addressed the intended outcome (heart rate impacts). Figure 4b indicated that number of embryos assessed was 67–80.
	Metric 18:	Consistency of Outcome Assessment	High	Heart rate: There was no evidence to indicate that the outcome assessment methodology was not employed consistently across groups.
Domain 6: Confounding / Variable Control				

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Study Citation:	Ye, G., Chen, Y., Wang, H. O., Ye, T., Lin, Y., Huang, Q., Chi, Y., Dong, S. (2016). Metabolomics approach reveals metabolic disorders and potential biomarkers associated with the developmental toxicity of tetrabromobisphenol A and tetrachlorobisphenol A. Scientific Reports 6:35257.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Embryo
Health Outcome:	Cardiovascular
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555779

Domain	Metric		Rating	Comments
	Metric 19:	Confounding Variables in Test Design and Procedures	Medium	Heart rate: There were no reported differences among the study groups in environmental conditions (e.g., number of organisms per experimental unit), however, authors did not clarify other factors (e.g., temperature) that could influence the outcome assessment.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Heart rate: Statistical methods were described (nonparametric test, two-tailed Mann-Whitney U test).
	Metric 22:	Reporting of Data	Medium	Heart rate: Data were presented for 0, 50, and 200 ug/L (Figure 4b). It was unclear why the 800 ug/L group was not assessed.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: The goal of the study was to investigate the effects of TBBPA on hatching, development (morphology), and global metabolism and nervous system assessed by a metabolomics analysis in marine medaka (Oryzias melastigma) embryos. This forms was used to evaluate heart rate.

Overall Quality Determination Medium

Study Citation:	Ye, G., Chen, Y., Wang, H. O., Ye, T., Lin, Y., Huang, Q., Chi, Y., Dong, S. (2016). Metabolomics approach reveals metabolic disorders and potential biomarkers associated with the developmental toxicity of tetrabromobisphenol A and tetrachlorobisphenol A. Scientific Reports 6:35257.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555779

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only, and no CASRN or other structural descriptors were reported.
	Metric 2:	Test Substance Source	Low	The source of the test substance was reported (Tokyo Chemical Industry, Tokyo, Japan), but the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity was reported as 97%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Developmental toxicology/morphological changes: Authors reported using a concurrent control (Figure 1S b & c), but details of the use of a control group were not offered in the exposure methodology section for this outcome (last paragraph of page 8). In addition, because details were not offered, the authors did not explicitly indicate if the control was a solvent control (DMSO) or if the control was in reality a negative control.
	Metric 5:	Negative Control Response	High	Developmental toxicology/morphological changes: The biological response of the negative control group was reported and adequate (Figure 1S b & c).
	Metric 6:	Randomized Allocation	Medium	Developmental toxicology/morphological changes: Authors reported that embryos were randomly selected andcultured in 90-mm Petri dishes with 20-mL artificial seawater (page 8).
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	Developmental toxicology/morphological changes: Authors reported preparing TBBPA in DMSO to prepare the stock solution and that the final DMSO concentration in artificial seawater was 0.2%. They also reported that 20-mL artificial seawater was added to 90-mm Petri dishes for embryos or 150-mL artificial seawater to 300-mL glass containers for larvae and that culture liquid was refreshed daily. However, the authors did not offer a rationale for the use of 0.2% DMSO. For example, OECD Test 236 Fish Embryo Acute Toxicity (FET) Test or OCSPP 850.1075 recommends that the final concentration of a solvent should not exceed 100 µl/L or 0.01%.
	Metric 8:	Consistency of Exposure Administration	Low	The was no evidence to suggest that the treatment concentrations were consistently administered across groups. But it was unclear if the control group received DMSO alone.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	Developmental toxicology/morphological changes: The duration of exposure (from 2 to 10 dpf = 8 day exposure) was reported in the methods (p. 8) and appropriate for assessing development and morphological changes.

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Study Citation:	Ye, G., Chen, Y., Wang, H. O., Ye, T., Lin, Y., Huang, Q., Chi, Y., Dong, S. (2016). Metabolomics approach reveals metabolic disorders and potential biomarkers associated with the developmental toxicity of tetrabromobisphenol A and tetrachlorobisphenol A. Scientific Reports 6:35257.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555779

Domain	Metric		Rating	Comments
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Developmental toxicology/morphological changes: The number of exposure groups and spacing was reported for a dose response (0, 50, 200 and 800 μ g/L TBBPA). However, the spacing factor was 4 and exceeded recommended spacing factors of test guidelines for the embryonic or aquatic toxicity studies (e.g., OECD Test 236 or OCSPP 850.1000).
	Metric 12:	Testing at or Below Solubility Limit	High	The exposure concentrations were 0, 50, 200 and 800 μ g/L TBBPA; below the water solubility limit. In addition, authors used a solvent (0.2% DMSO), and the exposure was conducted in artificial seawater (3%), so, under these conditions solubility of TBBPA was expected to be higher than 4.15 mg/L at 25 C, pH 7.56.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The test organisms (marine medaka, Oryzias melastigma, embryos) were adequately described, but it was unclear if embryos were from an established laboratory culture or from another source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Developmental toxicology/morphological changes: Authors reported that synchronized fertilized embryos were obtained within 2 hours after the initiation of photophase, and fertilization was confirmed using a dissecting microscope. Embryos were exposed to TBBPA from 2 to 10 dpf (days post fertilization). There is no evidence to indicate that embryos were not treated under the same pretreatment conditions.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Developmental toxicology/morphological changes: The numbers of test organisms (35 embryos) and replicates (three) were reported and sufficient.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Developmental toxicology/morphological changes: 35 embryos were cultured in 90-mm Petri dishes with 20-mL artificial seawater and newly hatched larvae were transferred to a 300-mL glass break added with 150-mL artificial seawater. The culture liquid was refreshed daily. Marine medaka embryos hatch around 10 dpf, so exogenous feeding had not started and external feeding was unnecessary. However, it was unclear if the embryos and larvae were kept at the same temperature and salinity conditions as the adults (cultured in 28 $\pm$ 1 °C, 3% salinity).
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology (microscopic observation of pericardial and abdominal edemas, marked with arrows in Figure 1S c) addressed the intended outcome (morphological changes).
	Metric 18:	Consistency of Outcome Assessment	High	Developmental toxicology/morphological changes: There was no evidence to indicate that the outcome assessment methodology was not employed consistently across groups.
Domain 6: Confounding / Variable Control				

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Study Citation:	Ye, G., Chen, Y., Wang, H. O., Ye, T., Lin, Y., Huang, Q., Chi, Y., Dong, S. (2016). Metabolomics approach reveals metabolic disorders and potential biomarkers associated with the developmental toxicity of tetrabromobisphenol A and tetrachlorobisphenol A. Scientific Reports 6:35257.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555779

Domain	Metric		Rating	Comments
	Metric 19:	Confounding Variables in Test Design and Procedures	Medium	Developmental toxicology/morphological changes: There were no reported differences among the study groups in environmental conditions (e.g., number of organisms per experimental unit), however, authors did not clarify other factors (e.g., temperature) that could influence the outcome assessment.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Uninformative	Developmental toxicology/morphological changes: Statistical analysis was not conducted for developmental endpoints. Authors only provided representative photographs of an embryo per treatment group (Figure 1S b, c). Moreover, incidence of morphological changes (i.e., pericardial and abdominal edemas) across treatment groups was not reported.
	Metric 22:	Reporting of Data	Low	Developmental toxicology/morphological changes: Authors only provided representative photographs of an embryo per treatment group (Figure 1S c). Moreover, incidence of morphological changes (i.e., pericardial and abdominal edemas) across treatment groups was not reported.
	Metric 23:	Explanation of Unexpected Outcomes	Low	Developmental toxicology/morphological changes: The study did not report any measures of variability or quantify results.

Additional Comments: The goal of the study was to investigate the effects of TBBPA on hatching, development (morphology), and global metabolism and nervous system assessed by a metabolomics analysis in marine medaka (Oryzias melastigma) embryos. This forms was used to evaluate developmental toxicology/morphological changes.

Overall Quality Determination — Uninformative

Study Citation:	Industries,, Cytec (1992). Tetrabromobisphenol a: report of analytical results - cam bioassay screen in pimephales promelas (fathead minnows) with cover letter dated 122292.
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*; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547575

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Uninformative	purity reported as 1-3% TBBPA
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Control group was reported but no details given.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Uninformative	The type of experimental system or test media preparation methods were not reported.
	Metric 8:	Consistency of Exposure Administration	Low	Details of exposure administration were not reported. Reporting omissions are likely to have a substantial impact on results. It appears that the pdf is an analytical report of California Hazardous waste assessment bioassay screening conducted on aqueous samples that contain 2-3% TBBPA.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured or measurements were not reported.
	Metric 10:	Exposure Duration and Frequency	Medium	Standard test duration (96 hrs) was used but no details given.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	Based on the table given on page 5/5, there appears to be two exposure groups (250 mg/L and 750 mg/L) but the column heading says 'dilution'. This is not a standard toxicity test. This test is a screening bioassay conducted to test whether a sample should be classified as a hazardous waste or not. The test sample was diluted with laboratory water to produce specific test concentrations but these are not TBBPA specific test concentrations.
	Metric 12:	Testing at or Below Solubility Limit	Low	Reporting omissions prevented determination of whether exposure concentrations exceeded the water solubility limit (or dispersibility limit if applicable).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Test organisms were obtained from Thomas Fish company and the length and weight of test organisms were given on page 5/5.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized or not. No details on pre-treatment conditions were given in the report.

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Study Citation:	Industries,, Cytec (1992). Tetrabromobisphenol a: report of analytical results - cam bioassay screen in pimephales promelas (fathead minnows) with cover letter dated 122292.
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*; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547575

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Low	10 organisms per dilution of sample used and tested in duplicates.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reported environmental conditions were not sufficient to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Uninformative	Results are from a screening bioassay. No statistical test was conducted.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	Low	Insufficient information was provided to determine if unexpected outcomes occurred

Additional Comments: It appears that the pdf is an analytical report of California Hazardous waste assessment bioassay screening conducted on samples (that contain 2-3% TBBPA) to determine whether a sample should be classified as a hazardous waste or not. The test sample was diluted with laboratory water to produce specific test concentrations but these are not TBBPA specific test concentrations.

Overall Quality Determination Uninformative

Study Citation:	Springborn Laboratories, (1988). Acute toxicity of tetrabromobisphenol A to fathead minnow (Pimephales promelas) under flow-through conditions with attachments and cover letter dated 111788.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Pimephales promelas*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547583

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 100%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using appropriate concurrent negative control groups.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were impartially allocated into study group.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical methods (HPLC, Page 15).
	Metric 10:	Exposure Duration and Frequency	Medium	48 hours longer than a standard 96 hr test (144 hours total).
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups (5) and spacing of exposure levels were suitable for a dose response by study authors.
	Metric 12:	Testing at or Below Solubility Limit	Medium	The solvent concentration was appropriate although some precipitate was noted at higher concentrations.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The test organisms were obtained from a reliable source but age 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	Number of organisms was adequate but only two reps were used.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health.

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Study Citation:	Springborn Laboratories, (1988). Acute toxicity of tetrabromobisphenol A to fathead minnow (Pimephales promelas) under flow-through conditions with attachments and cover letter dated 111788.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Pimephales promelas*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547583

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 were no differences reported among groups that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described (Page 16/85).
	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: Table 4 on page 29/85 presents the LC50 and CI values at 24, 28, 72, 96, 120, and 144 hours of exposure.

Overall Quality Determination High

Study Citation:	Springborn Laboratories, (1989). Bioconcentration and elimination of C-residues by eastern oysters (Crassostrea virginica) exposed to tetrabromobisphenol A (final report) with attached studies and letter 081889.
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; *Pimephales promelas*; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547108

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 100%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using appropriate concurrent negative control groups.
	Metric 5:	Negative Control Response	Medium	Less than 90% hatch reported in some control replicates. Specific hatch rates are presented in Table 4 on pave 33/58.
	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 and are similar to nominal concentrations, less so at higher concentrations.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type (30 days).
	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. (5 treatment concentrations).
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit (4.1 mg/L).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health (page 20/58).

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Study Citation:	Springborn Laboratories, (1989). Bioconcentration and elimination of C-residues by eastern oysters (Crassostrea virginica) exposed to tetrabromobisphenol A (final report) with attached studies and letter 081889.
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; *Pimephales promelas*; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547108

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 were no information presented to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described in section 3.8 on page 17/58.
	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 with hatch rate were identified. Minor study deviations were reported on pave 24/58.

Additional Comments: All mortality metrics (hatch and larvae survival at 30 days) are reported in Table 4 on page 33/58.

Overall Quality Determination High

Study Citation:	Springborn Laboratories, (1989). Bioconcentration and elimination of C-residues by eastern oysters (Crassostrea virginica) exposed to tetrabromobisphenol A (final report) with attached studies and letter 081889.
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; *Pimephales promelas*; Juvenile
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547108

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Only the test substance name was reported.
	Metric 2:	Test Substance Source	Low	The radiolabeled TBBPA was received from Midwest Research Institute, Kansas City, Missouri. It was not reported whether it was analytically verified or not.
	Metric 3:	Test Substance Purity	Medium	The test substance had a radiopurity of 96.0%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	A solvent control was used.
	Metric 5:	Negative Control Response	High	The detectable concentration in the control fish was <0.5% of the mean residue concentrations measured in fish exposed to TBBPA.
	Metric 6:	Randomized Allocation	Low	It was not reported if test fish were randomly allocated to treatment groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The test system was described adequately.
	Metric 8:	Consistency of Exposure Administration	High	The exposure was administered consistently across treatment groups.
	Metric 9:	Measurement of Test Substance Concentration	High	The test concentration was measured at various times throughout the study.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration was for 24 days and that was adequate to assess the outcome of interest.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	There was only one treatment concentration besides the control.
	Metric 12:	Testing at or Below Solubility Limit	High	A solvent was used.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described in section 3.2.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The test fish were acclimated to test conditions.
	Metric 15:	Number of Organisms and Replicates per Group	Low	There was no mention of replicates. Ninety-one fish were placed in each test aquarium.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Test conditions were adequate.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology was adequately reported.

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Study Citation:	Springborn Laboratories, (1989). Bioconcentration and elimination of C-residues by eastern oysters (Crassostrea virginica) exposed to tetrabromobisphenol A (final report) with attached studies and letter 081889.
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; *Pimephales promelas*; Juvenile
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547108

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	High	The methods were carried out consistently.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	Fish were acclimated and test conditions were described and adequate.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There were no reported differences in organisms among test groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were described in section 3.8.
	Metric 22:	Reporting of Data	High	Data and results were adequately reported for the treatment and control group.
	Metric 23:	Explanation of Unexpected Outcomes	Low	Measures of variability were not shown in the figure.

Additional Comments: This evaluation is for the assessment of TBBPA bioconcentration in fathead minnows.

Overall Quality Determination High

Study Citation:	Springborn Laboratories, (1989). Bioconcentration and elimination of C-residues by eastern oysters (Crassostrea virginica) exposed to tetrabromobisphenol A (final report) with attached studies and letter 081889.
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; *Pimephales promelas*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547108

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 100%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using appropriate concurrent negative control groups.
	Metric 5:	Negative Control Response	Medium	Less than 90% hatch reported in some control replicates in table 4, page 33/58.
	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 and are similar to nominal concentrations, less so at higher concentrations Table 3, page 32/58.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were justified for a dose response by study authors.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit (4.1 mg/L).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.

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Study Citation:	Springborn Laboratories, (1989). Bioconcentration and elimination of C-residues by eastern oysters (Crassostrea virginica) exposed to tetrabromobisphenol A (final report) with attached studies and letter 081889.
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; *Pimephales promelas*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547108

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 were no information presented to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described in Section 3.8 on page 17/58.
	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 metrics (total length and mean wet weight) are presented for each treatment group in table 4, page 33/58.

Overall Quality Determination High

Study Citation:	Battelle, (2018). Fish short-term reproduction assay of 3,3',5,5'-tetrabromobisphenol A (TBBPA) in Pimephales promelas.
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; *Pimephelas promelas*; Adult
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064188

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 not 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 for assessed outcomes
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	Medium	Stock solution exceeded solubility but tanks were measured and were comparable to nominal values
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from lab culture
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health

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Study Citation:	Battelle, (2018). Fish short-term reproduction assay of 3,3',5,5'-tetrabromobisphenol A (TBBPA) in Pimephales promelas.
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; *Pimephelas promelas*; Adult
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064188

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

Additional Comments: This evaluation is for the signs of clinical toxicity that were assessed in the reference, including hyperventilation, loss of equilibrium, uncoordinated swimming, atypical acquiescence, or feeding abstinence. The behavioral outcome was selected as the outcome interest.

Overall Quality Determination High

Study Citation:	Battelle, (2018). Fish short-term reproduction assay of 3,3',5,5'-tetrabromobisphenol A (TBBPA) in Pimephales promelas.
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; *Pimephelas promelas*; Adult
Health Outcome:	Endocrine
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064188

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 not 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 for assessed outcomes
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	Medium	Stock solution exceeded solubility but tanks were measured and were comparable to nominal values
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from lab culture
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	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:	Battelle, (2018). Fish short-term reproduction assay of 3,3',5,5'-tetrabromobisphenol A (TBBPA) in Pimephales promelas.
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; *Pimephelas promelas*; Adult
Health Outcome:	Endocrine
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064188

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	N/A	Due to no mortality in control or high concentration, authors reported not statistically analyzing survival data
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: This evaluation is for the histology conducted on the thyroid gland, thus the endocrine outcome was selected as the outcome of interest.

Overall Quality Determination High

Study Citation:	Battelle, (2018). Fish short-term reproduction assay of 3,3',5,5'-tetrabromobisphenol A (TBBPA) in Pimephales promelas.
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; *Pimephelas promelas*; Adult
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064188

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 not 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 for assessed outcomes
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	Medium	Stock solution exceeded solubility but tanks were measured and were comparable to nominal values
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from lab culture
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	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:	Battelle, (2018). Fish short-term reproduction assay of 3,3',5,5'-tetrabromobisphenol A (TBBPA) in Pimephales promelas.
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; *Pimephelas promelas*; Adult
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064188

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

Additional Comments: body length, weight

Overall Quality Determination High

Study Citation:	Battelle, (2018). Fish short-term reproduction assay of 3,3',5,5'-tetrabromobisphenol A (TBBPA) in Pimephales promelas.
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; *Pimephelas promelas*; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064188

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 not 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 for assessed outcomes
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	Medium	Stock solution exceeded solubility but tanks were measured and were comparable to nominal values
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from lab culture
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	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:	Battelle, (2018). Fish short-term reproduction assay of 3,3',5,5'-tetrabromobisphenol A (TBBPA) in Pimephales promelas.
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; *Pimephelas promelas*; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064188

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	N/A	Due to no mortality in control or high concentration, authors reported not statistically analyzing survival data
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: None

Overall Quality Determination High

Study Citation:	Battelle, (2018). Fish short-term reproduction assay of 3,3',5,5'-tetrabromobisphenol A (TBBPA) in Pimephales promelas.
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; *Pimephelas promelas*; Adult
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064188

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 not 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 for assessed outcomes
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	Medium	Stock solution exceeded solubility but tanks were measured and were comparable to nominal values
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from lab culture
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	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:	Battelle, (2018). Fish short-term reproduction assay of 3,3',5,5'-tetrabromobisphenol A (TBBPA) in Pimephales promelas.
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; *Pimephelas promelas*; Adult
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064188

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

Additional Comments: VTG, estradiol, testosterone

Overall Quality Determination High

Study Citation:	Battelle, (2018). Fish short-term reproduction assay of 3,3',5,5'-tetrabromobisphenol A (TBBPA) in Pimephales promelas.
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; *Pimephelas promelas*; Adult
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064188

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 not 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 for assessed outcomes
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	Medium	Stock solution exceeded solubility but tanks were measured and were comparable to nominal values
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from lab culture
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	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:	Battelle, (2018). Fish short-term reproduction assay of 3,3',5,5'-tetrabromobisphenol A (TBBPA) in Pimephales promelas.
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; *Pimephelas promelas*; Adult
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064188

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

Additional Comments: gonad condition, GSI, nuptial tubule, fecundity, fertilization

Overall Quality Determination High

Study Citation:	Veldhoen, N., Boggs, A., Walzak, K., Helbing, C. C. (2006). Exposure to tetrabromobisphenol-A alters TH-associated gene expression and tadpole metamorphosis in the Pacific tree frog Pseudacris regilla. Aquatic Toxicology 78(3):292-302.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Pseudacris regilla*; Larvae
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1414891

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance was purchased from AccuStandard but identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent solvent control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations and/or minimize loss of test substance before and during the exposure.
	Metric 8:	Consistency of Exposure Administration	Medium	Reporting omissions are likely to have a substantial impact on results. Exposure concentration was unclear, but authors indicated that test concentrations were renewed after 48 hr.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured and test concentration was not clearly reported, ”10 nM or 100nM TBBPA or a combinationof T3 and TBBPA concentrations at an application ratio of 1:10000 (chemical: water).”
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type and is consistent with other amphibian developmental studies quantifying similar effects.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Uninformative	Only two concentrations were tested (10nM TBBPA, 100nM TBBPA) and, in addition to being very unclearly described, were insufficient to characterize a significant response in the developmental endpoints tested.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	There are minor reservations over the use of wild collected egg masses, but as no significant mortality or disease effects were reported in the study, these were unlikely to affect the outcome of the study. The lifestage (Gosner stages 30–31) was acceptable because at this lifestage, it is likely that the thyroid hasn’t developed yet.

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Study Citation:	Veldhoen, N., Boggs, A., Walzak, K., Helbing, C. C. (2006). Exposure to tetrabromobisphenol-A alters TH-associated gene expression and tadpole metamorphosis in the Pacific tree frog Pseudacris regilla. Aquatic Toxicology 78(3):292-302.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Pseudacris regilla*; Larvae
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1414891

Domain	Metric		Rating	Comments
	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 numbers of test organisms (8) and replicates (1) were lower than the typical number used in studies of the same or similar type and affected the statistical power of the results.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reporting of housing and/or environmental conditions and/or food, water, and nutrients and/or biomass loading was not sufficiently reported to evaluate if adequate and whether differences occurred between control and exposed populations.
	Metric 17:	Outcome Assessment Methodology	Uninformative	The reported outcome assessment methodology was not sensitive for the outcomes of interest. No significant affect on developmental effects were observed.
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: The test concentrations were not reported clearly. Without sufficient explanation of the methodology used to create the test concentrations, a comparison with other similar studies is not possible. In addition, the test concentrations selected were insufficient to characterize an effect on growth and development. Mixture with T3 also reported.

Overall Quality Determination — Uninformative

Study Citation:	Veldhoen, N., Boggs, A., Walzak, K., Helbing, C. C. (2006). Exposure to tetrabromobisphenol-A alters TH-associated gene expression and tadpole metamorphosis in the Pacific tree frog Pseudacris regilla. Aquatic Toxicology 78(3):292-302.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Pseudacris regilla*; Larvae
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1414891

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance was purchased from AccuStandard but identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent solvent control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations and/or minimize loss of test substance before and during the exposure.
	Metric 8:	Consistency of Exposure Administration	Medium	Reporting omissions are likely to have a substantial impact on results. Exposure concentration was unclear, but authors indicated that test concentrations were renewed after 48 hr.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured and test concentration was not clearly reported, ”10 nM or 100nM TBBPA or a combination of T3 and TBBPA concentrations at an application ratio of 1:10000 (chemical: water).”
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type and is consistent with other amphibian developmental studies quantifying similar effects.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	Only two concentrations were tested (10nM TBBPA, 100nM TBBPA) which were very unclearly described, but were sufficient to characterize a significant response in the mechanistic endpoints tested.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	There are minor reservations over the use of wild collected egg masses, but as no significant mortality or disease effects were reported in the study, these were unlikely to affect the outcome of the study. The lifestage (Gosner stages 30–31) was acceptable because at this lifestage, it is likely that the thyroid hasn’t developed yet.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups

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Study Citation:	Veldhoen, N., Boggs, A., Walzak, K., Helbing, C. C. (2006). Exposure to tetrabromobisphenol-A alters TH-associated gene expression and tadpole metamorphosis in the Pacific tree frog Pseudacris regilla. Aquatic Toxicology 78(3):292-302.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Pseudacris regilla*; Larvae
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1414891

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Low	The numbers of test organisms (8) and replicates (1) were lower than the typical number used in studies of the same or similar type and affected the statistical power of the results.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reporting of housing and/or environmental conditions and/or food, water, and nutrients and/or biomass loading was not sufficiently reported to evaluate if adequate and whether differences occurred between control and exposed populations.
	Metric 17:	Outcome Assessment Methodology	Medium	The outcome assessment methodology addressed or reported the intended outcome but there are minor uncertainties that were identified.
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: mixture with T3 also reported

Overall Quality Determination Medium

Study Citation:	Yang, S. W., Yan, Z. G., Xu, F. F., Wang, S. R., Wu, F. C. (2012). Development of freshwater aquatic life criteria for tetrabromobisphenol A in China. Environmental Pollution 169:59-63.
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; *Pseudorasbora parva*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2562857

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%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using appropriate concurrent negative control groups
	Metric 5:	Negative Control Response	High	Mortality was less than 8 percent
	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	The study provided few details on exposure administration
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and 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 above the solubility limit listed in the Final TBBPA Scope (4.15 mg/L) were included, however an ethanol solvent was used and a solvent control was indicated.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	There are minor reservations about the source of test organisms
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	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	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed

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Study Citation:	Yang, S. W., Yan, Z. G., Xu, F. F., Wang, S. R., Wu, F. C. (2012). Development of freshwater aquatic life criteria for tetrabromobisphenol A in China. Environmental Pollution 169:59-63.
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; *Pseudorasbora parva*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2562857

Domain	Metric		Rating	Comments
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the outcome assessment were not reported.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	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
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: None

Overall Quality Determination Medium

Study Citation:	Carlsson, G., Norrgren, L. (2014). Comparison of embryo toxicity using two classes of aquatic vertebrates. Environmental Toxicology and Pharmacology 37(1):24-7. [Environmental toxicology and pharmacology].
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Rana arvalis*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5518140

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity 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	Low	The biological response of the negative control groups was not reported.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Medium	Reporting omissions were unlikely to have a substantial impact on results
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure 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 were obtained from a unreliable 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 replicates was not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Reported environmental conditions were not sufficient to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported

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Study Citation:	Carlsson, G., Norrgren, L. (2014). Comparison of embryo toxicity using two classes of aquatic vertebrates. Environmental Toxicology and Pharmacology 37(1):24-7. [Environmental toxicology and pharmacology].
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Rana arvalis*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5518140

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22:	Reporting of Data	Low	Data were only reported for some outcomes
	Metric 23:	Explanation of Unexpected Outcomes	Low	insufficient information was provided to determine if excessive variability or unexpected outcomes occurred.

Additional Comments: None

Overall Quality Determination Medium

Study Citation:	Carlsson, G., Norrgren, L. (2014). Comparison of embryo toxicity using two classes of aquatic vertebrates. Environmental Toxicology and Pharmacology 37(1):24-7. [Environmental toxicology and pharmacology].
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Rana arvalis*; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5518140

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity 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	Low	The biological response of the negative control groups was not reported.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Medium	Reporting omissions were unlikely to have a substantial impact on results
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure 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 were obtained from a unreliable 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 replicates was not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Reported environmental conditions were not sufficient to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported.

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Study Citation:	Carlsson, G., Norrgren, L. (2014). Comparison of embryo toxicity using two classes of aquatic vertebrates. Environmental Toxicology and Pharmacology 37(1):24-7. [Environmental toxicology and pharmacology].
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Rana arvalis*; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5518140

Domain		Metric	Rating	Comments
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22:	Reporting of Data	Low	Data were only reported for some outcomes
	Metric 23:	Explanation of Unexpected Outcomes	Low	insufficient information was provided to determine if excessive variability or unexpected outcomes occurred.

Additional Comments: None

Overall Quality Determination Medium

Study Citation:	Yang, S. W., Yan, Z. G., Xu, F. F., Wang, S. R., Wu, F. C. (2012). Development of freshwater aquatic life criteria for tetrabromobisphenol A in China. Environmental Pollution 169:59-63.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Rana limnocharis*; Larvae
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2562857

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%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using appropriate concurrent negative control groups
	Metric 5:	Negative Control Response	High	Mortality was less than 8 percent
	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	The study provided few details on exposure administration
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and 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 above the solubility limit listed in the Final TBBPA Scope (4.15 mg/L) were included, however an ethanol solvent was used and a solvent control was indicated.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	There are minor reservations about the source of test organisms
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	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	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed

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Study Citation:	Yang, S. W., Yan, Z. G., Xu, F. F., Wang, S. R., Wu, F. C. (2012). Development of freshwater aquatic life criteria for tetrabromobisphenol A in China. Environmental Pollution 169:59-63.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Rana limnocharis*; Larvae
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2562857

Domain	Metric		Rating	Comments
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the outcome assessment were not reported.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	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
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: None

Overall Quality Determination Medium

Study Citation:	Yang, S. W., Yan, Z. G., Xu, F. F., Wang, S. R., Wu, F. C. (2012). Development of freshwater aquatic life criteria for tetrabromobisphenol A in China. Environmental Pollution 169:59-63.
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; Amphibian; *Rana limnocharis*; Larvae
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2562857

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%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using appropriate concurrent negative control groups
	Metric 5:	Negative Control Response	High	Mortality was less than 8 percent
	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	The study provided few details on exposure administration
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and 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 above the solubility limit listed in the Final TBBPA Scope (4.15 mg/L) were included, however an ethanol solvent was used and a solvent control was indicated.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	There are minor reservations about the source of test organisms
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	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	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed

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Study Citation:	Yang, S. W., Yan, Z. G., Xu, F. F., Wang, S. R., Wu, F. C. (2012). Development of freshwater aquatic life criteria for tetrabromobisphenol A in China. Environmental Pollution 169:59-63.
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; Amphibian; *Rana limnocharis*; Larvae
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2562857

Domain	Metric		Rating	Comments
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the outcome assessment were not reported.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	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
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: None

Overall Quality Determination Medium

Study Citation:	Goto, Y., Kitamura, S., Kashiwagi, K., Oofusa, K., Tooi, O., Yoshizato, K., Sato, J., Ohta, S., Kashiwagi, A. (2006). Suppression of amphibian metamorphosis by bisphenol A and related chemical substances. Journal of Health Science 52(2):160-168.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Rana rugosa*; Larvae
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4003309

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity 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 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	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	Medium	Reporting omissions are unlikely to have a substantial impact on results
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	The number of exposure groups and spacing of exposure levels were adequate
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	Initial stage or range of stages was not reported
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of test organisms was not reported
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reported environmental conditions were not sufficient to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest

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Study Citation:	Goto, Y., Kitamura, S., Kashiwagi, K., Oofusa, K., Tooi, O., Yoshizato, K., Sato, J., Ohta, S., Kashiwagi, A. (2006). Suppression of amphibian metamorphosis by bisphenol A and related chemical substances. Journal of Health Science 52(2):160-168.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Rana rugosa*; Larvae
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4003309

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	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: T3 treated organism results also reported

Overall Quality Determination Medium

Study Citation:	Goto, Y., Kitamura, S., Kashiwagi, K., Oofusa, K., Tooi, O., Yoshizato, K., Sato, J., Ohta, S., Kashiwagi, A. (2006). Suppression of amphibian metamorphosis by bisphenol A and related chemical substances. Journal of Health Science 52(2):160-168.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Rana rugosa*; Larvae
Health Outcome:	Mechanistic-Genotox (including DNA repair)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4003309

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity 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 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	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	Medium	Reporting omissions are unlikely to have a substantial impact on results
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	The number of exposure groups and spacing of exposure levels were adequate
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	Initial stage or range of stages was not reported
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of test organisms was not reported
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reported environmental conditions were not sufficient to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups

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Study Citation:	Goto, Y., Kitamura, S., Kashiwagi, K., Oofusa, K., Tooi, O., Yoshizato, K., Sato, J., Ohta, S., Kashiwagi, A. (2006). Suppression of amphibian metamorphosis by bisphenol A and related chemical substances. Journal of Health Science 52(2):160-168.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Rana rugosa*; Larvae
Health Outcome:	Mechanistic-Genotox (including DNA repair)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4003309

Domain	Metric		Rating	Comments
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	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: DNA fragmentation, T3 treated organism results also reported

Overall Quality Determination Medium

Study Citation:	Union Carbide Corporation Environmental Services, (1978). Acute toxicity of FMBP4A (tetrabromobisphenol A) to the rainbow trout, Salmo gairdneri Richardson.
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 gairdneri*; Synonym: Oncorhynchus mykiss; Juvenile
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5549807

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Test substance was identified by name (tetrabromobisphenol A) and CASRN (79-94-7).
	Metric 2:	Test Substance Source	Low	Source was not reported and chemical was not analytically verified.
	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	Water control and solvent control groups were included in the study.
	Metric 5:	Negative Control Response	High	Biological response in the control groups was appropriate: No mortality was observed (Table 1).
	Metric 6:	Randomized Allocation	Medium	Authors reported that fish were randomly selected from the stock culture.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	Some details of test media such as renewal of solutions throughout the exposure duration was not reported. The authors reported that the test was prepared by weight to a precision of 0.1 mg and diluted to volume in volumetric glassware with acetone. The test was conducted in 5 gallon, chemically clean, glass jars containing 15 liters of water. The amount of solvent in the solvent control equaled that amount used in the highest concentration.
	Metric 8:	Consistency of Exposure Administration	High	There was no evidence to suggest that the exposure was not administered consistently across groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured, water renewal were not reported, so it is unclear if the nominal concentrations would be similar to the measured concentrations.
	Metric 10:	Exposure Duration and Frequency	High	The study was an acute toxicity study to determine the 96 h LC50 value; so a 96 h study was appropriate.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing was appropriate (control, solvent control, 0.1, 0.18. 0.32, 0.56, and 1.0 mg/L).
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit of TBBPA (4.15 mg/L at 25 C pH 7.56). While the exposure was performed at 13 C and solubility decreases with temperature, the authors used a solvent control (acetone).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described (rainbow trout, Salmo gairdneri, 3 months old, and length of 41 mm and weight of 0.51 grams) and were obtained from a reliable source (commercial hatchery in Washington).

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Study Citation:	Union Carbide Corporation Environmental Services, (1978). Acute toxicity of FMBP4A (tetrabromobisphenol A) to the rainbow trout, Salmo gairdneri Richardson.
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 gairdneri*; Synonym: Oncorhynchus mykiss; Juvenile
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5549807

Domain	Metric		Rating	Comments
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Fish were acclimated to the test water for 2.4 hours prior to testing.
	Metric 15:	Number of Organisms and Replicates per Group	Low	Ten individuals were placed in each of the 5 gallon test vessels but replicates were not used.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Culture conditions (temperature, Do, pH, biological loading, feeding regimen) were reported and appropriate.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology (mortality counts) was reported and appropriate for the intended outcome (deriving the 96 h LC50 value).
	Metric 18:	Consistency of Outcome Assessment	High	The outcome was assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in culture conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Spearman-Karber Estimator was used to calculate the LC50 value.
	Metric 22:	Reporting of Data	High	Mortality data was reported for all treatment groups (Table 1).
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: The study was an acute toxicity test in rainbow trout to derive a 96 h LC50 value. The authors reported using standardized guidelines (US Environmental Protection Agency, 1975). In addition to the mortality data, the authors reported anecdotal behavioral observations (twitching, erratic swimming, dark discoloration and labored respiration) in rainbow trout exposed to concentrations of 0.18 mg/l and higher; however, discrete data were not collected (such as incidence and differences across treatment groups). This form was used to evaluate the mortality assessment.

Overall Quality Determination High

Study Citation:	Union Carbide Corporation Environmental Services, (1978). Acute toxicity of FMBP4A (tetrabromobisphenol A) to the rainbow trout, Salmo gairdneri Richardson.
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 gairdneri*; Synonym: Oncorhynchus mykiss; Juvenile
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5549807

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Test substance was identified by name (tetrabromobisphenol A) and CASRN (79-94-7).
	Metric 2:	Test Substance Source	Low	Source was not reported and chemical was not analytically verified.
	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	Water control and solvent control groups were included in the study.
	Metric 5:	Negative Control Response	High	Biological response in the control groups was appropriate: No mortality was observed (Table 1).
	Metric 6:	Randomized Allocation	Medium	Authors reported that fish were randomly selected from the stock culture.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	Some details of test media such as renewal of solutions throughout the exposure duration was not reported. The authors reported that the test was prepared by weight to a precision of 0.1 mg and diluted to volume in volumetric glassware with acetone. The test was conducted in 5 gallon, chemically clean, glass jars containing 15 liters of water. The amount of solvent in the solvent control equalled thatamount used in the highest concentration.
	Metric 8:	Consistency of Exposure Administration	High	There was no evidence to suggest that the exposure was not administered consistently across groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured, water renewal were not reported, so it is unclear if the nominal concentrations would be similar to the measured concentrations.
	Metric 10:	Exposure Duration and Frequency	High	The study was an acute toxicity study to determine the 96 h LC50 value; so a 96 h study was appropriate.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing was appropriate (control, solvent control, 0.1, 0.18. 0.32, 0.56, and 1.0 mg/L).
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit of TBBPA (4.15 mg/L at 25 C pH 7.56). While the exposure was performed at 13 C and solubility decreases with temperature, the authors used a solvent control (acetone).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described (rainbow trout, Salmo gairdneri, 3 months old, and length of 41 mm and weight of 0.51 grams) and were obtained from a reliable source (commercial hatchery in Washington).
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Fish were acclimated to the test water for 2.4 hours prior to testing.

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Study Citation:	Union Carbide Corporation Environmental Services, (1978). Acute toxicity of FMBP4A (tetrabromobisphenol A) to the rainbow trout, Salmo gairdneri Richardson.
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 gairdneri*; Synonym: Oncorhynchus mykiss; Juvenile
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5549807

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Low	Ten individuals were placed in each of the 5 gallon test vessels but replicates were not used.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Culture conditions (temperature, Do, pH, biological loading, feeding regimen) were reported and appropriate.
	Metric 17:	Outcome Assessment Methodology	Uninformative	The authors reported anecdotal behavioral observations (twitching, erratic swimming, dark discoloration and labored respiration) in rainbow trout exposed to concentrations of 0.18 mg/l and higher; however, discrete data were not collected (such as incidence for each of the endpoints and statistical differences across treatment groups).
	Metric 18:	Consistency of Outcome Assessment	Low	It is unclear what protocol for the behavioral observations was consistent because a protocol for the endpoints was not reported.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in culture conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Uninformative	The authors reported anecdotal behavioral observations (twitching, erratic swimming, dark discoloration and labored respiration) in rainbow trout exposed to concentrations of 0.18 mg/l and higher; however, discrete data were not collected (e.g., statistical differences across treatment groups).
	Metric 22:	Reporting of Data	Low	The authors reported anecdotal behavioral observations (twitching, erratic swimming, dark discoloration and labored respiration) in rainbow trout exposed to concentrations of 0.18 mg/l and higher.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: The study was an acute toxicity test in rainbow trout to derive a 96 h LC50 value. The authors reported using standardized guidelines (US Environmental Protection Agency, 1975). In addition to the mortality data, the authors reported anecdotal behavioral observations (twitching, erratic swimming, dark discoloration and labored respiration) in rainbow trout exposed to concentrations of 0.18 mg/l and higher; however, discrete data were not collected (such as incidence and differences across treatment groups). This form was used to evaluate the behavioral observations as reported by authors.

Overall Quality Determination Uninformative

Study Citation:	Jurgella, G. F., Marwah, A., Malison, J. A., Peterson, R., Barry, T. P. (2006). Effects of xenobiotics and steroids on renal and hepatic estrogen metabolism in lake trout. General and Comparative Endrocrinology 148(2):273-281.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Salvelinus namaycush*; in vitro study with liver and kidney tissue; Juvenile
Health Outcome:	Mechanistic-Endocrine toxicity-Kidney/renal-Liver toxicology
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2159104

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The chemical was identified by name only.
	Metric 2:	Test Substance Source	Low	The test substance identity was NOT analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Chemical purity and grade were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	In the methods section it is slightly unclear what the control consisted of. It was mentioned that "Controls were no-tissue and time 0 incubates." In the text under Table 2, study authors mention that liver and kidney tissues were incubated for 1 hour in vitro in the absence (which is the control) or in the presence of the test chemicals.
	Metric 5:	Negative Control Response	High	There would not be a biological response with a blank used as a control.
	Metric 6:	Randomized Allocation	Low	There was no mention on how the tissues were allocated for testing.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were either not measured or not reported.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	There was only one exposure level for each chemical, but it was sufficient to get the data the researchers were looking for.
	Metric 12:	Testing at or Below Solubility Limit	High	An appropriate solvent was used to aid in dissolution.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Pre treatment conditions were the same for the control and treatment organisms. In vitro cultures were kept at similar temperatures as the pre treatment organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The number of replicates in the in vitro test appeared sufficient to characterize toxicological effects. Experiments used tissues fromone fish cultured in duplicate incubations, and each experiment was conducted two to four times.
Domain 5: Outcome Assessment				

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Study Citation:	Jurgella, G. F., Marwah, A., Malison, J. A., Peterson, R., Barry, T. P. (2006). Effects of xenobiotics and steroids on renal and hepatic estrogen metabolism in lake trout. General and Comparative Endrocrinology 148(2):273-281.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Salvelinus namaycush*; in vitro study with liver and kidney tissue; Juvenile
Health Outcome:	Mechanistic-Endocrine toxicity-Kidney/renal-Liver toxicology
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2159104

Domain	Metric		Rating	Comments
	Metric 16:	Adequacy of Test Conditions	High	Pre-treatment environmental conditions were conducive to the organism's health, and tissues were maintained in appropriate culture media during treatment.
	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 outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to the exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described in section 2.6.
	Metric 22:	Reporting of Data	Medium	Summarized results were shown in Table 2. No raw data was shown. Results were also described in the text.
	Metric 23:	Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained. Measures of variability were shown in Table 2.

Additional Comments: None

Overall Quality Determination Medium

Study Citation:	Goto, Y., Kitamura, S., Kashiwagi, K., Oofusa, K., Tooi, O., Yoshizato, K., Sato, J., Ohta, S., Kashiwagi, A. (2006). Suppression of amphibian metamorphosis by bisphenol A and related chemical substances. Journal of Health Science 52(2):160-168.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Silurana tropicalis*; Larvae
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4003309

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity 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 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	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	Medium	Reporting omissions are unlikely to have a substantial impact on results
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	The number of exposure groups and spacing of exposure levels were adequate
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	Initial stage or range of stages was not reported
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of test organisms was not reported
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reported environmental conditions were not sufficient to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest

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Study Citation:	Goto, Y., Kitamura, S., Kashiwagi, K., Oofusa, K., Tooi, O., Yoshizato, K., Sato, J., Ohta, S., Kashiwagi, A. (2006). Suppression of amphibian metamorphosis by bisphenol A and related chemical substances. Journal of Health Science 52(2):160-168.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Silurana tropicalis*; Larvae
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4003309

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	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: T3 treated organism results also reported

Overall Quality Determination Medium

Study Citation:	Carlsson, G., Pohl, J., Athanassiadis, I., Norrgren, L., Weiss, J. (2019). Thyroid disruption properties of three indoor dust chemicals tested in Silurana tropicalis tadpoles. Journal of Applied Toxicology 07(9):1248-1256.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Silurana tropicalis*; Larvae
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5518130

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	Source was Sigma-Aldrich.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 97%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	Medium	No mention of irregularities in exposure administration
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical methods.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	For some endpoints (BL, SBL), significant differences from control were observed at the lowest exposure level (0.125 mg/L)
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms whether pretreatment conditions were the same for control and exposed groups
	Metric 15:	Number of Organisms and Replicates per Group	Medium	12 organisms were exposed individually in separate 1L containers per treatment.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reported environmental conditions were not sufficient to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest

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Study Citation:	Carlsson, G., Pohl, J., Athanassiadis, I., Norrgren, L., Weiss, J. (2019). Thyroid disruption properties of three indoor dust chemicals tested in Silurana tropicalis tadpoles. Journal of Applied Toxicology 07(9):1248-1256.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Silurana tropicalis*; Larvae
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5518130

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described
	Metric 22:	Reporting of Data	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 High

Study Citation:	Fini, J. B., Riu, A., Debrauwer, L., Hillenweck, A., Mével, Le, S., Chevolleau, S., Boulahtouf, A., Palmier, K., Balaguer, P., Cravedi, J. P., Demeneix, B. A., Zalko, D. (2012). Parallel biotransformation of tetrabromobisphenol A in Xenopus laevis and mammals: Xenopus as a model for endocrine perturbation studies. Toxicological Sciences 125(2):359-367.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus laevis*; TH/bZIP-GFP; Larvae
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1293799

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was only identified by name (tetrabromobisphenol A, TBBPA), but no other descriptors such as CASRN or structure were reported.
	Metric 2:	Test Substance Source	High	Tadpole morphology assay and TH reporter assay: TBBPA, TBBPA-sulfate, and TBBPA-disulfate. TBBPA (>97% purity) was purchased from Sigma-Aldrich. TBBPA-sulfate, and TBBPA-disulfate were synthesized by the laboratory.Kinetic assay: 14C-TBBPA was synthesized in laboratory and confirmed via multiple analytical techniques (HPLC, NMR, etc).
	Metric 3:	Test Substance Purity	High	Tadpole morphology assay and TH reporter assay: TBBPA (>97% purity) was purchased from Sigma-Aldrich. TBBPA-sulfate, and TBBPA-disulfate were synthesized by the laboratory.Kinetic assay: 14C-TBBPA was synthesized in laboratory and confirmed via multiple analytical techniques (HPLC, NMR, etc). Purity confirmed >99.8% via HPLC.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	TH reporter assay: Details are missing in the methods, but authors indicated a control group in Figure 3. Moreover, it is unclear if the control group was a solvent control (ethanol) since as per the authors TBBPA, in ethanol, was added to dechlorinated water. Also, there was no indication of the % ethanol used.
	Metric 5:	Negative Control Response	High	TH reporter assay: The biological response (repression of T3-induced GFP expression to measure the antithyroidal effect of TBBPA and metabolites) of the control group was reported and appeared appropriate (Figure 3D).
	Metric 6:	Randomized Allocation	Low	Randomized allocation not stated.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study lacked details on how the exposure solutions were prepared for the various assessments. Some details appear in the methods, others in the figure captions or results, but overall details were missing.
	Metric 8:	Consistency of Exposure Administration	High	Evidence suggests that exposure administration was administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	TBBPA was measured in water and tadpoles as part of the kinetic assessment of the study.
	Metric 10:	Exposure Duration and Frequency	High	TH reporter assay: The exposure duration (3 days) was reported and appropriate for the endpoint (antithyroid effect of TBBPA and its metabolites).

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Study Citation:	Fini, J. B., Riu, A., Debrauwer, L., Hillenweck, A., Mével, Le, S., Chevolleau, S., Boulahtouf, A., Palmier, K., Balaguer, P., Cravedi, J. P., Demeneix, B. A., Zalko, D. (2012). Parallel biotransformation of tetrabromobisphenol A in Xenopus laevis and mammals: Xenopus as a model for endocrine perturbation studies. Toxicological Sciences 125(2):359-367.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus laevis*; TH/bZIP-GFP; Larvae
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1293799

Domain	Metric		Rating	Comments
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	The goal of the study was not to have a dose-dependent effect; study had a single TBBBA dose and a single metabolite dose.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentration was below the solubility limit; TBBPA exposure concentration was 10^-6 M = 0.5439 mg/L and TBBPA's solubility limit is 4.15 mg/L at 25 C pH 7.56.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	Test organisms were adequately described (X. laevis NF45 tadpoles). However, the authors did not specify if organisms were from a X. laevis culture they maintained or obtained from an independent source for the experiments.
	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	TH reporter assay: Information was reported in the methods and Figure 3 caption. Fifteen NF45 tadpoles were added per well (experimental unit). No replicate wells were included, but authors indicated that experiments were performed three times in the caption of Figure 3D.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	The care and treatment of animals were in accordance with institutional and national guidelines (Charte Nationale Portant sur l'Ethique de l'Expe´rimentation Animale, 2009).
	Metric 17:	Outcome Assessment Methodology	High	TH reporter assay: The outcome assessment methodology (use of THbZIP eGFP transgenic tadpoles to measure TH expression) was reported and appropriate for the intended outcome of interest (antithyroidal effect of TBBPA and metabolites).
	Metric 18:	Consistency of Outcome Assessment	High	The outcome assessment was consistent across 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.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were described (one-way ANOVA followed by Dunn's test).
	Metric 22:	Reporting of Data	High	TH reporter assay: Data were presented for each control and treatment groups (Figure 3D).

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Study Citation:	Fini, J. B., Riu, A., Debrauwer, L., Hillenweck, A., Mével, Le, S., Chevolleau, S., Boulahtouf, A., Palmier, K., Balaguer, P., Cravedi, J. P., Demeneix, B. A., Zalko, D. (2012). Parallel biotransformation of tetrabromobisphenol A in Xenopus laevis and mammals: Xenopus as a model for endocrine perturbation studies. Toxicological Sciences 125(2):359-367.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus laevis*; TH/bZIP-GFP; Larvae
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1293799

Domain	Metric		Rating	Comments
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This study examined thyroid disruption effects of TBBPA in Xenopus laevis. To do this the authors (1) assessed the kinetics of TBBPA (uptake and excretion) in X. laevis tadpoles, (2) investigated if antithyroid effects are due to the parent TBBPA or a metabolite using two readouts: tadpole morphology and an in vivo TH reporter assay, and (3) examined the ability of sulfate metabolites andTBBPA to exert direct actions on the TRalpha using a reporter in vitro cell assay. This form was used to evaluate the tadpole TH reporter assay with THbZIP eGFP transgenic tadpoles following a 3-day TBBPA exposure.

Overall Quality Determination High

Study Citation:	Fini, J. B., Riu, A., Debrauwer, L., Hillenweck, A., Mével, Le, S., Chevolleau, S., Boulahtouf, A., Palmier, K., Balaguer, P., Cravedi, J. P., Demeneix, B. A., Zalko, D. (2012). Parallel biotransformation of tetrabromobisphenol A in Xenopus laevis and mammals: Xenopus as a model for endocrine perturbation studies. Toxicological Sciences 125(2):359-367.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus laevis*; Larvae
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1293799

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was only identified by name (tetrabromobisphenol A, TBBPA), but no other descriptors such as CASRN or structure were reported.
	Metric 2:	Test Substance Source	High	Tadpole morphology assay and TH reporter assay: TBBPA, TBBPA-sulfate, and TBBPA-disulfate. TBBPA (>97% purity) was purchased from Sigma-Aldrich. TBBPA-sulfate, and TBBPA-disulfate were synthesized by the laboratory.Kinetic assay: 14C-TBBPA was synthesized in laboratory and confirmed via multiple analytical techniques (HPLC, NMR, etc).
	Metric 3:	Test Substance Purity	High	Tadpole morphology assay and TH reporter assay: TBBPA (>97% purity) was purchased from Sigma-Aldrich. TBBPA-sulfate, and TBBPA-disulfate were synthesized by the laboratory.Kinetic assay: 14C-TBBPA was synthesized in laboratory and confirmed via multiple analytical techniques (HPLC, NMR, etc). Purity confirmed >99.8% via HPLC.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Uninformative	Radiolabeled TBBPA experiment. No water only control.
	Metric 5:	Negative Control Response	N/A	No water only control.
	Metric 6:	Randomized Allocation	Low	Randomized allocation not stated.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study lacked details on how the exposure solutions were prepared for the various assessments. Some details appear in the methods, others in the figure captions or results, but overall details were missing.
	Metric 8:	Consistency of Exposure Administration	High	Evidence suggested that exposure administration was administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	TBBPA was measured in water and tadpoles as part of the kinetic assessment of the study.
	Metric 10:	Exposure Duration and Frequency	High	Kinetic assay (uptake and excretion): The duration of exposure (72 hours) was reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Kinetic assay (uptake and excretion): The authors reported the number of exposures concentrations (10^-6 and 10^-7 M), but there was not rationale on why these concentrations were selected.
	Metric 12:	Testing at or Below Solubility Limit	High	Kinetic assay (uptake and excretion): Exposure concentrations were below the solubility limit; TBBPA exposure concentration were 10^-6 M = 0.5439 mg/L and 10^-7 M = 0.05439 mg/L. TBBPA's solubility limit is 4.15 mg/L at 25 C pH 7.56.

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Study Citation:	Fini, J. B., Riu, A., Debrauwer, L., Hillenweck, A., Mével, Le, S., Chevolleau, S., Boulahtouf, A., Palmier, K., Balaguer, P., Cravedi, J. P., Demeneix, B. A., Zalko, D. (2012). Parallel biotransformation of tetrabromobisphenol A in Xenopus laevis and mammals: Xenopus as a model for endocrine perturbation studies. Toxicological Sciences 125(2):359-367.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus laevis*; Larvae
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1293799

Domain	Metric		Rating	Comments
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	Test organisms were adequately described (X. laevis NF45 tadpoles). However, the authors did not specify if organisms were from a X. laevis culture they maintained or obtained from an independent source for the experiments.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.
	Metric 15:	Number of Organisms and Replicates per Group	Low	Kinetic assay (uptake and excretion): The number of organisms per group was reported (15 organisms), but the authors did not report using replicate groups. They indicated that "Each group of NF45 tadpoles (n =15) was placed in a silanized glass tube" and they also reported "At 72 h, all tadpoles (15) from each group were pooled and deep frozen on dry ice until extracted". On the other hand, data in the results were expressed as mean ± SD; unclear if this was pseudoreplication.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	The care and treatment of animals were in accordance with institutional and national guidelines (Charte Nationale Portant sur l'Ethique de l'Expe´rimentation Animale, 2009).
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology (measurement of radio-labeled and unlabeled TBBPA and its metabolites in water and tissues by integrating the area of the radio-chromatographic peaks) was reported and appropriate for the intended outcome of interest (uptake and excretion).
	Metric 18:	Consistency of Outcome Assessment	High	The outcome assessment was consistent across 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.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	N/A	Statistical analysis was not necessary to assess how much TBBPA was present in the water or taken up by tadpoles during the 72 hour exposure. Statistical analysis was not conducted for the kinetic assay (results were expressed as mean ± SD).
	Metric 22:	Reporting of Data	High	Results were reported for all groups and expressed as mean ± SD (Figures 1 and 2).
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

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Study Citation:	Fini, J. B., Riu, A., Debrauwer, L., Hillenweck, A., Mével, Le, S., Chevolleau, S., Boulahtouf, A., Palmier, K., Balaguer, P., Cravedi, J. P., Demeneix, B. A., Zalko, D. (2012). Parallel biotransformation of tetrabromobisphenol A in Xenopus laevis and mammals: Xenopus as a model for endocrine perturbation studies. Toxicological Sciences 125(2):359-367.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus laevis*; Larvae
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1293799

Domain	Metric	Rating	Comments
Additional Comments:	This study examined thyroid disruption effects of TBBPA in Xenopus laevis. To do this the authors (1) assessed the kinetics of TBBPA (uptake and excretion) in X. laevis tadpoles, (2) investigated if antithyroid effects are due to the parent TBBPA or a metabolite using two readouts: tadpole morphology and an in vivo TH reporter assay, and (3) examined the ability of sulfate metabolites andTBBPA to exert direct actions on the TRalpha using a reporter in vitro cell assay. This form was used to evaluate the kinetics of TBBPA (uptake and excretion) in X. laevis tadpoles.		

Overall Quality Determination Uninformative

Study Citation:	Jagnytsch, O., Opitz, R., Lutz, I., Kloas, W. (2006). Effects of tetrabromobisphenol A on larval development and thyroid hormone-regulated biomarkers of the amphibian Xenopus laevis. Environmental Research 101(3):340-348.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus laevis*; Juvenile
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1298685

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 >98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent solvent control group and 2 positive controls
	Metric 5:	Negative Control Response	High	The biological response of the solvent 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	Reporting omissions were unlikely to have a substantial impact on results
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable
	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 (bred in the Department of Inland Fisheries, Leibniz-Institute of Freshwater Ecology and Inland Fisheries, Berlin, Germany).
	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 numbers of test organisms and replicates were lower than the typical number used in studies of the same or similar type but sufficient for statistical analysis
Domain 5: Outcome Assessment				

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Study Citation:	Jagnytsch, O., Opitz, R., Lutz, I., Kloas, W. (2006). Effects of tetrabromobisphenol A on larval development and thyroid hormone-regulated biomarkers of the amphibian Xenopus laevis. Environmental Research 101(3):340-348.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus laevis*; Juvenile
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1298685

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

Additional Comments: None

Overall Quality Determination High

Study Citation:	Jagnytsch, O., Opitz, R., Lutz, I., Kloas, W. (2006). Effects of tetrabromobisphenol A on larval development and thyroid hormone-regulated biomarkers of the amphibian Xenopus laevis. Environmental Research 101(3):340-348.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus laevis*; Juvenile
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1298685

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 was obtained from Sigman, but the identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent solvent control group and 2 positive controls
	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	Reporting omissions were unlikely to have a substantial impact on results
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit and a solvent was used to facilitate incorporation into the test media.
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 numbers of test organisms and replicates were lower than the typical number used in studies of the same or similar type but sufficient for statistical analysis
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:	Jagnytsch, O., Opitz, R., Lutz, I., Kloas, W. (2006). Effects of tetrabromobisphenol A on larval development and thyroid hormone-regulated biomarkers of the amphibian Xenopus laevis. Environmental Research 101(3):340-348.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus laevis*; Juvenile
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1298685

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

Additional Comments: None

Overall Quality Determination High

Study Citation:	Fini, J. B., Riu, A., Debrauwer, L., Hillenweck, A., Mével, Le, S., Chevolleau, S., Boulahtouf, A., Palmier, K., Balaguer, P., Cravedi, J. P., Demeneix, B. A., Zalko, D. (2012). Parallel biotransformation of tetrabromobisphenol A in Xenopus laevis and mammals: Xenopus as a model for endocrine perturbation studies. Toxicological Sciences 125(2):359-367.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus laevis*; Larvae
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1293799

Domain		Metric	Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was only identified by name (tetrabromobisphenol A, TBBPA), but no other descriptors such as CASRN or structure were reported.
	Metric 2:	Test Substance Source	High	Tadpole morphology assay and TH reporter assay: TBBPA, TBBPA-sulfate, and TBBPA-disulfate. TBBPA (>97% purity) was purchased from Sigma-Aldrich. TBBPA-sulfate, and TBBPA-disulfate were synthesized by the laboratory.Kinetic assay: 14C-TBBPA was synthesized in laboratory and confirmed via multiple analytical techniques (HPLC, NMR, etc).
	Metric 3:	Test Substance Purity	High	Tadpole morphology assay and TH reporter assay: TBBPA (>97% purity) was purchased from Sigma-Aldrich. TBBPA-sulfate, and TBBPA-disulfate were synthesized by the laboratory.Kinetic assay: 14C-TBBPA was synthesized in laboratory and confirmed via multiple analytical techniques (HPLC, NMR, etc). Purity confirmed >99.8% via HPLC.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Tadpole morphology assay: Details are missing in the methods, but authors indicated a control group in Figure 3. Moreover, it is unclear if the control group was a solvent control (ethanol) since as per the authors TBBPA, in ethanol, was added to dechlorinated water. Also, there was no indication of the % ethanol used.
	Metric 5:	Negative Control Response	High	Tadpole morphology assay: The biological response (head areas) of the control group were reported and appeared appropriate.
	Metric 6:	Randomized Allocation	Low	Randomized allocation not stated.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study lacked details on how the exposure solutions were prepared for the various assessments. Some details appear in the methods, others in the figure captions or results, but overall details were missing.
	Metric 8:	Consistency of Exposure Administration	High	Evidence suggests that exposure administration was administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	TBBPA was measured in water and tadpoles as part of the kinetic assessment of the study.
	Metric 10:	Exposure Duration and Frequency	High	Tadpole morphology assay: The exposure duration (6 days) was reported and appropriate for the organism age (larval, NF45 tadpoles).

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Study Citation:	Fini, J. B., Riu, A., Debrauwer, L., Hillenweck, A., Mével, Le, S., Chevolleau, S., Boulahtouf, A., Palmier, K., Balaguer, P., Cravedi, J. P., Demeneix, B. A., Zalko, D. (2012). Parallel biotransformation of tetrabromobisphenol A in Xenopus laevis and mammals: Xenopus as a model for endocrine perturbation studies. Toxicological Sciences 125(2):359-367.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus laevis*; Larvae
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1293799

Domain	Metric		Rating	Comments
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	The goal of the study was not to have a dose-dependent effect; study had a single TBBBA dose and a single metabolite dose.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentration was below the solubility limit; TBBPA exposure concentration was 10^-6 M = 0.5439 mg/L and TBBPA's solubility limit is 4.15 mg/L at 25 C pH 7.56.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	Test organisms were adequately described (X. laevis NF45 tadpoles). However, the authors did not specify if organisms were from a X. laevis culture they maintained or obtained from an independent source for the experiments.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized.
	Metric 15:	Number of Organisms and Replicates per Group	Low	Tadpole morphology assay: Information was reported in the methods and Figure 3 caption. Fifteen NF45 tadpoles were added per well (experimental unit). No replicate wells were included, but authors indicated that experiments were performed twice in the caption of Figure 3.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	The care and treatment of animals were in accordance with institutional and national guidelines (Charte Nationale Portant sur l'Ethique de l'Expe´rimentation Animale, 2009).
	Metric 17:	Outcome Assessment Methodology	High	Tadpole morphology assay: The outcome assessment methodology (measurement of tadpoles' head areas) was reported and appropriate for the intended outcome of interest (morphological effects of TBBPA during development).
	Metric 18:	Consistency of Outcome Assessment	High	The outcome assessment was consistent across 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.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were described (one-way ANOVA followed by Dunn's test).
	Metric 22:	Reporting of Data	High	Tadpole morphology assay: Data were presented for each control and treatment groups (Figure 3).

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Study Citation:	Fini, J. B., Riu, A., Debrauwer, L., Hillenweck, A., Mével, Le, S., Chevolleau, S., Boulahtouf, A., Palmier, K., Balaguer, P., Cravedi, J. P., Demeneix, B. A., Zalko, D. (2012). Parallel biotransformation of tetrabromobisphenol A in Xenopus laevis and mammals: Xenopus as a model for endocrine perturbation studies. Toxicological Sciences 125(2):359-367.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus laevis*; Larvae
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1293799

Domain	Metric		Rating	Comments
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This study examined thyroid disruption effects of TBBPA in Xenopus laevis. To do this the authors (1) assessed the kinetics of TBBPA (uptake and excretion) in X. laevis tadpoles, (2) investigated if antithyroid effects are due to the parent TBBPA or a metabolite using two readouts: tadpole morphology and an in vivo TH reporter assay, and (3) examined the ability of sulfate metabolites andTBBPA to exert direct actions on the TRalpha receptor using a reporter in vitro cell assay. This form was used to evaluate the tadpole morphology assay following a 6-day TBBPA exposure.

Overall Quality Determination High

Study Citation:	Mengeling, B. J., Wei, Y., Dobrawa, L. N., Streekstra, M., Louisse, J., Singh, V., Singh, L., Lein, P. J., Wulff, H., Murk, A. J., Furlow, J. D. (2017). A multi-tiered, in vivo, quantitative assay suite for environmental disruptors of thyroid hormone signaling. Aquatic Toxicology 190:1-10.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus laevis*; TRE-Luciferase reporter (transgenic); Larvae
Health Outcome:	Mechanistic-Receptor binding/ regulation of receptor activity
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3975102

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Accepted common name and definitive chemical structure are given.
	Metric 2:	Test Substance Source	Low	Source of test substance was reported as Sigma-Aldrich (St.Louis, MO), but identity was not analytically verified
	Metric 3:	Test Substance Purity	Low	No purity was reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Controls were reported and were vehicle controls with 0.2% DMSO.
	Metric 5:	Negative Control Response	High	Control morphological responses were reported in Fig 7A,B,C. Survival was reported to be impacted at 2 uM and results for morphology were not conducted due to mortality. Survival was not reported for 0.5 and 1 uM. "Unless mentioned specifically in the results (e.g. 500 nM BDE-47 or 2 μM TBBPA), treatment conditions did not result in animal mortality."
	Metric 6:	Randomized Allocation	Low	Random allocation was not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	Renewal was conducted every 2 days for the 5 day exposure period. The methods of stock solution and concentrations for TBBPA were not detailed. The methods do not report TBBPA treatment concentrations.
	Metric 8:	Consistency of Exposure Administration	High	Exposure conditions among treatment and control appear to be consistently applied.
	Metric 9:	Measurement of Test Substance Concentration	Low	Compound concentrations are reported as nominal, no verification performed.
	Metric 10:	Exposure Duration and Frequency	High	The exposure spans a 5 day period at 10 days post fertilization to observe morphological and cellular impacts of the test compound.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	There are only two treatment concentrations as the highest (2 uM) produced mortality. The authors do not report any metrics of mortality at this concentration. Treatment concentrations are 0.5 and 1 uM.
	Metric 12:	Testing at or Below Solubility Limit	High	The treatment concentrations are within the solubility of TBBPA as reported in the final scope (4.1 g/L).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	Source was not reported. Age and culture methods for larvae in the study were reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Medium	The larval conditions during exposure are consistent with pre-treatment conditions. Density of larvae was different before the exposure period.

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Study Citation:	Mengeling, B. J., Wei, Y., Dobrawa, L. N., Streekstra, M., Louisse, J., Singh, V., Singh, L., Lein, P. J., Wulff, H., Murk, A. J., Furlow, J. D. (2017). A multi-tiered, in vivo, quantitative assay suite for environmental disruptors of thyroid hormone signaling. Aquatic Toxicology 190:1-10.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus laevis*; TRE-Luciferase reporter (transgenic); Larvae
Health Outcome:	Mechanistic-Receptor binding/ regulation of receptor activity
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3975102

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Medium	There were triplicates at each concentration and control with 5 tadpoles per replicate.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	This study is proposing and validating an alternative approach to the Amphibian Metamorphosis Assay (AMA) which is OECD 231. The authors used stages before feeding, and had shorter exposure periods that were initiated earlier when compared to OECD 231. Authors did not report initial water quality parameters but reported using 0.1x Marks modified ringer solution for all incubations.
	Metric 17:	Outcome Assessment Methodology	High	Methods section 2.3 details the morphology endpoints, Section 2.4 details the immunochemistry endpoints, and Section 2.5 details the use of transgenic frogs with TRE-Luciferase reporters of GFP for TH responsiveness.
	Metric 18:	Consistency of Outcome Assessment	High	outcome assessment appear to be consistent among treatment and controls.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	No differences were reported to indicate environmental or other factors influencing outcome assessment.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	Nothing was reported to indicate differences based on animal attrition of health outcomes.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	1-Way ANOVA with Dunnet's or Sidak's Multiple Comparison Test (MCT) was used to determine significance.
	Metric 22:	Reporting of Data	High	Results are presented as means and variance components in figure 7.
	Metric 23:	Explanation of Unexpected Outcomes	High	No unexpected outcomes were reported.

Additional Comments: This form represents the TRE-Luciferace reporter frogs (transgenic) for TH responsiveness endpoints in the paper. Results are presented for 0, 0.5, and 1 uM TBBPA exposures for cell proliferation in figure 7E.

Overall Quality Determination High

Study Citation:	Mengeling, B. J., Wei, Y., Dobrawa, L. N., Streekstra, M., Louisse, J., Singh, V., Singh, L., Lein, P. J., Wulff, H., Murk, A. J., Furlow, J. D. (2017). A multi-tiered, in vivo, quantitative assay suite for environmental disruptors of thyroid hormone signaling. Aquatic Toxicology 190:1-10.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus laevis*; Larvae
Health Outcome:	Mechanistic-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3975102

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Accepted common name and definitive chemical structure are given.
	Metric 2:	Test Substance Source	Low	Source of test substance was reported as Sigma-Aldrich (St.Louis, MO), but identity was not analytically verified
	Metric 3:	Test Substance Purity	Low	No purity was reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Controls were reported and were vehicle controls with 0.2% DMSO.
	Metric 5:	Negative Control Response	High	Control morphological responses were reported in Fig 7A,B,C. Survival was reported to be impacted at 2 uM and results for morphology were not conducted due to mortality. Survival was not reported for 0.5 and 1 uM. "Unless mentioned specifically in the results (e.g. 500 nM BDE-47 or 2 μM TBBPA), treatment conditions did not result in animal mortality."
	Metric 6:	Randomized Allocation	Low	Random allocation was not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	Renewal was conducted every 2 days for the 5 day exposure period. The methods of stock solution and concentrations for TBBPA were not detailed. The methods do not report TBBPA treatment concentrations.
	Metric 8:	Consistency of Exposure Administration	High	Exposure conditions among treatment and control appear to be consistently applied.
	Metric 9:	Measurement of Test Substance Concentration	Low	Compound concentrations are reported as nominal, no verification performed.
	Metric 10:	Exposure Duration and Frequency	High	The exposure spans a 5 day period at 10 days post fertilization to observe morphological and cellular impacts of the test compound.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	There are only two treatment concentrations as the highest (2 uM) produced mortality. The authors do not report any metrics of mortality at this concentration. Treatment concentrations are 0.5 and 1 uM.
	Metric 12:	Testing at or Below Solubility Limit	High	The treatment concentrations are within the solubility of TBBPA as reported in the final scope (4.1 g/L).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	Source was not reported. Age and culture methods for larvae in the study were reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Medium	The larval conditions during exposure are consistent with pre-treatment conditions. Density of larvae was different before the exposure period.

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Study Citation:	Mengeling, B. J., Wei, Y., Dobrawa, L. N., Streekstra, M., Louisse, J., Singh, V., Singh, L., Lein, P. J., Wulff, H., Murk, A. J., Furlow, J. D. (2017). A multi-tiered, in vivo, quantitative assay suite for environmental disruptors of thyroid hormone signaling. Aquatic Toxicology 190:1-10.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus laevis*; Larvae
Health Outcome:	Mechanistic-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3975102

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Medium	There were triplicates at each concentration and control with 5 tadpoles per replicate.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	This study is proposing and validating an alternative approach to the Amphibian Metamorphosis Assay (AMA) which is OECD 231. The authors used stages before feeding, and had shorter exposure periods that were initiated earlier when compared to OECD 231. Authors did not report initial water quality parameters but reported using 0.1x Marks modified ringer solution for all incubations.
	Metric 17:	Outcome Assessment Methodology	High	Methods section 2.3 details the morphology endpoints, Section 2.4 details the immunochemistry endpoints, and Section 2.5 details the use of transgenic frogs with TRE-Luciferase reporters of GFP for TH responsiveness.
	Metric 18:	Consistency of Outcome Assessment	High	outcome assessment appear to be consistent among treatment and controls.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	No differences were reported to indicate environmental or other factors influencing outcome assessment.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	Nothing was reported to indicate differences based on animal attrition of health outcomes.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	1-Way ANOVA with Dunnet's or Sidak's Multiple Comparison Test (MCT) was used to determine significance.
	Metric 22:	Reporting of Data	High	Results are presented as means and variance components in figure 7.
	Metric 23:	Explanation of Unexpected Outcomes	High	No unexpected outcomes were reported.

Additional Comments: This form represents the cell proliferation endpoints in the paper. Results are presented for 0, 0.5, and 1 uM TBBPA exposures for cell proliferation in figure 7D.

Overall Quality Determination High

Study Citation:	Mengeling, B. J., Wei, Y., Dobrawa, L. N., Streekstra, M., Louisse, J., Singh, V., Singh, L., Lein, P. J., Wulff, H., Murk, A. J., Furlow, J. D. (2017). A multi-tiered, in vivo, quantitative assay suite for environmental disruptors of thyroid hormone signaling. Aquatic Toxicology 190:1-10.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus laevis*; Larvae
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3975102

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Accepted common name and definitive chemical structure are given.
	Metric 2:	Test Substance Source	Low	Source of test substance was reported as Sigma-Aldrich (St.Louis, MO), but identity was not analytically verified
	Metric 3:	Test Substance Purity	Low	No purity was reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Controls were reported and were vehicle controls with 0.2% DMSO.
	Metric 5:	Negative Control Response	High	Control morphological responses were reported in Fig 7A,B,C. Survival was reported to be impacted at 2 uM and results for morphology were not conducted due to mortality. Survival was not reported for 0.5 and 1 uM. "Unless mentioned specifically in the results (e.g. 500 nM BDE-47 or 2 μM TBBPA), treatment conditions did not result in animal mortality."
	Metric 6:	Randomized Allocation	Low	Random allocation was not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	Renewal was conducted every 2 days for the 5 day exposure period. The methods of stock solution and concentrations for TBBPA were not detailed. The methods do not report TBBPA treatment concentrations.
	Metric 8:	Consistency of Exposure Administration	High	Exposure conditions among treatment and control appear to be consistently applied.
	Metric 9:	Measurement of Test Substance Concentration	Low	Compound concentrations are reported as nominal, no verification performed.
	Metric 10:	Exposure Duration and Frequency	High	The exposure spans a 5 day period at 10 days post fertilization to observe morphological and cellular impacts of the test compound.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	There are only two treatment concentrations as the highest (2 uM) produced mortality. The authors do not report any metrics of mortality at this concentration. Treatment concentrations are 0.5 and 1 uM.
	Metric 12:	Testing at or Below Solubility Limit	High	The treatment concentrations are within the solubility of TBBPA as reported in the final scope (4.1 g/L).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	Source was not reported. Age and culture methods for larvae in the study were reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	Medium	The larval conditions during exposure are consistent with pre-treatment conditions. Density of larvae was different before the exposure period.

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Study Citation:	Mengeling, B. J., Wei, Y., Dobrawa, L. N., Streekstra, M., Louisse, J., Singh, V., Singh, L., Lein, P. J., Wulff, H., Murk, A. J., Furlow, J. D. (2017). A multi-tiered, in vivo, quantitative assay suite for environmental disruptors of thyroid hormone signaling. Aquatic Toxicology 190:1-10.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus laevis*; Larvae
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3975102

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Medium	There were triplicates at each concentration and control with 5 tadpoles per replicate.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	This study is proposing and validating an alternative approach to the Amphibian Metamorphosis Assay (AMA) which is OECD 231. The authors used stages before feeding, and had shorter exposure periods that were initiated earlier when compared to OECD 231. Authors did not report initial water quality parameters but reported using 0.1x Marks modified ringer solution for all incubations.
	Metric 17:	Outcome Assessment Methodology	High	Methods section 2.3 details the morphology endpoints, Section 2.4 details the immunochemistry endpoints, and Section 2.5 details the use of transgenic frogs with TRE-Luciferase reporters of GFP for TH responsiveness.
	Metric 18:	Consistency of Outcome Assessment	High	outcome assessment appear to be consistent among treatment and controls.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	No differences were reported to indicate environmental or other factors influencing outcome assessment.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	Nothing was reported to indicate differences based on animal attrition of health outcomes.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	1-Way ANOVA with Dunnet's or Sidak's Multiple Comparison Test (MCT) was used to determine significance.
	Metric 22:	Reporting of Data	High	Results are presented as means and variance components in figure 7.
	Metric 23:	Explanation of Unexpected Outcomes	High	No unexpected outcomes were reported.

Additional Comments: This form represents the Morphology endpoints in the paper. Results are presented for 0, 0.5, and 1 uM TBBPA exposures for Brain, head area, and olfaction organ to brain distance in figures, 7A, 7B, and 7C, respectively.

Overall Quality Determination High

Study Citation:	Zhang, Y. F., Xu, W., Lou, Q. Q., Li, Y. Y., Zhao, Y. X., Wei, W. J., Qin, Z. F., Wang, H. L., Li, J. Z. (2014). Tetrabromobisphenol A disrupts vertebrate development via thyroid hormone signaling pathway in a developmental stage-dependent manner. Environmental Science and Technology 48(14):8227-8234.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus laevis*; Larvae
Health Outcome:	Mechanistic-Cell signaling/function-Gastrointestinal
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2927119

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Compound was only referred to by name, Tetrabromobisphenol A (TBBPA). No structure or CASRN provided.
	Metric 2:	Test Substance Source	Low	Source was provided at Geel Belgium (New Jersey, USA), no analytical verification was conducted by the performing lab.
	Metric 3:	Test Substance Purity	Low	Purity was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	DMSO was used for the preparation of the stock solution, but the authors do not state if the control group contains the DMSO carrier.
	Metric 5:	Negative Control Response	Medium	Histology from T3 and TBBPA treated tadpoles are represented in Figure 2A and the corresponding narrative (pages 3-4 /8). Mortality among treatment and control groups were not described.
	Metric 6:	Randomized Allocation	Low	Random allocation was not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	Water and chemicals were exchanged daily. The exposure system (volume, water used, environmental conditions) were not described. Authors indicated that a previous publication described "housing and breeding conditions" and that publication is HERO ID 2174138, but experimental conditions were not described in the current work. Concentrations of test substance were not measured during the study.
	Metric 8:	Consistency of Exposure Administration	High	Exposure conditions among treatment and control groups were consistent.
	Metric 9:	Measurement of Test Substance Concentration	Medium	TBBPA concentrations (10, 100, 500, 1000 nM) were not measured and are reported as nominal.
	Metric 10:	Exposure Duration and Frequency	High	The methods are not related to OCSSP or OECD test guidelines, however, 6 days of exposure with the test compound (TBBPA) and T3 to induce metamorphosis would allow enough time for measures of morphological change to occur and be measured.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The authors use 10, 100, 500, 1000 nM concentrations and a control. The highest value is 0.53 mg/L. The number of exposure groups and spacing of exposure levels were to address the purpose of the study.

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Study Citation:	Zhang, Y. F., Xu, W., Lou, Q. Q., Li, Y. Y., Zhao, Y. X., Wei, W. J., Qin, Z. F., Wang, H. L., Li, J. Z. (2014). Tetrabromobisphenol A disrupts vertebrate development via thyroid hormone signaling pathway in a developmental stage-dependent manner. Environmental Science and Technology 48(14):8227-8234.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus laevis*; Larvae
Health Outcome:	Mechanistic-Cell signaling/function-Gastrointestinal
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2927119

Domain	Metric		Rating	Comments
	Metric 12:	Testing at or Below Solubility Limit	High	The highest concentration used was 1000nM, which is 0.53 mg/L, which is under the solubility listed in the final scope of 4.1 mg/L.The solvent concentration was appropriate.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The source and morphological stage of the larvae were described.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	Authors did not describe the pre-treatment and treatment water conditions or exposure environments well in the study. There was no mention of acclimatization. The authors referred to a previous publication (HERO 2174138) for "housing and breeding conditions", but it is not clear if this paper also describes the treatment conditions for the current study.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The treatments were performed in triplicate with 10 individuals per replicate. 3/tank (9 total) for histology, 3/tank (9 total) for TUNEL assay, and 4/tank (12 total) for gene expression. developmental stage and hind limb length were assessed for all tadpoles.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions, food and water quality parameters were poorly reported.
	Metric 17:	Outcome Assessment Methodology	High	The outcomes assessment for TUNEL assay were reported in the supplemental notes on page S3-S4/8.
	Metric 18:	Consistency of Outcome Assessment	High	Details of outcome assessments were reported and consistent among treatment and control groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	Nothing was reported among study groups with environmental conditions or other factors, however, the study overall did not describe environmental conditions throughout the study.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	No information in the study suggests that differences were from animal attrition or health outcomes.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Detailed methods for stats are presented in the supplemental notes.
	Metric 22:	Reporting of Data	High	These data are presented as means and SEM in Table S3 (page 8/8) of the supplemental notes.
	Metric 23:	Explanation of Unexpected Outcomes	High	Nothing unexpected was reported.

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Study Citation:	Zhang, Y. F., Xu, W., Lou, Q. Q., Li, Y. Y., Zhao, Y. X., Wei, W. J., Qin, Z. F., Wang, H. L., Li, J. Z. (2014). Tetrabromobisphenol A disrupts vertebrate development via thyroid hormone signaling pathway in a developmental stage-dependent manner. Environmental Science and Technology 48(14):8227-8234.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus laevis*; Larvae
Health Outcome:	Mechanistic-Cell signaling/function-Gastrointestinal
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2927119

Domain	Metric	Rating	Comments
Additional Comments:	This form is for the TUNEL assay for intestinal changes with cell staining. The quantitative results are presented in Table S3 of the supplemental notes.		

Overall Quality Determination Medium

Study Citation:	Zhang, Y. F., Xu, W., Lou, Q. Q., Li, Y. Y., Zhao, Y. X., Wei, W. J., Qin, Z. F., Wang, H. L., Li, J. Z. (2014). Tetrabromobisphenol A disrupts vertebrate development via thyroid hormone signaling pathway in a developmental stage-dependent manner. Environmental Science and Technology 48(14):8227-8234.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus laevis*; Larvae
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2927119

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Compound was only referred to by name, Tetrabromobisphenol A (TBBPA). No structure or CASRN provided.
	Metric 2:	Test Substance Source	Low	Source was provided at Geel Belgium (New Jersey, USA), no analytical verification was conducted by the performing lab.
	Metric 3:	Test Substance Purity	Low	Purity was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	DMSO was used for the preparation of the stock solution, but the authors do not state if the control group contains the DMSO carrier.
	Metric 5:	Negative Control Response	Medium	Control responses for morphology and detailed in Figure 1 and figure S1. Table S2 details hind limb length among control and treatment groups. Mortality among treatment and control groups were not described.
	Metric 6:	Randomized Allocation	Low	Random allocation was not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	Water and chemcials were exchanged daily. The exposure system (volume, water used, environmental conditions) were no described. Authors indicated that a previous publication described "housing and breeding conditions" and that publication is HERO ID 2174138, but experimental conditions were not described in the current work. Concentrations of test substance were not measured during the study,
	Metric 8:	Consistency of Exposure Administration	High	Exposure conditions among treatment and control groups were consistent.
	Metric 9:	Measurement of Test Substance Concentration	Medium	TBBPA concentrations (10, 100, 500, 1000 nM) were not measured and are reported as nominal.
	Metric 10:	Exposure Duration and Frequency	High	The methods are not related to OCSSP or OECD test guidelines, however, 6 days of exposure with the test compound (TBBPA) and T3 to induce metamorphosis would allow enough time for measures of morphological change to occur and be measured.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The authors use 10, 100, 500, 1000 nM concentrations and a control. The highest value is 0.53 mg/L. The number of exposure groups and spacing of exposure levels were adequate to address the purpose of the study.
	Metric 12:	Testing at or Below Solubility Limit	High	The highest concentration used was 1000nM, which is 0.53 mg/L, which is under the solubility listed in the final scope of 4.1 mg/L.The solvent concentration was appropriate.
Domain 4: Test Organism				

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Study Citation:	Zhang, Y. F., Xu, W., Lou, Q. Q., Li, Y. Y., Zhao, Y. X., Wei, W. J., Qin, Z. F., Wang, H. L., Li, J. Z. (2014). Tetrabromobisphenol A disrupts vertebrate development via thyroid hormone signaling pathway in a developmental stage-dependent manner. Environmental Science and Technology 48(14):8227-8234.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus laevis*; Larvae
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2927119

Domain	Metric		Rating	Comments
	Metric 13:	Test Organism Characteristics	High	The source and morphological stage of the larvae were described.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	Authors did not describe the pre-treatment and treatment water conditions or exposure environments well in the study. There was no mention of acclimatization in the study. The authors referred to a previous publication (HERO 2174138) for pre-treatment conditions, but it is not clear if this paper also describes the treatment conditions for the current study.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The treatments were performed in triplicate with 10 individuals per replicate. 3/tank (9 total) for histology, 3/tank (9 total) for TUNEL assay, and 4/tank (12 total) for gene expression. developmental stage and hind limb length were assessed for all tadpoles.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions, food and water quality parameters were poorly reported.
	Metric 17:	Outcome Assessment Methodology	High	Endpoints for morphology (developmental stage and hind limb length), qPCR of target genes, TUNEL assay (histology with cellular markers), and overall histology for intestinal remodeling were well described in the main document as well as the supplemental data/methods.
	Metric 18:	Consistency of Outcome Assessment	High	Details of outcome assessments were reported and consistent among treatment and control groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	Nothing was reported among study groups with environmental conditions or other factors, however, the study overall did not describe environmental conditions throughout the study.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	No information in the study suggests that differences were from animal attrition or health outcomes.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	The supplemental methods and data has a detailed section on the ANOVA and Chi-square analysis for the paper.
	Metric 22:	Reporting of Data	High	Morphology outputs are detailed in Figure 1 and figure S1. Table S2 details hind limb length among control and treatment groups.
	Metric 23:	Explanation of Unexpected Outcomes	High	Nothing unexpected was reported.

Additional Comments: This form is for TBBPA impacts to morphology from 6 day exposure of tadpoles administered T3 to produce metamorphosis. Morphology outputs are detailed in Figure 1 and figure S1. Table S2 details hind limb length among control and treatment groups.

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Study Citation:	Zhang, Y. F., Xu, W., Lou, Q. Q., Li, Y. Y., Zhao, Y. X., Wei, W. J., Qin, Z. F., Wang, H. L., Li, J. Z. (2014). Tetrabromobisphenol A disrupts vertebrate development via thyroid hormone signaling pathway in a developmental stage-dependent manner. Environmental Science and Technology 48(14):8227-8234.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus laevis*; Larvae
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2927119

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

Study Citation:	Zhang, Y. F., Xu, W., Lou, Q. Q., Li, Y. Y., Zhao, Y. X., Wei, W. J., Qin, Z. F., Wang, H. L., Li, J. Z. (2014). Tetrabromobisphenol A disrupts vertebrate development via thyroid hormone signaling pathway in a developmental stage-dependent manner. Environmental Science and Technology 48(14):8227-8234.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus laevis*; Larvae
Health Outcome:	Mechanistic-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2927119

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Compound was only referred to by name, Tetrabromobisphenol A (TBBPA). No structure or CASRN provided.
	Metric 2:	Test Substance Source	Low	Source was provided at Geel Belgium (New Jersey, USA), no analytical verification was conducted by the performing lab.
	Metric 3:	Test Substance Purity	Low	Purity was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	DMSO was used for the preparation of the stock solution, but the authors do not state if the control group contains the DMSO carrier.
	Metric 5:	Negative Control Response	Medium	Control responses were detailed for qPCR genes of interest in Figure 3. Mortality among treatment and control groups were not described.
	Metric 6:	Randomized Allocation	Low	Random allocation was not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	Water and chemicals were exchanged daily. The exposure system (volume, water used, environmental conditions) were not described. Authors indicated that a previous publication described "housing and breeding conditions" and that publication is HERO ID 2174138, but experimental conditions were not described in the current work. Concentrations of test substance were not measured during the study.
	Metric 8:	Consistency of Exposure Administration	High	Exposure conditions among treatment and control groups were consistent.
	Metric 9:	Measurement of Test Substance Concentration	Medium	TBBPA concentrations (10, 100, 500, 1000 nM) were not measured and are reported as nominal.
	Metric 10:	Exposure Duration and Frequency	High	The methods are not related to OCSSP or OECD test guidelines, however, 6 days of exposure with the test compound (TBBPA) and T3 to induce metamorphosis would allow enough time for measures of morphological change to occur and be measured.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The authors use 10, 100, 500, 1000 nM concentrations and a control. The highest value is 0.53 mg/L. The number of exposure groups and spacing of exposure levels were adequate to address the purpose of the study.
	Metric 12:	Testing at or Below Solubility Limit	High	The highest concentration used was 1000nM, which is 0.53 mg/L, which is under the solubility listed in the final scope of 4.1 mg/L. The solvent concentration was appropriate ((i.e., no effects on biological responses were observed in the solvent control and no interactions were expected between the solvent and test substance).
Domain 4: Test Organism				

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Study Citation:	Zhang, Y. F., Xu, W., Lou, Q. Q., Li, Y. Y., Zhao, Y. X., Wei, W. J., Qin, Z. F., Wang, H. L., Li, J. Z. (2014). Tetrabromobisphenol A disrupts vertebrate development via thyroid hormone signaling pathway in a developmental stage-dependent manner. Environmental Science and Technology 48(14):8227-8234.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus laevis*; Larvae
Health Outcome:	Mechanistic-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2927119

Domain	Metric		Rating	Comments
	Metric 13:	Test Organism Characteristics	High	The source and morphological stage of the larvae were described.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	Authors did not describe the pre-treatment and treatment water conditions or exposure environments well in the study. There was no mention of acclimatization in the study. The authors referred to a previous publication (HERO 2174138) for "housing and breeding conditions", but it is not clear if this paper also describes the treatment conditions for the current study.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The treatments were performed in triplicate with 10 individuals per replicate. 3/tank (9 total) for histology, 3/tank (9 total) for TUNEL assay, and 4/tank (12 total) for gene expression. developmental stage and hind limb length were assessed for all tadpoles.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions, food and water quality parameters were poorly reported.
	Metric 17:	Outcome Assessment Methodology	High	Endpoints for morphology (developmental stage and hind limb length), qPCR of target genes, TUNEL assay (histology with cellular markers), and overall histology for intestinal remodeling were well described in the main document as well as the supplemental data/methods.
	Metric 18:	Consistency of Outcome Assessment	High	Details of outcome assessments were reported and consistent among treatment and control groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	Nothing was reported among study groups with environmental conditions or other factors, however, the study overall did not describe environmental conditions throughout the study.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	No information in the study suggests that differences were from animal attrition or health outcomes.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	The supplemental methods and data has a detailed section on the ANOVA and Chi-square analysis for the paper.
	Metric 22:	Reporting of Data	High	Figure 3 (page 4/8) presents gene activity at control and treatment concentrations.
	Metric 23:	Explanation of Unexpected Outcomes	High	Nothing unexpected was reported.

Additional Comments: This form is for TBBPA impacts to gene expression from 6 day exposure of tadpoles administered T3 to produce metamorphosis. Figure 3 (page 4/8) presents gene activity at control and treatment concentrations.

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Study Citation:	Zhang, Y. F., Xu, W., Lou, Q. Q., Li, Y. Y., Zhao, Y. X., Wei, W. J., Qin, Z. F., Wang, H. L., Li, J. Z. (2014). Tetrabromobisphenol A disrupts vertebrate development via thyroid hormone signaling pathway in a developmental stage-dependent manner. Environmental Science and Technology 48(14):8227-8234.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus laevis*; Larvae
Health Outcome:	Mechanistic-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2927119

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

Study Citation:	Zhang, Y. F., Xu, W., Lou, Q. Q., Li, Y. Y., Zhao, Y. X., Wei, W. J., Qin, Z. F., Wang, H. L., Li, J. Z. (2014). Tetrabromobisphenol A disrupts vertebrate development via thyroid hormone signaling pathway in a developmental stage-dependent manner. Environmental Science and Technology 48(14):8227-8234.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus laevis*; Larvae
Health Outcome:	Gastrointestinal
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2927119

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Compound was only referred to by name, Tetrabromobisphenol A (TBBPA). No structure or CASRN provided.
	Metric 2:	Test Substance Source	Low	Source was provided at Geel Belgium (New Jersey, USA), no analytical verification was conducted by the performing lab.
	Metric 3:	Test Substance Purity	Low	Purity was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	DMSO was used for the preparation of the stock solution, but the authors do not state if the control group contains the DMSO carrier.
	Metric 5:	Negative Control Response	Medium	Histology from T3 and TBBPA treated tadpoles are represented in Figure 2A and the corresponding narrative (pages 3-4 /8). Mortality among treatment and control groups were not described.
	Metric 6:	Randomized Allocation	Low	Random allocation was not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	Water and chemicals were exchanged daily. The exposure system (volume, water used, environmental conditions) were no described. Authors indicated that a previous publication described "housing and breeding conditions" and that publication is HERO ID 2174138, but experimental conditions were not described in the current work. Concentrations of test substance were not measured during the study
	Metric 8:	Consistency of Exposure Administration	High	Exposure conditions among treatment and control groups were consistent.
	Metric 9:	Measurement of Test Substance Concentration	Medium	TBBPA concentrations (10, 100, 500, 1000 nM) were not measured and are reported as nominal.
	Metric 10:	Exposure Duration and Frequency	High	The methods are not related to OCSSP or OECD test guidelines, however, 6 days of exposure with the test compound (TBBPA) and T3 to induce metamorphosis would allow enough time for measures of morphological change to occur and be measured.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The authors use 10, 100, 500, 1000 nM concentrations and a control. The highest value is 0.53 mg/L. The number of exposure groups and spacing of exposure levels were adequate to address the purpose of the study.
	Metric 12:	Testing at or Below Solubility Limit	High	The highest concentration used was 1000nM, which is 0.53 mg/L, which is under the solubility listed in the final scope of 4.1 mg/L.The solvent concentration was appropriate.
Domain 4: Test Organism				

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Study Citation:	Zhang, Y. F., Xu, W., Lou, Q. Q., Li, Y. Y., Zhao, Y. X., Wei, W. J., Qin, Z. F., Wang, H. L., Li, J. Z. (2014). Tetrabromobisphenol A disrupts vertebrate development via thyroid hormone signaling pathway in a developmental stage-dependent manner. Environmental Science and Technology 48(14):8227-8234.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus laevis*; Larvae
Health Outcome:	Gastrointestinal
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2927119

Domain	Metric		Rating	Comments
	Metric 13:	Test Organism Characteristics	High	The source and morphological stage of the larvae were described.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	Authors did not describe the pre-treatment and treatment water conditions or exposure environments well in the study. there was no mention of acclimatization. The authors referred to a previous publication (HERO 2174138) for "housing and breeding conditions", but it is not clear if this paper also describes the treatment conditions for the current study.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The treatments were performed in triplicate with 10 individuals per replicate. 3/tank (9 total) for histology, 3/tank (9 total) for TUNEL assay, and 4/tank (12 total) for gene expression. developmental stage and hind limb length were assessed for all tadpoles.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	The reporting of environmental conditions, food, water quality parameters are poorly reported.
	Metric 17:	Outcome Assessment Methodology	Low	The outcomes assessment for intestine histology were qualitative and observational. Detailed assessment criteria were not reported.
	Metric 18:	Consistency of Outcome Assessment	High	Details of outcome assessments were reported and consistent among treatment and control groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	Nothing was reported among study groups with environmental conditions or other factors, however, the study overall did not describe environmental conditions throughout the study.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	No information in the study suggests that differences were from animal attrition or health outcomes.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	N/A	These results were qualitative and presented in the narrative of the results section.
	Metric 22:	Reporting of Data	Low	These are observation responses based on an assessment of intestinal morphology after 6 days of metamorphosis.
	Metric 23:	Explanation of Unexpected Outcomes	High	Nothing unexpected was reported.

Additional Comments: This form is for histology of the intestine in figure 2A and a qualitative description is presented in the narrative in the last paragraph on page 3.

Overall Quality Determination Medium

Study Citation:	Jagnytsch, O., Opitz, R., Lutz, I., Kloas, W. (2006). Effects of tetrabromobisphenol A on larval development and thyroid hormone-regulated biomarkers of the amphibian Xenopus laevis. Environmental Research 101(3):340-348.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus laevis*; Juvenile
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1298685

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	Test substance was obtained from Sigma, but the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent solvent control group and 2 positive controls
	Metric 5:	Negative Control Response	High	The biological response of the solvent control group was suitable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The experimental system and/or test media preparation methods were adequately reported. Reporting omissions were unlikely to have a substantial impact on results
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable, as significant differences were seen at the highest test concentration but not at the lower test concentrations.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Low	The numbers of test organisms (20 tadpoles/tank) and replicates were lower than the typical number used in studies of the same or similar type but sufficient for statistical analysis

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Study Citation:	Jagnytsch, O., Opitz, R., Lutz, I., Kloas, W. (2006). Effects of tetrabromobisphenol A on larval development and thyroid hormone-regulated biomarkers of the amphibian Xenopus laevis. Environmental Research 101(3):340-348.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus laevis*; Juvenile
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1298685

Domain	Metric		Rating	Comments
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health, as no undue mortality was reported in the controls.
	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 with regard to the environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups unrelated to exposure, as evidenced by the results of the positive controls.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were clearly described: The length data (whole body length, hind limb length, and tail length) were checked for normal distribution (Kolmogorov–Smirnov test) and homogeneity of variance (Levene test). The Dunnett's test was used to compare length data from the SC group to all other treatment groups.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: None

Overall Quality Determination High

Study Citation:	Jagnytsch, O., Opitz, R., Lutz, I., Kloas, W. (2006). Effects of tetrabromobisphenol A on larval development and thyroid hormone-regulated biomarkers of the amphibian Xenopus laevis. Environmental Research 101(3):340-348.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus laevis*; Juvenile
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1298685

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 was obtained from Sigma, but the identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent solvent control group (etOH) and 2 positive controls
	Metric 5:	Negative Control Response	High	The biological response of the solvent control group was suitable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The experimental system and/or test media preparation methods were adequately reported and reporting omissions were unlikely to have a substantial impact on results
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type as evidenced by the significant effects seen in the results.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable, as significant reductions in development parameters were observed in a dose-responsive manner.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Low	The numbers of test organisms and replicates were lower than the typical number used in studies of the same or similar type but sufficient for statistical analysis
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:	Jagnytsch, O., Opitz, R., Lutz, I., Kloas, W. (2006). Effects of tetrabromobisphenol A on larval development and thyroid hormone-regulated biomarkers of the amphibian Xenopus laevis. Environmental Research 101(3):340-348.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus laevis*; Juvenile
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1298685

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 clearly described
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: None

Overall Quality Determination High

Study Citation:	Zhang, Y. F., Xu, W., Lou, Q. Q., Li, Y. Y., Zhao, Y. X., Wei, W. J., Qin, Z. F., Wang, H. L., Li, J. Z. (2014). Tetrabromobisphenol A disrupts vertebrate development via thyroid hormone signaling pathway in a developmental stage-dependent manner. Environmental Science and Technology 48(14):8227-8234.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus laevis*; Larvae
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2927119

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Compound was only referred to by name, Tetrabromobisphenol A (TBBPA). No structure or CASRN provided.
	Metric 2:	Test Substance Source	Low	Source was provided at Geel Belgium (New Jersey, USA), no analytical verification was conducted by the performing lab.
	Metric 3:	Test Substance Purity	Low	Purity was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	DMSO was used for the preparation of the stock solution, but the authors do not state if the control group contains the DMSO carrier.
	Metric 5:	Negative Control Response	Medium	Control responses for % morphological stage and hind limb length are presented in figure 4. Survival was not reported.
	Metric 6:	Randomized Allocation	Low	Random allocation was not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	Water and chemicals were exchanged daily. The exposure system (volume, water used, environmental conditions) were not described. Authors indicated that a previous publication described "housing and breeding conditions" and that publication is HERO ID 2174138, but experimental conditions were not described in the current work. Concentrations of test substance were not measured during the study.
	Metric 8:	Consistency of Exposure Administration	High	Exposure conditions among treatment and control groups were consistent.
	Metric 9:	Measurement of Test Substance Concentration	Medium	TBBPA concentrations (10, 100, 500, 1000 nM) were not measured and are reported as nominal.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was based on the natural progression of metamorphosis in this species.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The authors use 10, 100, 500, 1000 nM concentrations and a control. The highest value is 0.53 mg/L. The number of exposure groups and spacing of exposure levels were adequate to address the purpose of the study.
	Metric 12:	Testing at or Below Solubility Limit	High	The highest concentration used was 1000nM, which is 0.53 mg/L, which is under the solubility listed in the final scope of 4.1 mg/L.The solvent concentration was appropriate.
Domain 4: Test Organism				

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Study Citation:	Zhang, Y. F., Xu, W., Lou, Q. Q., Li, Y. Y., Zhao, Y. X., Wei, W. J., Qin, Z. F., Wang, H. L., Li, J. Z. (2014). Tetrabromobisphenol A disrupts vertebrate development via thyroid hormone signaling pathway in a developmental stage-dependent manner. Environmental Science and Technology 48(14):8227-8234.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus laevis*; Larvae
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2927119

Domain	Metric		Rating	Comments
	Metric 13:	Test Organism Characteristics	High	The source and morphological stage of the larvae were described.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	Authors did not describe the pre-treatment and treatment water conditions or exposure environments well in the study. There was no mention of acclimatization in the study. The authors referred to a previous publication (HERO 2174138) for "housing and breeding", but it is not clear if this paper also describes the treatment conditions for the current study.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The treatments were performed in triplicate with 10 individuals per replicate and the experiment was conduced three times. Data for each was presented.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	The reporting of environmental conditions, food, water quality parameters are poorly reported.
	Metric 17:	Outcome Assessment Methodology	High	Endpoints for morphology (developmental stage and hind limb length), qPCR of target genes, TUNEL assay (histology with cellular markers), and overall histology for intestinal remodeling were well described in the main document as well as the supplemental data/methods.
	Metric 18:	Consistency of Outcome Assessment	High	Details of outcome assessments were reported and consistent among treatment and control groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	Nothing was reported among study groups with environmental conditions or other factors, however, the study overall did not describe environmental conditions throughout the study.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	No information in the study suggests that differences were from animal attrition or health outcomes.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	The supplemental methods and data has a detailed section on the ANOVA and Chi-square analysis for the paper.
	Metric 22:	Reporting of Data	High	Hind limb length (Mean and SEM) and % morphological stage at 7, 14, and 21 days of exposure are presented in figure 4 with stats.
	Metric 23:	Explanation of Unexpected Outcomes	High	Nothing unexpected was reported.

Additional Comments: This form is for TBBPA impacts to morphology/metamorphosis from 21 day exposure to TBBPA concentrations starting with tadpoles at stage 51. The experiment was conducted three times and hindlimb and % stages at days 7, 14, and 21 days are presented in figure 4 on page 5/8.

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Study Citation:	Zhang, Y. F., Xu, W., Lou, Q. Q., Li, Y. Y., Zhao, Y. X., Wei, W. J., Qin, Z. F., Wang, H. L., Li, J. Z. (2014). Tetrabromobisphenol A disrupts vertebrate development via thyroid hormone signaling pathway in a developmental stage-dependent manner. Environmental Science and Technology 48(14):8227-8234.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus laevis*; Larvae
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2927119

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

Study Citation:	Zhang, Y. F., Xu, W., Lou, Q. Q., Li, Y. Y., Zhao, Y. X., Wei, W. J., Qin, Z. F., Wang, H. L., Li, J. Z. (2014). Tetrabromobisphenol A disrupts vertebrate development via thyroid hormone signaling pathway in a developmental stage-dependent manner. Environmental Science and Technology 48(14):8227-8234.
Duration:	Overall Duration: > 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus laevis*; Larvae
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2927119

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Compound was only referred to by name, Tetrabromobisphenol A (TBBPA). No structure or CASRN provided.
	Metric 2:	Test Substance Source	Low	Source was provided at Geel Belgium (New Jersey, USA), no analytical verification was conducted by the performing lab.
	Metric 3:	Test Substance Purity	Low	Purity was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	DMSO was used for the preparation of the stock solution, but the authors do not state if the control group contains the DMSO carrier.
	Metric 5:	Negative Control Response	Medium	Control responses for % morphological stage and hind limb length are presented in figure 5. Survival was not reported.
	Metric 6:	Randomized Allocation	Low	Random allocation was not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	Water and chemicals were exchanged daily. The exposure system (volume, water used, environmental conditions) were not described. Authors indicated that a previous publication described "housing and breeding conditions" and that publication is HERO ID 2174138, but experimental conditions were not described in the current work. Concentrations of test substance were not measured during the study.
	Metric 8:	Consistency of Exposure Administration	High	Exposure conditions among treatment and control groups were consistent.
	Metric 9:	Measurement of Test Substance Concentration	Medium	TBBPA concentrations (10, 100, 500, 1000 nM) were not measured and are reported as nominal.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure were dependent on natural progression of metamorphosis in this species.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The authors use 10, 100, 500, 1000 nM concentrations and a control. The highest value is 0.53 mg/L. The number of exposure groups and spacing of exposure levels were adequate to address the purpose of the study.
	Metric 12:	Testing at or Below Solubility Limit	High	The highest concentration used was 1000nM, which is 0.53 mg/L, which is under the solubility listed in the final scope of 4.1 mg/L.The solvent concentration was appropriate.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The source and morphological stage of the larvae were described.

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Study Citation:	Zhang, Y. F., Xu, W., Lou, Q. Q., Li, Y. Y., Zhao, Y. X., Wei, W. J., Qin, Z. F., Wang, H. L., Li, J. Z. (2014). Tetrabromobisphenol A disrupts vertebrate development via thyroid hormone signaling pathway in a developmental stage-dependent manner. Environmental Science and Technology 48(14):8227-8234.
Duration:	Overall Duration: > 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus laevis*; Larvae
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2927119

Domain	Metric		Rating	Comments
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	Authors did not describe the pre-treatment and treatment water conditions or exposure environments well in the study. There was no mention of acclimatization in the study. The authors referred to a previous publication (HERO 2174138) for "housing and breeding conditions", but it is not clear if this paper also describes the treatment conditions for the current study.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The treatments were performed in triplicate with 10 individuals per replicate. This experiment was conducted once.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	The reporting of environmental conditions, food, water quality parameters are poorly reported.
	Metric 17:	Outcome Assessment Methodology	High	Endpoints for morphology (developmental stage and hind limb length), qPCR of target genes, TUNEL assay (histology with cellular markers), and overall histology for intestinal remodeling were well described in the main document as well as the supplemental data/methods.
	Metric 18:	Consistency of Outcome Assessment	High	Details of outcome assessments were reported and consistent among treatment and control groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	Nothing was reported among study groups with environmental conditions or other factors, however, the study overall did not describe environmental conditions throughout the study.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	No information in the study suggests that differences were from animal attrition or health outcomes.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	The supplemental methods and data has a detailed section on the ANOVA and Chi-square analysis for the paper.
	Metric 22:	Reporting of Data	High	The results of these data are presented in the narrative and on page 6/8 in figure 5 as % tadpoles at morphological stages after 14 days exposure, and time from stage 57 to 66 in mean days with SEM.
	Metric 23:	Explanation of Unexpected Outcomes	High	Nothing unexpected was reported.

Additional Comments: This form is for TBBPA impacts to morphology/metamorphosis from stage 57 tadpoles, which the authors described as near morphological climax. The results of these data are presented in the narrative and on page 6/8 in figure 5 as % tadpoles at morphological stages after 14 days exposure, and time from stage 57 to 66 in mean days with SEM.

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Study Citation:	Zhang, Y. F., Xu, W., Lou, Q. Q., Li, Y. Y., Zhao, Y. X., Wei, W. J., Qin, Z. F., Wang, H. L., Li, J. Z. (2014). Tetrabromobisphenol A disrupts vertebrate development via thyroid hormone signaling pathway in a developmental stage-dependent manner. Environmental Science and Technology 48(14):8227-8234.
Duration:	Overall Duration: > 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus laevis*; Larvae
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2927119

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

Study Citation:	Carlsson, G., Norrgren, L. (2014). Comparison of embryo toxicity using two classes of aquatic vertebrates. Environmental Toxicology and Pharmacology 37(1):24-7. [Environmental toxicology and pharmacology].
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus tropicalis*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5518140

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity 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	Low	The biological response of the negative control groups was not reported.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Medium	Reporting omissions were unlikely to have a substantial impact on results
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure 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	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 replicates was not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Reported environmental conditions were not sufficient to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported

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Study Citation:	Carlsson, G., Norrgren, L. (2014). Comparison of embryo toxicity using two classes of aquatic vertebrates. Environmental Toxicology and Pharmacology 37(1):24-7. [Environmental toxicology and pharmacology].
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus tropicalis*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5518140

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22:	Reporting of Data	Low	Data were only reported for some outcomes
	Metric 23:	Explanation of Unexpected Outcomes	Low	insufficient information was provided to determine if excessive variability or unexpected outcomes occurred.

Additional Comments: None

Overall Quality Determination Medium

Study Citation:	Carlsson, G., Norrgren, L. (2014). Comparison of embryo toxicity using two classes of aquatic vertebrates. Environmental Toxicology and Pharmacology 37(1):24-7. [Environmental toxicology and pharmacology].
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus tropicalis*; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5518140

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity 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	Low	The biological response of the negative control groups was not reported.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Medium	Reporting omissions were unlikely to have a substantial impact on results
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure 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	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 replicates was not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Reported environmental conditions were not sufficient to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported.

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Study Citation:	Carlsson, G., Norrgren, L. (2014). Comparison of embryo toxicity using two classes of aquatic vertebrates. Environmental Toxicology and Pharmacology 37(1):24-7. [Environmental toxicology and pharmacology].
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus tropicalis*; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5518140

Domain	Metric		Rating	Comments
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22:	Reporting of Data	Low	Data were only reported for some outcomes
	Metric 23:	Explanation of Unexpected Outcomes	Low	insufficient information was provided to determine if excessive variability or unexpected outcomes occurred.

Additional Comments: None

Overall Quality Determination Medium

Study Citation:	Shi, H., Qian, L., Guo, S., Zhang, X., Liu, J., Cao, Q. (2010). Teratogenic effects of tetrabromobisphenol A on Xenopus tropicalis embryos. Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 152(1):62-68.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus tropicalis*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1403499

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 97%.
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	Medium	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	Methods for preparation of test media were not described in adequate detail (e.g., stock preparation was not reported), but the authors did report using a solvent to dissolve TBBPA (DMSO, concentration <0.1% DMSO).
	Metric 8:	Consistency of Exposure Administration	Low	Details of exposure administration (exposure solution volume, final solvent concentration in test media, etc.) were not adequately reported. Authors reported that test media was renewed every 24 hours but unclear whether this was consistently done for all treatments groups and controls.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported (48 h) and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	The number of exposure groups and spacing of exposure levels were suitable.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit. The solvent concentration was appropriate ((i.e., no effects on biological responses were observed in the solvent control and no interactions were expected between the solvent and test substance).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.

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Study Citation:	Shi, H., Qian, L., Guo, S., Zhang, X., Liu, J., Cao, Q. (2010). Teratogenic effects of tetrabromobisphenol A on Xenopus tropicalis embryos. Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 152(1):62-68.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus tropicalis*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1403499

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects. Three exposures groups and control, 4 replicate dishes per concentration and 20 embryos per dish were used.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest. "The body length was measured from the tip of the head to the tip of the tail using computer-assisted image analysis."
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups. Body length measurement was recorded at 24 hour intervals.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical differences were analyzed using one way ANOVA followed by Dunnett's test. But the authors do not use statistical results to explain the effects of TBBPA on body length (Section 3.1).
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: The study examined the effects of TBBPA on survival, growth and teratogenic effects on Xenopus tropicalis embryos. This evaluation form is relevant for the body length measurements.

Overall Quality Determination Medium

Study Citation:	Shi, H., Qian, L., Guo, S., Zhang, X., Liu, J., Cao, Q. (2010). Teratogenic effects of tetrabromobisphenol A on Xenopus tropicalis embryos. Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 152(1):62-68.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus tropicalis*; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1403499

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 97%.
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	Medium	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	Methods for preparation of test media were not described in adequate detail (e.g., stock preparation was not reported), but the authors did report using a solvent to dissolve TBBPA (DMSO, concentration <0.1% DMSO).
	Metric 8:	Consistency of Exposure Administration	Low	Details of exposure administration (exposure solution volume, final solvent concentration in test media, etc.) were not adequately reported. Authors reported that test media was renewed every 24hours but unclear whether this was consistently done for all treatments groups and controls.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported (48 h) and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	The number of exposure groups and spacing of exposure levels were suitable.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit. The solvent concentration was appropriate ((i.e., no effects on biological responses were observed in the solvent control and no interactions were expected between the solvent and test substance).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	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. Three exposures groups and control, 4 replicate dishes per concentration and 20 embryos per dish were used.

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Study Citation:	Shi, H., Qian, L., Guo, S., Zhang, X., Liu, J., Cao, Q. (2010). Teratogenic effects of tetrabromobisphenol A on Xenopus tropicalis embryos. Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 152(1):62-68.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus tropicalis*; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1403499

Domain	Metric		Rating	Comments
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups. Dead embryos were removed a 24 hour intervals.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical differences were analyzed using one way ANOVA followed by Dunnett's test. But the authors do not use statistical results to explain the effects of TBBPA on survival rate (Section 3.1).
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group (Figure 1).
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: The study examined the effects of TBBPA on survival, growth and teratogenic effects on Xenopus tropicalis embryos. This evaluation form is relevant for the mortality outcome.

Overall Quality Determination Medium

Study Citation:	Shi, H., Qian, L., Guo, S., Zhang, X., Liu, J., Cao, Q. (2010). Teratogenic effects of tetrabromobisphenol A on Xenopus tropicalis embryos. Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 152(1):62-68.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus tropicalis*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1403499

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 97%.
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	Medium	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	Methods for preparation of test media were not described in adequate detail (e.g., stock preparation was not reported), but the authors did report using a solvent to dissolve TBBPA (DMSO, concentration <0.1% DMSO).
	Metric 8:	Consistency of Exposure Administration	Low	Details of exposure administration (exposure solution volume, final solvent concentration in test media, etc.) were not adequately reported. Authors reported that test media was renewed every 24 hours but unclear whether this was consistently done for all treatments groups and controls.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	The number of exposure groups and spacing of exposure levels were suitable.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit. The solvent concentration was appropriate ((i.e., no effects on biological responses were observed in the solvent control and no interactions were expected between the solvent and test substance).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	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. Three exposures groups and control, 4 replicate dishes per concentration and 20 embryos per dish were used.

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Study Citation:	Shi, H., Qian, L., Guo, S., Zhang, X., Liu, J., Cao, Q. (2010). Teratogenic effects of tetrabromobisphenol A on Xenopus tropicalis embryos. Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 152(1):62-68.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Xenopus tropicalis*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1403499

Domain	Metric		Rating	Comments
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health.
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported. The grading of eye malformations were not fully reported. It was unclear whether the same embryos were used for both to examine eye and whole body malformations.
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were confusing. In section 2.5, it was reported that ”15–20 embryos were randomly selected from each grade (0–3) for histological sectioning” to examine eye malformations. But in Figure 5, data was reported as mean of 4 replicates of 20 embryos/concentration. Also, it was unclear whether the same embryos were used for both to examine eye and whole body malformations.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical differences were analyzed using one way ANOVA followed by Dunnett’s test (Fig 5 caption). But the authors do not use statistical results to explain the effects of TBBPA on the incidence on malformations.
	Metric 22:	Reporting of Data	High	Total incidence of malformations and index of eye malformations were quantified for each treatment group and control (Figure 5).
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: The study examined the effects of TBBPA on survival, growth and teratogenic effects on Xenopus tropicalis embryos. This evaluation form is relevant for the histological examination of malformations in embryos.

Overall Quality Determination Medium

Study Citation:	Behl, M., Hsieh, J. H., Shafer, T. J., Mundy, W. R., Rice, J. R., Boyd, W. A., Freedman, J. H., Hunter, E. S., Jarema, K. A., Padilla, S., Tice, R. R. (2015). Use of alternative assays to identify and prioritize organophosphorus flame retardants for potential developmental and neurotoxicity. Neurotoxicology and Teratology 52(Pt B):181-193.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Caenorhabditis elegans*; Bristol N2 wild type; Larvae
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3479540

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent solvent control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided few details on exposure media preparation
	Metric 8:	Consistency of Exposure Administration	Low	The study provided few details on exposure administration
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were justified for a dose response by study authors
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups
	Metric 15:	Number of Organisms and Replicates per Group	Medium	three replicates were used, 50 larvae per well
Domain 5: Outcome Assessment				

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Study Citation:	Behl, M., Hsieh, J. H., Shafer, T. J., Mundy, W. R., Rice, J. R., Boyd, W. A., Freedman, J. H., Hunter, E. S., Jarema, K. A., Padilla, S., Tice, R. R. (2015). Use of alternative assays to identify and prioritize organophosphorus flame retardants for potential developmental and neurotoxicity. Neurotoxicology and Teratology 52(Pt B):181-193.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Caenorhabditis elegans*; Bristol N2 wild type; Larvae
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3479540

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

Additional Comments: None

Overall Quality Determination Medium

Study Citation:	Behl, M., Rice, J. R., Smith, M. V., Co, C. A., Bridge, M. F., Hsieh, J. H., Freedman, J. H., Boyd, W. A. (2016). Editor's highlight: Comparative toxicity of organophosphate flame retardants and polybrominated diphenyl ethers to Caenorhabditis elegans. Toxicological Sciences 154(2):241-252.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Caenorhabditis elegans*; Bristol N2; Larvae
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3975281

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 98.93%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Medium	Reporting omissions are unlikely to have a substantial impact on results
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure 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	solvents at an appropriate level aided in dissolution
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reporting of environmental conditions was not sufficiently reported to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest

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Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Caenorhabditis elegans*; Bristol N2; Larvae
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3975281

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	Medium	outcomes were assessed consistently across study groups but few details were provided
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	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 appropriate for dataset
	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 High

Study Citation:	Behl, M., Rice, J. R., Smith, M. V., Co, C. A., Bridge, M. F., Hsieh, J. H., Freedman, J. H., Boyd, W. A. (2016). Editor's highlight: Comparative toxicity of organophosphate flame retardants and polybrominated diphenyl ethers to Caenorhabditis elegans. Toxicological Sciences 154(2):241-252.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Caenorhabditis elegans*; Bristol N2; Juvenile
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3975281

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 98.93%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Medium	Reporting omissions are unlikely to have a substantial impact on results
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure 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	solvents at an appropriate level aided in dissolution
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reporting of environmental conditions was not sufficiently reported to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	Medium	outcomes were assessed consistently across study groups but few details were provided

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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); 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*; Bristol N2; Juvenile
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3975281

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 appropriate for dataset
	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 evaluation was for the study conducted with young adult C. elegans that were assessed for feeding rate. This falls under a behavioral outcome.

Overall Quality Determination High

Study Citation:	Behl, M., Rice, J. R., Smith, M. V., Co, C. A., Bridge, M. F., Hsieh, J. H., Freedman, J. H., Boyd, W. A. (2016). Editor's highlight: Comparative toxicity of organophosphate flame retardants and polybrominated diphenyl ethers to Caenorhabditis elegans. Toxicological Sciences 154(2):241-252.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Caenorhabditis elegans*; Bristol N2; Larvae
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3975281

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 98.93%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Medium	Reporting omissions are unlikely to have a substantial impact on results
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure 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	solvents at an appropriate level aided in dissolution
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reporting of environmental conditions was not sufficiently reported to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	Medium	outcomes were assessed consistently across study groups but few details were provided

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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); 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*; Bristol N2; Larvae
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3975281

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 appropriate for dataset
	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 High

Study Citation:	Ju, J., Lieke, T., Saul, N., Pu, Y., Yin, L., Kochan, C., Putschew, A., Baberschke, N., Steinberg, C. E. (2014). Neurotoxic evaluation of two organobromine model compounds and natural AOBr-containing surface water samples by a Caenorhabditis elegans test. Ecotoxicology and Environmental Safety 104:194-201.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Caenorhabditis elegans*; Wild Type N2; Larvae
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5497653

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by accepted common name, tetrabromobisphenol-A; TBBP-A.
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	Medium	The biological response of the negative control group was reported and reasonable for reported outcomes, survival or health outcomes were 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 few details on exposure media preparation
	Metric 8:	Consistency of Exposure Administration	Low	The study provided few details on exposure administration
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	Medium	The duration of exposure was reported and suitable for the study type, unclear if longer duration would provide clearer response
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	There is only one exposure concentration with an appropriate solvent concentration.
	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	Medium	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
	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	Reported environmental conditions were not sufficient to evaluate if adequate

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Study Citation:	Ju, J., Lieke, T., Saul, N., Pu, Y., Yin, L., Kochan, C., Putschew, A., Baberschke, N., Steinberg, C. E. (2014). Neurotoxic evaluation of two organobromine model compounds and natural AOBr-containing surface water samples by a Caenorhabditis elegans test. Ecotoxicology and Environmental Safety 104:194-201.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Caenorhabditis elegans*; Wild Type N2; Larvae
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5497653

Domain	Metric		Rating	Comments
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described
	Metric 22:	Reporting of Data	High	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: comparison with contaminated surface waters also reported

Overall Quality Determination Medium

Study Citation:	Liu, F., Zaman, W. Q., Peng, H., Li, C., Cao, X., Huang, K., Cui, C., Zhang, W., Lin, K., Luo, Q. (2019). Ecotoxicity of Caenorhabditis elegans following a step and repeated chronic exposure to tetrabromobisphenol A. Ecotoxicology and Environmental Safety 169:273-281.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater), Terrestrial; Water, 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:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5497487

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was 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	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for this study type
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of replicates was not reported
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Reporting omissions were unlikely to have a substantial impact on results
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest

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Study Citation:	Liu, F., Zaman, W. Q., Peng, H., Li, C., Cao, X., Huang, K., Cui, C., Zhang, W., Lin, K., Luo, Q. (2019). Ecotoxicity of Caenorhabditis elegans following a step and repeated chronic exposure to tetrabromobisphenol A. Ecotoxicology and Environmental Safety 169:273-281.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater), Terrestrial; Water, 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:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5497487

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
	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: None

Overall Quality Determination High

Study Citation:	Liu, F., Zaman, W. Q., Peng, H., Li, C., Cao, X., Huang, K., Cui, C., Zhang, W., Lin, K., Luo, Q. (2019). Ecotoxicity of Caenorhabditis elegans following a step and repeated chronic exposure to tetrabromobisphenol A. Ecotoxicology and Environmental Safety 169:273-281.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater), Terrestrial; Water, 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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5497487

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was 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	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for this study type
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of replicates was not reported
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Reporting omissions were unlikely to have a substantial impact on results
	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				

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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), Terrestrial; Water, 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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5497487

Domain	Metric		Rating	Comments
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	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 High

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Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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*; sod-3; Larvae
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
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Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
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	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	only one concentration tested
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	details of pretreatment were not provided
	Metric 15:	Number of Organisms and Replicates per Group	Low	The numbers of replicates were not reported
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Reporting omissions were unlikely to have a substantial impact on results
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest

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Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5497487

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
	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
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Additional Comments: None

Overall Quality Determination High

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Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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*; sod-3; Larvae
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Oxidative stress (including redox biology)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
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Domain 2: Test Design				
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Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Reporting omissions were unlikely to have a substantial impact on results
	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				

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Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Oxidative stress (including redox biology)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
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Domain	Metric		Rating	Comments
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	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

Overall Quality Determination High

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Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Caenorhabditis elegans*; ctl-2; Larvae
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Oxidative stress (including redox biology)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5497487

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	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
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Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Caenorhabditis elegans*; ctl-2; Larvae
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Oxidative stress (including redox biology)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
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	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups
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Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described
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Additional Comments: None

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Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Caenorhabditis elegans*; wild type; Larvae
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5497487

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
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Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
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	Metric 16:	Adequacy of Test Conditions	Medium	Reporting omissions were unlikely to have a substantial impact on results
	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				

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Study Citation:	Liu, F., Zaman, W. Q., Peng, H., Li, C., Cao, X., Huang, K., Cui, C., Zhang, W., Lin, K., Luo, Q. (2019). Ecotoxicity of Caenorhabditis elegans following a step and repeated chronic exposure to tetrabromobisphenol A. Ecotoxicology and Environmental Safety 169:273-281.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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-Biomarkers (exposure and effect)-Cell signaling/function-Oxidative stress (including redox biology)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5497487

Domain	Metric		Rating	Comments
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	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: gene expression, enzyme activity, ROS activity

Overall Quality Determination High

Study Citation:	Liu, F., Zaman, W. Q., Peng, H., Li, C., Cao, X., Huang, K., Cui, C., Zhang, W., Lin, K., Luo, Q. (2019). Ecotoxicity of Caenorhabditis elegans following a step and repeated chronic exposure to tetrabromobisphenol A. Ecotoxicology and Environmental Safety 169:273-281.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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-Biomarkers (exposure and effect)-Cell signaling/function-Oxidative stress (including redox biology)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5497487

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was 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	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for this study type
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of replicates was not reported
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Reporting omissions were unlikely to have a substantial impact on results
	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				

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Study Citation:	Liu, F., Zaman, W. Q., Peng, H., Li, C., Cao, X., Huang, K., Cui, C., Zhang, W., Lin, K., Luo, Q. (2019). Ecotoxicity of Caenorhabditis elegans following a step and repeated chronic exposure to tetrabromobisphenol A. Ecotoxicology and Environmental Safety 169:273-281.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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-Biomarkers (exposure and effect)-Cell signaling/function-Oxidative stress (including redox biology)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5497487

Domain	Metric		Rating	Comments
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	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: gene expression, enzyme activity, ROS activity

Overall Quality Determination High

Study Citation:	Liu, F., Zaman, W. Q., Peng, H., Li, C., Cao, X., Huang, K., Cui, C., Zhang, W., Lin, K., Luo, Q. (2019). Ecotoxicity of Caenorhabditis elegans following a step and repeated chronic exposure to tetrabromobisphenol A. Ecotoxicology and Environmental Safety 169:273-281.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5497487

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was 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	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for this study type
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of replicates was not reported
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Reporting omissions were unlikely to have a substantial impact on results
	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				

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Study Citation:	Liu, F., Zaman, W. Q., Peng, H., Li, C., Cao, X., Huang, K., Cui, C., Zhang, W., Lin, K., Luo, Q. (2019). Ecotoxicity of Caenorhabditis elegans following a step and repeated chronic exposure to tetrabromobisphenol A. Ecotoxicology and Environmental Safety 169:273-281.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5497487

Domain		Metric	Rating	Comments
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	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 High

Study Citation:	Liu, F., Zaman, W. Q., Peng, H., Li, C., Cao, X., Huang, K., Cui, C., Zhang, W., Lin, K., Luo, Q. (2019). Ecotoxicity of Caenorhabditis elegans following a step and repeated chronic exposure to tetrabromobisphenol A. Ecotoxicology and Environmental Safety 169:273-281.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5497487

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was 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	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for this study type
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of replicates was not reported
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Reporting omissions were unlikely to have a substantial impact on results
	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				

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Study Citation:	Liu, F., Zaman, W. Q., Peng, H., Li, C., Cao, X., Huang, K., Cui, C., Zhang, W., Lin, K., Luo, Q. (2019). Ecotoxicity of Caenorhabditis elegans following a step and repeated chronic exposure to tetrabromobisphenol A. Ecotoxicology and Environmental Safety 169:273-281.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5497487

Domain		Metric	Rating	Comments
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	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 High

Study Citation:	Liu, F., Zaman, W. Q., Peng, H., Li, C., Cao, X., Huang, K., Cui, C., Zhang, W., Lin, K., Luo, Q. (2019). Ecotoxicity of Caenorhabditis elegans following a step and repeated chronic exposure to tetrabromobisphenol A. Ecotoxicology and Environmental Safety 169:273-281.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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*; ctl-2; Larvae
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5497487

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was 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	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	only one concentration tested
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	details of pretreatment were not provided
	Metric 15:	Number of Organisms and Replicates per Group	Low	The numbers of replicates were not reported
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Reporting omissions were unlikely to have a substantial impact on results
	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				

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Study Citation:	Liu, F., Zaman, W. Q., Peng, H., Li, C., Cao, X., Huang, K., Cui, C., Zhang, W., Lin, K., Luo, Q. (2019). Ecotoxicity of Caenorhabditis elegans following a step and repeated chronic exposure to tetrabromobisphenol A. Ecotoxicology and Environmental Safety 169:273-281.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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*; ctl-2; Larvae
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5497487

Domain	Metric		Rating	Comments
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	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 High

Study Citation:	Yang, S. W., Yan, Z. G., Xu, F. F., Wang, S. R., Wu, F. C. (2012). Development of freshwater aquatic life criteria for tetrabromobisphenol A in China. Environmental Pollution 169:59-63.
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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2562857

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%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using appropriate concurrent negative control groups
	Metric 5:	Negative Control Response	High	Mortality was less than 8 percent
	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	The study provided few details on exposure administration
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and 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 above the solubility limit listed in the Final TBBPA Scope (4.15 mg/L) were included, however an ethanol solvent was used and a solvent control was indicated.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed

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Study Citation:	Yang, S. W., Yan, Z. G., Xu, F. F., Wang, S. R., Wu, F. C. (2012). Development of freshwater aquatic life criteria for tetrabromobisphenol A in China. Environmental Pollution 169:59-63.
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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2562857

Domain	Metric		Rating	Comments
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the outcome assessment were not reported.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	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
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: None

Overall Quality Determination Medium

Study Citation:	Ltd,, W.I. (2005). Tetrabromobisphenol-A (TBBPA): A 28-day sediment toxicity test with Chironomus riparius using spiked sediment.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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 riparius*; Larvae
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	7415562

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 an independent laboratory
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.2%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using appropriate concurrent negative control groups
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via sediment
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health

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Study Citation:	Ltd,, W.I. (2005). Tetrabromobisphenol-A (TBBPA): A 28-day sediment toxicity test with Chironomus riparius using spiked sediment.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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 riparius*; Larvae
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	7415562

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

Additional Comments: emergence ratios, development times and development rates

Overall Quality Determination High

Study Citation:	Ltd,, W.I. (2005). Tetrabromobisphenol-A (TBBPA): A 28-day sediment toxicity test with Chironomus riparius using spiked sediment.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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 riparius*; Larvae
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	7415562

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 an independent laboratory
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.2%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using appropriate concurrent negative control groups
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via sediment
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups

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Study Citation:	Ltd,, W.I. (2005). Tetrabromobisphenol-A (TBBPA): A 28-day sediment toxicity test with Chironomus riparius using spiked sediment.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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 riparius*; Larvae
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	7415562

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

Additional Comments: This evaluation is for the assessment of mortality (daily observations).

Overall Quality Determination High

Study Citation:	Ltd,, W.I. (2005). Tetrabromobisphenol-A (TBBPA): A 28-day sediment toxicity test with Chironomus riparius using spiked sediment.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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 riparius*; Larvae
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	7415562

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 an independent laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity was reported as 99.2%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using appropriate concurrent negative control groups.
	Metric 5:	Negative Control Response	High	There were no abnormal behaviors in the negative and solvent control groups.
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for this type of study.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via sediment.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18:	Consistency of Outcome Assessment	High	Observations of any abnormal behavior were made daily.

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Study Citation:	Ltd,, W.I. (2005). Tetrabromobisphenol-A (TBBPA): A 28-day sediment toxicity test with Chironomus riparius using spiked sediment.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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 riparius*; Larvae
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	7415562

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 were no differences in test organisms between treatment groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group in Table 9.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This evaluation is for the assessment of behavior.

Overall Quality Determination High

Study Citation:	Springborn Laboratories, (1989). The subchronic toxicity of sediment-sorbed tetrabromobisphenol A to Chironomus tentans under flow-through conditions (final report) with cover sheet and letter dated 101689.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547095

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	Source was reported as Brominated Flame Retardant Industry Panel. Lot Numbers and manufacturers reported on page 79/100 in PDF.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.15%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies.
	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	Exposures were in sediment and those concentrations and the water were monitored.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				

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Study Citation:	Springborn Laboratories, (1989). The subchronic toxicity of sediment-sorbed tetrabromobisphenol A to Chironomus tentans under flow-through conditions (final report) with cover sheet and letter dated 101689.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547095

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

Additional Comments: Percent survival and wet weight from aqueous testing is on 47/100Percent survival and mean wet weight and dry weight from high organic sediment tests is on page 57/100Percent survival and mean wet weight and dry weight from middle organic sediment tests is on page 58/100Percent survival and mean wet weight and dry weight from low organic sediment tests is on page 59/100Data on tissue concentrations and BDF for TBBPA is on page 60/100

Overall Quality Determination High

Study Citation:	Springborn Laboratories, (1989). The subchronic toxicity of sediment-sorbed tetrabromobisphenol A to Chironomus tentans under flow-through conditions (final report) with cover sheet and letter dated 101689.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547095

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	Source was reported as Brominated Flame Retardant Industry Panel. Lot Numbers and manufacturers reported on page 79/100 in PDF.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.15%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies
	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	Exposures were in sediment and those concentrations and the water were monitored.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest

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Study Citation:	Springborn Laboratories, (1989). The subchronic toxicity of sediment-sorbed tetrabromobisphenol A to Chironomus tentans under flow-through conditions (final report) with cover sheet and letter dated 101689.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547095

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	No information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained

Additional Comments: Percent survival and wet weight from aqueous testing is on 47/100Percent survival and mean wet weight and dry weight from high organic sediment tests is on page 57/100Percent survival and mean wet weight and dry weight from middle organic sediment tests is on page 58/100Percent survival and mean wet weight and dry weight from low organic sediment tests is on page 59/100Data on tissue concentrations and BDF for TBBPA is on page 60/100

Overall Quality Determination High

Study Citation:	Springborn Laboratories, (1989). The subchronic toxicity of sediment-sorbed tetrabromobisphenol A to Chironomus tentans under flow-through conditions (final report) with cover sheet and letter dated 101689.
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; *Chironomus tentans*; Larvae
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547095

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	Source was reported as Brominated Flame Retardant Industry Panel. Lot Numbers and manufacturers reported on page 79/100 in PDF.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.15%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	Medium	The solvent concentration was appropriate. One highest nominal concentration was 5 mg/L, the final scope lists the solubility at 25C at 4.19 mg/L.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health

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Study Citation:	Springborn Laboratories, (1989). The subchronic toxicity of sediment-sorbed tetrabromobisphenol A to Chironomus tentans under flow-through conditions (final report) with cover sheet and letter dated 101689.
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; *Chironomus tentans*; Larvae
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547095

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

Additional Comments: Percent survival and wet weight from aqueous testing is on 47/100Percent survival and mean wet weight and dry weight from high organic sediment tests is on page 57/100Percent survival and mean wet weight and dry weight from middle organic sediment tests is on page 58/100Percent survival and mean wet weight and dry weight from low organic sediment tests is on page 59/100Data on tissue concentrations and BDF for TBBPA is on page 60/100

Overall Quality Determination High

Study Citation:	Springborn Laboratories, (1989). The subchronic toxicity of sediment-sorbed tetrabromobisphenol A to Chironomus tentans under flow-through conditions (final report) with cover sheet and letter dated 101689.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547095

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	Source was reported as Brominated Flame Retardant Industry Panel. Lot Numbers and manufacturers reported on page 79/100 in PDF.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.15%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies
	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	Exposures were in sediment and those concentrations and the water were monitored.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest

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Study Citation:	Springborn Laboratories, (1989). The subchronic toxicity of sediment-sorbed tetrabromobisphenol A to Chironomus tentans under flow-through conditions (final report) with cover sheet and letter dated 101689.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547095

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	No information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained

Additional Comments: Percent survival and wet weight from aqueous testing is on 47/100Percent survival and mean wet weight and dry weight from high organic sediment tests is on page 57/100Percent survival and mean wet weight and dry weight from middle organic sediment tests is on page 58/100Percent survival and mean wet weight and dry weight from low organic sediment tests is on page 59/100Data on tissue concentrations and BDF for TBBPA is on page 60/100

Overall Quality Determination High

Study Citation:	Springborn Laboratories, (1989). The subchronic toxicity of sediment-sorbed tetrabromobisphenol A to Chironomus tentans under flow-through conditions (final report) with cover sheet and letter dated 101689.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547095

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	Source was reported as Brominated Flame Retardant Industry Panel. Lot Numbers and manufacturers reported on page 79/100 in PDF.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.15%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposures were in sediment and those concentrations and the water were monitored.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest

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Study Citation:	Springborn Laboratories, (1989). The subchronic toxicity of sediment-sorbed tetrabromobisphenol A to Chironomus tentans under flow-through conditions (final report) with cover sheet and letter dated 101689.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547095

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	No information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained

Additional Comments: Percent survival and wet weight from aqueous testing is on 47/100Percent survival and mean wet weight and dry weight from high organic sediment tests is on page 57/100Percent survival and mean wet weight and dry weight from middle organic sediment tests is on page 58/100Percent survival and mean wet weight and dry weight from low organic sediment tests is on page 59/100Data on tissue concentrations and BDF for TBBPA is on page 60/100

Overall Quality Determination High

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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	Source was reported as Brominated Flame Retardant Industry Panel. Lot Numbers and manufacturers reported on page 79/100 in PDF.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.15%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposures were in sediment and those concentrations and the water were monitored.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest

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Domain 7: Data Presentation and Analysis				
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	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposures were in sediment and those concentrations and the water were monitored.
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	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
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	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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Taxa, Species, Age:	Invertebrate; Arthropods; *Chironomus tentans*; Larvae
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547095

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Domain 2: Test Design				
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	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	Medium	The solvent concentration was appropriate. One highest nominal concentration was 5 mg/L, the final scope lists the solubility at 25C at 4.19 mg/L
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
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	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: Percent survival and wet weight from aqueous testing is on 47/100Percent survival and mean wet weight and dry weight from high organic sediment tests is on page 57/100Percent survival and mean wet weight and dry weight from middle organic sediment tests is on page 58/100Percent survival and mean wet weight and dry weight from low organic sediment tests is on page 59/100Data on tissue concentrations and BDF for TBBPA is on page 60/100

Overall Quality Determination High

Study Citation:	He, Q., Wang, X., Sun, P., Wang, Z., Wang, L. (2015). Acute and chronic toxicity of tetrabromobisphenol A to three aquatic species under different pH conditions. Aquatic Toxicology 164:145-154.
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:	Immobilization
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3350555

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	High	The test substance identity was not analytically verified by the performing laboratory but source is clearly identified.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	Low	The biological response of the negative control groups was not reported
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The experimental 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 but few details were provided
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods
	Metric 10:	Exposure Duration and Frequency	Medium	Exposure duration was shorter than standard
	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	all pretreatment conditions were the same for control and exposed organisms,
	Metric 15:	Number of Organisms and Replicates per Group	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the amount of details provided in the study being reviewed.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Few details provided so difficult to assess

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Study Citation:	He, Q., Wang, X., Sun, P., Wang, Z., Wang, L. (2015). Acute and chronic toxicity of tetrabromobisphenol A to three aquatic species under different pH conditions. Aquatic Toxicology 164:145-154.
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:	Immobilization
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3350555

Domain	Metric		Rating	Comments
	Metric 17:	Outcome Assessment Methodology	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
	Metric 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	pH was varied across experiments but was consistent across treatments
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups within an exposure
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described
	Metric 22:	Reporting of Data	Medium	only resultant EC50s were reported
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcome

Additional Comments: pH effects and mixture results with DMSO also reported

Overall Quality Determination High

Study Citation:	Lee, S. K., Freitag, D., Steinberg, C., Kettrup, A., Kim, Y. H. (1993). Effects of dissolved humic materials on acute toxicity of some organic chemicals to aquatic organisms. Water Research 27(2):199-204.
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:	Immobilization
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5518890

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Medium	Chemical was identified by name and structure
	Metric 2:	Test Substance Source	High	Sourced from "Kukdo Co, Korea". 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. "Technical grade" with no further details.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	use of appropriate controls was implied by Figure 1, but not stated outright.
	Metric 5:	Negative Control Response	Low	The biological response of the negative control groups was not reported.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The study provided few details on exposure media preparation
	Metric 8:	Consistency of Exposure Administration	Medium	Reporting omissions were unlikely to have a substantial impact on results
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	exposure levels are not specified, only a range implied in Figure 1
	Metric 12:	Testing at or Below Solubility Limit	Low	Reporting omissions prevented determination of whether exposure concentrations exceeded the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test animals was not reported
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				

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Study Citation:	Lee, S. K., Freitag, D., Steinberg, C., Kettrup, A., Kim, Y. H. (1993). Effects of dissolved humic materials on acute toxicity of some organic chemicals to aquatic organisms. Water Research 27(2):199-204.
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:	Immobilization
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5518890

Domain	Metric		Rating	Comments
	Metric 16:	Adequacy of Test Conditions	Medium	Reported environmental conditions were not sufficient to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were not reported
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	One series with no added humic acid was used
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among group
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical analysis was performed and described adequately.
	Metric 22:	Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	Medium	There were no unexpected outcomes but few details were provided

Additional Comments: None

Overall Quality Determination Low

Study Citation:	Ltd,, W.I. (2003). Tetrabromobisphenol A: A 48-hour flow-through acute toxicity test with the cladoceran (Daphnia magna).
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:	Immobilization
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5352358

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 was obtained from three manufacturers(Great Lakes Chemical Corporation iot# I008JE04B), AlbermarleCorporation (lot# 25115T-1), and Bromine Compounds, Ltd. (lot 010040)) and and a composite sample was prepared. Subsamples of the composite was analytically verified by a third party.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.17%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using appropriate concurrent negative control groups. A negative control (dilution water), and a solvent control (0.1 mL dimethyl formamide/L) were used.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and adequate for assessed outcomes.
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were indiscriminately allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies.
	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	Medium	The number of exposure groups and spacing of exposure levels were justified for a dose response as authors were only looking to see if toxic effects occurred above 1 mg/L, however no toxic effects were reported at the highest concentration. Only two exposure groups were tested.
	Metric 12:	Testing at or Below Solubility Limit	High	The DMF solvent concentration was appropriate.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.

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Study Citation:	Ltd,, W.I. (2003). Tetrabromobisphenol A: A 48-hour flow-through acute toxicity test with the cladoceran (Daphnia magna).
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:	Immobilization
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5352358

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Low	Only two replicates were used.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition.
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 immobilization outcome assess in Daphnia magna following exposure to two concentrations of TBBPA. Experiment was well described and exposure concentrations were measured during the study.

Overall Quality Determination High

Study Citation:	Union Carbide Corporation Environmental Services, (1978). Acute toxicity of FMBP4A (Tetrabromobisphenol A) to the water flea Daphnia magna straus.
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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5546496

Domain		Metric	Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using appropriate concurrent negative control groups.
	Metric 5:	Negative Control Response	High	The biological 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, concentration of acetone used was not reported. Concentrations of test substance were not measured during the study.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	Standard test duration (48 hr) was used for the daphnid test.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response. 5 exposure concentrations (0.32, 0.56, 1.0. 1.8 and 3.20 mg/l) of TBBPA were used.
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate ((i.e., no effects on biological responses were observed in the solvent control and no interactions were expected between the solvent and test substance).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a laboratory stock culture. Daphnids less than 20 hours old were used for the experiment.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The number of test organisms and/or replicates was suitable. 4 replicate beakers with 5 daphnids each were used.
Domain 5: Outcome Assessment				

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Study Citation:	Union Carbide Corporation Environmental Services, (1978). Acute toxicity of FMBP4A (Tetrabromobisphenol A) to the water flea Daphnia magna straus.
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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5546496

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	Medium	The outcome assessment methodology reported the intended outcome of interest, no mention of how they assessed mortalities.
	Metric 18:	Consistency of Outcome Assessment	Medium	Outcomes were assessed consistently across study groups but only few details were provided.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure (e.g., infection) that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22:	Reporting of Data	Medium	Data for exposure-related findings were presented for each treatment and control group. Not specified whether the data was mean mortality data from the four replicates and no measure of variability was provided.
	Metric 23:	Explanation of Unexpected Outcomes	Low	insufficient information was provided to determine if excessive variability or unexpected outcomes occurred.

Additional Comments: None

Overall Quality Determination Medium

Study Citation:	Waaijers, S. L., Hartmann, J., Soeter, A. M., Helmus, R., Kools, S. A., Voogt, de, P., Admiraal, W., Parsons, J. R., Kraak, M. H. (2013). Toxicity of new generation flame retardants to Daphnia magna. Science of the Total Environment 463-464:1042-1048.
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:	Immobilization
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1786503

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 97%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrentsolvent control group
	Metric 5:	Negative Control Response	Low	The biological response of the negative control groups was not reported
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study group
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	Medium	Few details of exposure administration were reported but administration seemed adequate
	Metric 9:	Measurement of Test Substance Concentration	Low	Concentrations were nominal
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	exposure groups were identified graphically
	Metric 12:	Testing at or Below Solubility Limit	Medium	Concentrations may have exceeded solubility but solvents at an appropriate level aided in dissolution
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health

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Study Citation:	Waaijers, S. L., Hartmann, J., Soeter, A. M., Helmus, R., Kools, S. A., Voogt, de, P., Admiraal, W., Parsons, J. R., Kraak, M. H. (2013). Toxicity of new generation flame retardants to Daphnia magna. Science of the Total Environment 463-464:1042-1048.
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:	Immobilization
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1786503

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

Additional Comments: None

Overall Quality Determination High

Study Citation:	Yang, S. W., Yan, Z. G., Xu, F. F., Wang, S. R., Wu, F. C. (2012). Development of freshwater aquatic life criteria for tetrabromobisphenol A in China. Environmental Pollution 169:59-63.
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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2562857

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%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using appropriate concurrent negative control groups
	Metric 5:	Negative Control Response	High	Mortality was less than 8 percent
	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	The study provided few details on exposure administration
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and 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 above the solubility limit listed in the Final TBBPA Scope (4.15 mg/L) were included, however an ethanol solvent was used and a solvent control was indicated.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed

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Study Citation:	Yang, S. W., Yan, Z. G., Xu, F. F., Wang, S. R., Wu, F. C. (2012). Development of freshwater aquatic life criteria for tetrabromobisphenol A in China. Environmental Pollution 169:59-63.
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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2562857

Domain	Metric		Rating	Comments
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the outcome assessment were not reported.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	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
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: None

Overall Quality Determination Medium

Study Citation:	Springborn Laboratories, (1989). The chronic toxicity of tetrabromobisphenol A to Daphnia magna under flow-through conditions (final report) with cover letter dated 081989.
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:	Immobilization
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547090

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	High	The test substance identity was analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 100% AI
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using appropriate concurrent negative control groups
	Metric 5:	Negative Control Response	Medium	Specific immobilization data for the control group was not provided. However, the mean percentage survival at day 21 was 98%. In the text, authors mentioned that no immobilization occurred at any of the tested treatment levels.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	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	Medium	The number of exposure groups and spacing of exposure levels were too low 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	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				

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Study Citation:	Springborn Laboratories, (1989). The chronic toxicity of tetrabromobisphenol A to Daphnia magna under flow-through conditions (final report) with cover letter dated 081989.
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:	Immobilization
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547090

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	Medium	The immobilization assessment was briefly described.
	Metric 18:	Consistency of Outcome Assessment	High	It was assumed 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 were no reported differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described.
	Metric 22:	Reporting of Data	Medium	No raw data was shown. But authors mentioned there were no observed immobilized organisms at any of the tested treatment levels (0.98-0.056 mg/L).
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This evaluation is for the assessment of immobilization.

Overall Quality Determination High

Study Citation:	Springborn Laboratories, (1989). The chronic toxicity of tetrabromobisphenol A to Daphnia magna under flow-through conditions (final report) with cover letter dated 081989.
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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547090

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only.
	Metric 2:	Test Substance Source	High	The test substance identity was analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 100% AI.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using appropriate concurrent negative control groups.
	Metric 5:	Negative Control Response	High	The biological 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	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	The number of exposure groups and spacing of exposure levels were suitable 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	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups.

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Study Citation:	Springborn Laboratories, (1989). The chronic toxicity of tetrabromobisphenol A to Daphnia magna under flow-through conditions (final report) with cover letter dated 081989.
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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547090

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

Additional Comments: Offspring produced are presented in Table 6 on page 36/60.

Overall Quality Determination High

Study Citation:	Springborn Laboratories, (1989). The chronic toxicity of tetrabromobisphenol A to Daphnia magna under flow-through conditions (final report) with cover letter dated 081989.
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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547090

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only.
	Metric 2:	Test Substance Source	High	The test substance identity was analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 100% AI.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using appropriate concurrent negative control groups.
	Metric 5:	Negative Control Response	High	The biological 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	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	The number of exposure groups and spacing of exposure levels were too low 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	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.

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Study Citation:	Springborn Laboratories, (1989). The chronic toxicity of tetrabromobisphenol A to Daphnia magna under flow-through conditions (final report) with cover letter dated 081989.
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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547090

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

Additional Comments: Growth metrics at 21 days are presented for each treatment concentration and control on page 37/60, table 7.

Overall Quality Determination High

Study Citation:	Springborn Laboratories, (1989). The chronic toxicity of tetrabromobisphenol A to Daphnia magna under flow-through conditions (final report) with cover letter dated 081989.
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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547090

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	High	The test substance identity was analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 100% AI
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using appropriate concurrent negative control groups
	Metric 5:	Negative Control Response	High	The biological 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	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	The number of exposure groups and spacing of exposure levels were too low 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	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.

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Study Citation:	Springborn Laboratories, (1989). The chronic toxicity of tetrabromobisphenol A to Daphnia magna under flow-through conditions (final report) with cover letter dated 081989.
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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547090

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

Additional Comments: Mortality/survival was reported in table 5 on page 35/60.

Overall Quality Determination High

Study Citation:	Yang, S. W., Yan, Z. G., Xu, F. F., Wang, S. R., Wu, F. C. (2012). Development of freshwater aquatic life criteria for tetrabromobisphenol A in China. Environmental Pollution 169:59-63.
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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2562857

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%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using appropriate concurrent negative control groups
	Metric 5:	Negative Control Response	High	Mortality was less than 8 percent
	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	The study provided few details on exposure administration
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and 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 above the solubility limit listed in the Final TBBPA Scope (4.15 mg/L) were included, however an ethanol solvent was used and a solvent control was indicated.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed

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Study Citation:	Yang, S. W., Yan, Z. G., Xu, F. F., Wang, S. R., Wu, F. C. (2012). Development of freshwater aquatic life criteria for tetrabromobisphenol A in China. Environmental Pollution 169:59-63.
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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2562857

Domain	Metric		Rating	Comments
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the outcome assessment were not reported.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	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
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: None

Overall Quality Determination Medium

Study Citation:	Yang, S. W., Yan, Z. G., Xu, F. F., Wang, S. R., Wu, F. C. (2012). Development of freshwater aquatic life criteria for tetrabromobisphenol A in China. Environmental Pollution 169:59-63.
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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2562857

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%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using appropriate concurrent negative control groups
	Metric 5:	Negative Control Response	High	Mortality was less than 8 percent
	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	The study provided few details on exposure administration
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and 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 above the solubility limit listed in the Final TBBPA Scope (4.15 mg/L) were included, however an ethanol solvent was used and a solvent control was indicated.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported

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Study Citation:	Yang, S. W., Yan, Z. G., Xu, F. F., Wang, S. R., Wu, F. C. (2012). Development of freshwater aquatic life criteria for tetrabromobisphenol A in China. Environmental Pollution 169:59-63.
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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2562857

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the outcome assessment were not reported.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	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
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: None

Overall Quality Determination Medium

Study Citation:	Zhang, S., Hagstrom, D., Hayes, P., Graham, A., Collins, E. S. (2019). Multi-behavioral endpoint testing of an 87-chemical compound library in freshwater planarians. Toxicological Sciences 167(1):26-44.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Dugesia japonica*; regenerating; Adult
Health Outcome:	Ocular and Sensory
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064285

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity or grade of test substance was not reported
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The control response for eye regeneration was shown in Figure 2H and seemed normal.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The study provided few details on exposure media preparation
	Metric 8:	Consistency of Exposure Administration	Medium	Reporting omissions were unlikely to have a substantial impact on results
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure 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 (0.5% DMSO was appropriate). The study authors had "previously shown that there are no effects onplanarian morphology or behavior at this concentration."
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects, tests were repeated three times
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:	Zhang, S., Hagstrom, D., Hayes, P., Graham, A., Collins, E. S. (2019). Multi-behavioral endpoint testing of an 87-chemical compound library in freshwater planarians. Toxicological Sciences 167(1):26-44.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Dugesia japonica*; regenerating; Adult
Health Outcome:	Ocular and Sensory
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064285

Domain	Metric		Rating	Comments
	Metric 17:	Outcome Assessment Methodology	Low	The assessment methodology reported the intended outcome of interest with uncertainty
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes
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 evaluation is for the eye regeneration test. At the highest test concentration for TBBPA in the eye regeneration test it appeared as though the organisms died for each replicate.

Overall Quality Determination Medium

Study Citation:	Zhang, S., Ireland, D., Sipes, N. S., Behl, M., Collins, E. S. (2019). Screening for neurotoxic potential of 15 flame retardants using freshwater planarians. Neurotoxicology and Teratology 73:54-66.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Dugesia japonica*; regenerative; Adult
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5469417

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Test substance clearly identified with name and CAS #
	Metric 2:	Test Substance Source	Low	Sourced from Sigma Aldrich, but chemical was not analytically verified by the laboratory.
	Metric 3:	Test Substance Purity	High	reported as 99.77% pure
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Authors did not use a true negative control. ”Negative” controls contained either acetaminophen or L-ascorbic acid. A positive solvent control (0.5% DMSO) was also used. The authors presented information from previous studies to verify that acetaminophen or L-ascorbic acid did not affect the test organisms.
	Metric 5:	Negative Control Response	High	100% survival in solvent control (0.5% DMSO), no mortality observed in ”negative” controls
	Metric 6:	Randomized Allocation	Low	No mention of randomization in this paper. Author did cite another paper which may have described randomization.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The experimental system and methods for preparation of test media were described in adequate detail and appropriately accounted for the physical-chemical properties of the test substance (solvent used due to the low solubility of the test material). Measured test concentrations were not provided.
	Metric 8:	Consistency of Exposure Administration	High	No inconsistencies were noticed or reported.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured, highest concentration listed as 99.5 uM in supplemental data instead of a nominal value of 100 uM as stated in text.
	Metric 10:	Exposure Duration and Frequency	High	Exposure duration (12 days) was adequate to elicit a response for the chemicals in this study.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Exposure concentrations seemed adequate to illicit a response (development) for most chemicals tested in this study.
	Metric 12:	Testing at or Below Solubility Limit	Medium	Solvent (0.5% DMSO) was used to facilitate the dissolution of the test material into the test medium. The two highest concentrations (100 and 10 uM) are above the solubility listed in the final scope (4.1 mg/L).
Domain 4: Test Organism				

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Study Citation:	Zhang, S., Ireland, D., Sipes, N. S., Behl, M., Collins, E. S. (2019). Screening for neurotoxic potential of 15 flame retardants using freshwater planarians. Neurotoxicology and Teratology 73:54-66.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Dugesia japonica*; regenerative; Adult
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5469417

Domain	Metric		Rating	Comments
	Metric 13:	Test Organism Characteristics	High	Supplemental methods description describes the test organism in more detail (Zhang et al., 2019). Test organisms were described as adults and "Test worms were manually selected to fall within a certain range of sizes and we found full worm length, after automated size measurement, to be 7.3 6 2.3 mm (mean 6 SD), and tail worm length to be 7.3 mm þ/- 2.7 mm (mean 6 SD).
	Metric 14:	Acclimatization and Pretreatment Conditions	High	In supplemental materials, pretreatment procedure was described as: "Animals were starved for at least 5 days before beingused for experiments and their containers were cleaned immediately prior to worm selection for experiments." This was adequate for gut-clearance.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	8 individuals were used for each replicate.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Test conditions were fully described in supplemental methods paper (Zhang et al., 2019): "Wells of a 48- well plate with 200 ml of IO water." The authors noted that in the present study, they updated the feeding procedure, but this was not described clearly, "we fed planarians used in FR screen 2 commercial freeze-dried organic chicken liver (Amazon, Seattle, WA) to better control food quality and thus minimize animal fitness variability." It was unclear when this supplemental feeding occurred during the exposure period or if the lack of feeding could have contributed to additional mortality in some chemicals in the screen.
	Metric 17:	Outcome Assessment Methodology	High	Outcome assessment was adequate.
	Metric 18:	Consistency of Outcome Assessment	High	Assessment methods seemed consistent across groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. Besides the feeding procedure, there were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure (e.g., infection) that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				

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Study Citation:	Zhang, S., Ireland, D., Sipes, N. S., Behl, M., Collins, E. S. (2019). Screening for neurotoxic potential of 15 flame retardants using freshwater planarians. Neurotoxicology and Teratology 73:54-66.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Dugesia japonica*; regenerative; Adult
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5469417

Domain	Metric		Rating	Comments
	Metric 21:	Statistical Methods	Low	The authors describe the statistical procedure as: "Statistical significance was determined using either a one-tailed Fisher's exact test for lethality, eye regeneration, phototaxis, and scrunching endpoints; Mann Whitney U test for thermotaxis and unstimulated behavior; or two-tailed t-test for unstimulated behavior (depending on normality of the sample) using a significance level of 0.05. This was adequate to determine if mortality was significant." This analysis and the raw data describing mortality were not presented in this study.
	Metric 22:	Reporting of Data	Low	A large amount of data was presented in the supplemental file (Zhang et al., 2019): https://doi.org/10.1016/j.ntt.2019.03.003. Data were not clear and interpretation was difficult.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes reported.

Additional Comments: This form is for eye regeneration for day 7 and detailed in Figure 1. Results are presented in supplement 1 xsl file.

Overall Quality Determination Medium

Study Citation:	Zhang, S., Ireland, D., Sipes, N. S., Behl, M., Collins, E. S. (2019). Screening for neurotoxic potential of 15 flame retardants using freshwater planarians. Neurotoxicology and Teratology 73:54-66.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Dugesia japonica*; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5469417

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Test substance clearly identified with name and CAS #
	Metric 2:	Test Substance Source	Low	Sourced from Sigma Aldrich, but chemical was not analytically verified by the laboratory.
	Metric 3:	Test Substance Purity	High	reported as 99.77% pure.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Authors did not use a true negative control. "Negative" controls contained either acetaminophen or L-ascorbic acid. A positive solvent control (0.5% DMSO) was also used. The authors presented information from previous studies to verify that acetaminophen or L-ascorbic acid did not affect the test organisms.
	Metric 5:	Negative Control Response	High	100% survival in solvent control (0.5% DMSO), no mortality observed in "negative" controls.
	Metric 6:	Randomized Allocation	Low	No mention of randomization in this paper. Author did cite another paper which may have described randomization.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The experimental system and methods for preparation of test media were described in adequate detail and appropriately accounted for the physical-chemical properties of the test substance (solvent used due to the low solubility of the test material). Measured test concentrations were not provided.
	Metric 8:	Consistency of Exposure Administration	High	No inconsistencies were noticed or reported
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured, highest concentration listed as 99.5 uM in supplemental data instead of a nominal value of 100 uM as stated in text
	Metric 10:	Exposure Duration and Frequency	High	Exposure duration (12 days) was adequate to cause mortality for some of the chemicals in this study (mortalities seen in TBBPA, but not TCEP).
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Exposure concentrations seemed adequate to illicit a response (mortality) for most chemicals tested in this study, TBBPA included. This was below the NOAEL for TCEP, but the purpose of the study was to compare the mortality between chemicals within a specific group, so this was not determined to affect the rating of this study.
	Metric 12:	Testing at or Below Solubility Limit	Medium	Solvent (0.5% DMSO) was used to facilitate the dissolution of the test material into the test medium. The two highest concentrations (100 and 10 uM) are above the solubility listed in the final scope (4.1 mg/L).
Domain 4: Test Organism				

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Study Citation:	Zhang, S., Ireland, D., Sipes, N. S., Behl, M., Collins, E. S. (2019). Screening for neurotoxic potential of 15 flame retardants using freshwater planarians. Neurotoxicology and Teratology 73:54-66.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Dugesia japonica*; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5469417

Domain	Metric		Rating	Comments
	Metric 13:	Test Organism Characteristics	High	Supplemental methods description describes the test organism in more detail (Zhang et al., 2019). Test organisms were described as adults and "Test worms were manually selected to fall within a certain range of sizes and we found full worm length, after automated size measurement, to be 7.3 6 2.3 mm (mean 6 SD), and tail worm length to be 7.3 mm þ/- 2.7 mm (mean 6 SD).
	Metric 14:	Acclimatization and Pretreatment Conditions	High	In supplemental materials, pretreatment procedure was described as: "Animals were starved for at least 5 days before beingused for experiments and their containers were cleaned immediately prior to worm selection for experiments." This was adequate for gut-clearance.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	8 individuals were used for each replicate.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Test conditions were fully described in supplemental methods paper (Zhang et al., 2019): "Wells of a 48- well plate with 200 ml of IO water." The authors noted that in the present study, they updated the feeding procedure, but this was not described clearly, "we fed planarians used in FR screen 2 commercial freeze-dried organic chicken liver (Amazon, Seattle, WA) to better control food quality and thus minimize animal fitness variability." It was unclear when this supplemental feeding occurred during the exposure period or if the lack of feeding could have contributed to additional mortality in some chemicals in the screen.
	Metric 17:	Outcome Assessment Methodology	High	Outcome assessment was adequate to describe the comparative mortality of several flame retardants following exposure to the test material.
	Metric 18:	Consistency of Outcome Assessment	High	Assessment methods seemed consistent across groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. Besides the feeding procedure, there were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure (e.g., infection) that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				

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Study Citation:	Zhang, S., Ireland, D., Sipes, N. S., Behl, M., Collins, E. S. (2019). Screening for neurotoxic potential of 15 flame retardants using freshwater planarians. Neurotoxicology and Teratology 73:54-66.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Dugesia japonica*; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5469417

Domain	Metric		Rating	Comments
	Metric 21:	Statistical Methods	Low	The authors describe the statistical procedure as: "Statistical significance was determined using either a one-tailed Fisher's exact test for lethality, eye regeneration, phototaxis, and scrunching endpoints; Mann Whitney U test for thermotaxis and unstimulated behavior; or two-tailed t-test for unstimulated behavior (depending on normality of the sample) using a significance level of 0.05. This was adequate to determine if mortality was significant." This analysis and the raw data describing mortality were not presented in this study.
	Metric 22:	Reporting of Data	Low	A large amount of data was presented in the supplemental file (Zhang et al., 2019): https://doi.org/10.1016/j.ntt.2019.03.003. Data were not clear and interpretation was difficult.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes reported.

Additional Comments: This form is for the mortality test for the adult organisms conducted on day 7. Mortality data is located in supplemental 1 xls.

Overall Quality Determination Medium

Study Citation:	Zhang, S., Ireland, D., Sipes, N. S., Behl, M., Collins, E. S. (2019). Screening for neurotoxic potential of 15 flame retardants using freshwater planarians. Neurotoxicology and Teratology 73:54-66.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Dugesia japonica*; regenerative; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5469417

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Test substance clearly identified with name and CAS #
	Metric 2:	Test Substance Source	Low	Sourced from Sigma Aldrich, but chemical was not analytically verified by the laboratory.
	Metric 3:	Test Substance Purity	High	reported as 99.77% pure
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Authors did not use a true negative control. "Negative" controls contained either acetaminophen or L-ascorbic acid. A positive solvent control (0.5% DMSO) was also used. The authors presented information from previous studies to verify that acetaminophen or L-ascorbic acid did not affect the test organisms.
	Metric 5:	Negative Control Response	High	100% survival in solvent control (0.5% DMSO), no mortality observed in "negative" controls
	Metric 6:	Randomized Allocation	Low	No mention of randomization in this paper. Author did cite another paper which may have described randomization.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The experimental system and methods for preparation of test media were described in adequate detail and appropriately accounted for the physical-chemical properties of the test substance (solvent used due to the low solubility of the test material). Measured test concentrations were not provided.
	Metric 8:	Consistency of Exposure Administration	High	No inconsistencies were noticed or reported
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured, highest concentration listed as 99.5 uM in supplemental data instead of a nominal value of 100 uM as stated in text
	Metric 10:	Exposure Duration and Frequency	High	Exposure duration (12 days) was adequate to cause mortality for some of the chemicals in this study (mortalities seen in TBBPA, but not TCEP).
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Exposure concentrations seemed adequate to illicit a response (mortality) for most chemicals tested in this study, TBBPA included. This was below the NOAEL for TCEP, but the purpose of the study was to compare the mortality between chemicals within a specific group, so this was not determined to affect the rating of this study.
	Metric 12:	Testing at or Below Solubility Limit	Medium	Solvent (0.5% DMSO) was used to facilitate the dissolution of the test material into the test medium. The two highest concentrations (100 and 10 uM) are above the solubility listed in the final scope (4.1 mg/L).
Domain 4: Test Organism				

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Study Citation:	Zhang, S., Ireland, D., Sipes, N. S., Behl, M., Collins, E. S. (2019). Screening for neurotoxic potential of 15 flame retardants using freshwater planarians. Neurotoxicology and Teratology 73:54-66.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Dugesia japonica*; regenerative; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5469417

Domain	Metric		Rating	Comments
	Metric 13:	Test Organism Characteristics	High	Supplemental methods description describes the test organism in more detail (Zhang et al., 2019). Test organisms were described as adults and "Test worms were manually selected to fall within a certain range of sizes and we found full worm length, after automated size measurement, to be 7.3 6 2.3 mm (mean 6 SD), and tail worm length to be 7.3 mm þ/- 2.7 mm (mean 6 SD).
	Metric 14:	Acclimatization and Pretreatment Conditions	High	In supplemental materials, pretreatment procedure was described as: "Animals were starved for at least 5 days before beingused for experiments and their containers were cleaned immediately prior to worm selection for experiments." This was adequate for gut-clearance.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	8 individuals were used for each replicate.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Test conditions were fully described in supplemental methods paper (Zhang et al., 2019): "Wells of a 48- well plate with 200 ml of IO water." The authors noted that in the present study, they updated the feeding procedure, but this was not described clearly, "we fed planarians used in FR screen 2 commercial freeze-dried organic chicken liver (Amazon, Seattle, WA) to better control food quality and thus minimize animal fitness variability." It was unclear when this supplemental feeding occurred during the exposure period or if the lack of feeding could have contributed to additional mortality in some chemicals in the screen.
	Metric 17:	Outcome Assessment Methodology	High	Outcome assessment was adequate to describe the comparative mortality of several flame retardants following exposure to the test material.
	Metric 18:	Consistency of Outcome Assessment	High	Assessment methods seemed consistent across groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. Besides the feeding procedure, there were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure (e.g., infection) that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				

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Study Citation:	Zhang, S., Ireland, D., Sipes, N. S., Behl, M., Collins, E. S. (2019). Screening for neurotoxic potential of 15 flame retardants using freshwater planarians. Neurotoxicology and Teratology 73:54-66.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Dugesia japonica*; regenerative; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5469417

Domain	Metric		Rating	Comments
	Metric 21:	Statistical Methods	Low	The authors describe the statistical procedure as: "Statistical significance was determined using either a one-tailed Fisher's exact test for lethality, eye regeneration, phototaxis, and scrunching endpoints; Mann Whitney U test for thermotaxis and unstimulated behavior; or two-tailed t-test for unstimulated behavior (depending on normality of the sample) using a significance level of 0.05. This was adequate to determine if mortality was significant." This analysis and the raw data describing mortality were not presented in this study.
	Metric 22:	Reporting of Data	Low	A large amount of data was presented in the supplemental file (Zhang et al., 2019): https://doi.org/10.1016/j.ntt.2019.03.003. Data were not clear and interpretation was difficult.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes reported.

Additional Comments: This form is for the mortality test for the regenerating adult organisms at dat 7. The data for mortality are located in Supplement 1 xls.

Overall Quality Determination Medium

Study Citation:	Zhang, S., Ireland, D., Sipes, N. S., Behl, M., Collins, E. S. (2019). Screening for neurotoxic potential of 15 flame retardants using freshwater planarians. Neurotoxicology and Teratology 73:54-66.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Dugesia japonica*; Adult
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5469417

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Test substance clearly identified with name and CAS #
	Metric 2:	Test Substance Source	Low	Sourced from Sigma Aldrich, but chemical was not analytically verified by the laboratory.
	Metric 3:	Test Substance Purity	High	reported as 99.77% pure
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Authors did not use a true negative control. ”Negative” controls contained either acetaminophen or L-ascorbic acid. A positive solvent control (0.5% DMSO) was also used. The authors presented information from previous studies to verify that acetaminophen or L-ascorbic acid did not affect the test organisms.
	Metric 5:	Negative Control Response	High	No effects observed in ”negative” or solvent controls
	Metric 6:	Randomized Allocation	Low	No mention of randomization in this paper. Author did cite another paper which may have described randomization.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The experimental system and methods for preparation of test media were described in adequate detail and appropriately accounted for the physical-chemical properties of the test substance (solvent used due to the low solubility of the test material). Measured test concentrations were not provided.
	Metric 8:	Consistency of Exposure Administration	High	No inconsistencies were noticed or reported
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured, highest concentration listed as 99.5 uM in supplemental data instead of a nominal value of 100 uM as stated in text
	Metric 10:	Exposure Duration and Frequency	High	Exposure duration (12 days) was adequate to elicit a response for some of the chemicals in this study.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Exposure concentrations seemed adequate to elicit a response (behavior) for most chemicals tested in this study.
	Metric 12:	Testing at or Below Solubility Limit	Medium	Solvent (0.5% DMSO) was used to facilitate the dissolution of the test material into the test medium. The two highest concentrations (100 and 10 uM) are above the solubility listed in the final scope (4.1 mg/L).
Domain 4: Test Organism				

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Study Citation:	Zhang, S., Ireland, D., Sipes, N. S., Behl, M., Collins, E. S. (2019). Screening for neurotoxic potential of 15 flame retardants using freshwater planarians. Neurotoxicology and Teratology 73:54-66.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Dugesia japonica*; Adult
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5469417

Domain	Metric		Rating	Comments
	Metric 13:	Test Organism Characteristics	High	Supplemental methods description describes the test organism in more detail (Zhang et al., 2019). Test organisms were described as adults and "Test worms were manually selected to fall within a certain range of sizes and we found full worm length, after automated size measurement, to be 7.3 6 2.3 mm (mean 6 SD), and tail worm length to be 7.3 mm þ/- 2.7 mm (mean 6 SD).
	Metric 14:	Acclimatization and Pretreatment Conditions	High	In supplemental materials, pretreatment procedure was described as: "Animals were starved for at least 5 days before beingused for experiments and their containers were cleaned immediately prior to worm selection for experiments." This was adequate for gut-clearance.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	8 individuals were used for each replicate.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Test conditions were fully described in supplemental methods paper (Zhang et al., 2019): "Wells of a 48- well plate with 200 ml of IO water." The authors noted that in the present study, they updated the feeding procedure, but this was not described clearly, "we fed planarians used in FR screen 2 commercial freeze-dried organic chicken liver (Amazon, Seattle, WA) to better control food quality and thus minimize animal fitness variability." It was unclear when this supplemental feeding occurred during the exposure period or if the lack of feeding could have contributed to additional mortality in some chemicals in the screen.
	Metric 17:	Outcome Assessment Methodology	High	Outcome assessment was adequate
	Metric 18:	Consistency of Outcome Assessment	High	Assessment methods seemed consistent across groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. Besides the feeding procedure, there were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure (e.g., infection) that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				

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Study Citation:	Zhang, S., Ireland, D., Sipes, N. S., Behl, M., Collins, E. S. (2019). Screening for neurotoxic potential of 15 flame retardants using freshwater planarians. Neurotoxicology and Teratology 73:54-66.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Dugesia japonica*; Adult
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5469417

Domain	Metric		Rating	Comments
	Metric 21:	Statistical Methods	Low	The authors describe the statistical procedure as: "Statistical significance was determined using either a one-tailed Fisher's exact test for lethality, eye regeneration, phototaxis, and scrunching endpoints; Mann Whitney U test for thermotaxis and unstimulated behavior; or two-tailed t-test for unstimulated behavior (depending on normality of the sample) using a significance level of 0.05. This was adequate to determine if mortality was significant." This analysis and the raw data describing mortality were not presented in this study.
	Metric 22:	Reporting of Data	Low	A large amount of data was presented in the supplemental file (Zhang et al., 2019): https://doi.org/10.1016/j.ntt.2019.03.003. Data were not clear and interpretation was difficult.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes reported.

Additional Comments: This form is for behavior at day 7 for adults. The data are located in Supplement 1 xls. Behavior data includes assays for: Speed, rest, and phototaxsis.

Overall Quality Determination Medium

Study Citation:	Zhang, S., Ireland, D., Sipes, N. S., Behl, M., Collins, E. S. (2019). Screening for neurotoxic potential of 15 flame retardants using freshwater planarians. Neurotoxicology and Teratology 73:54-66.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Dugesia japonica*; regenerative; Adult
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5469417

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Test substance clearly identified with name and CAS #
	Metric 2:	Test Substance Source	Low	Sourced from Sigma Aldrich, but chemical was not analytically verified by the laboratory.
	Metric 3:	Test Substance Purity	High	reported as 99.77% pure
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Authors did not use a true negative control. ”Negative” controls contained either acetaminophen or L-ascorbic acid. A positive solvent control (0.5% DMSO) was also used. The authors presented information from previous studies to verify that acetaminophen or L-ascorbic acid did not affect the test organisms.
	Metric 5:	Negative Control Response	High	No effects observed in ”negative” or solvent controls
	Metric 6:	Randomized Allocation	Low	No mention of randomization in this paper. Author did cite another paper which may have described randomization.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The experimental system and methods for preparation of test media were described in adequate detail and appropriately accounted for the physical-chemical properties of the test substance (solvent used due to the low solubility of the test material). Measured test concentrations were not provided.
	Metric 8:	Consistency of Exposure Administration	High	No inconsistencies were noticed or reported
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured, highest concentration listed as 99.5 uM in supplemental data instead of a nominal value of 100 uM as stated in text
	Metric 10:	Exposure Duration and Frequency	High	Exposure duration (12 days) was adequate to elicit a response for some of the chemicals in this study.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Exposure concentrations seemed adequate to elicit a response (behavior) for most chemicals tested in this study.
	Metric 12:	Testing at or Below Solubility Limit	Medium	Solvent (0.5% DMSO) was used to facilitate the dissolution of the test material into the test medium. The two highest concentrations (100 and 10 uM) are above the solubility listed in the final scope (4.1 mg/L).
Domain 4: Test Organism				

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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)
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Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
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Domain	Metric		Rating	Comments
	Metric 13:	Test Organism Characteristics	High	Supplemental methods description describes the test organism in more detail (Zhang et al., 2019). Test organisms were described as adults and "Test worms were manually selected to fall within a certain range of sizes and we found full worm length, after automated size measurement, to be 7.3 6 2.3 mm (mean 6 SD), and tail worm length to be 7.3 mm þ/- 2.7 mm (mean 6 SD).
	Metric 14:	Acclimatization and Pretreatment Conditions	High	In supplemental materials, pretreatment procedure was described as: "Animals were starved for at least 5 days before beingused for experiments and their containers were cleaned immediately prior to worm selection for experiments." This was adequate for gut-clearance.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	8 individuals were used for each replicate.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Test conditions were fully described in supplemental methods paper (Zhang et al., 2019): "Wells of a 48- well plate with 200 ml of IO water." The authors noted that in the present study, they updated the feeding procedure, but this was not described clearly, "we fed planarians used in FR screen 2 commercial freeze-dried organic chicken liver (Amazon, Seattle, WA) to better control food quality and thus minimize animal fitness variability." It was unclear when this supplemental feeding occurred during the exposure period or if the lack of feeding could have contributed to additional mortality in some chemicals in the screen.
	Metric 17:	Outcome Assessment Methodology	High	Outcome assessment was adequate
	Metric 18:	Consistency of Outcome Assessment	High	Assessment methods seemed consistent across groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. Besides the feeding procedure, there were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure (e.g., infection) that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				

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Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
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Domain	Metric		Rating	Comments
	Metric 21:	Statistical Methods	Low	The authors describe the statistical procedure as: "Statistical significance was determined using either a one-tailed Fisher's exact test for lethality, eye regeneration, phototaxis, and scrunching endpoints; Mann Whitney U test for thermotaxis and unstimulated behavior; or two-tailed t-test for unstimulated behavior (depending on normality of the sample) using a significance level of 0.05. This was adequate to determine if mortality was significant." This analysis and the raw data describing mortality were not presented in this study.
	Metric 22:	Reporting of Data	Low	A large amount of data was presented in the supplemental file (Zhang et al., 2019): https://doi.org/10.1016/j.ntt.2019.03.003. Data were not clear and interpretation was difficult.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes reported.

Additional Comments: This form is for behavior at day 7 for regenerative adults. The data are located in Supplement 1 xls. Behavior data includes assays for: Speed, rest, and phototaxsis.

Overall Quality Determination Medium

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Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Dugesia japonica*; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064285

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity or grade of test substance was not reported
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The study provided few details on exposure media preparation
	Metric 8:	Consistency of Exposure Administration	Medium	Reporting omissions were unlikely to have a substantial impact on results
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure 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 (0.5% DMSO was appropriate). The study authors had "previously shown that there are no effects onplanarian morphology or behavior at this concentration."
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects, tests were repeated three times
Domain 5: Outcome Assessment				

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Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Dugesia japonica*; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064285

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 reported the intended outcome of interest with some uncertainty related to some results considered invalid. The study authors reported "LC50 might not be accurate, due to low resolution of lethality data in tested concentrations." Supplementary Table 2. LC50 values in full worms and regenerating tails at day 12.
	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	Low	There was no information in the study to suggest differences among groups in animal attrition or health outcomes. The low resolution may have impacted the LC50 for TBBPA.
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: A concern exists regarding the certainty of the LC50 value due to the low resolution. Additionally, the methodology focused on high throughput screen as opposed to focusing on the specifics of any given study with any of the 87 chemicals assessed.

Overall Quality Determination Medium

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Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10064285

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity or grade of test substance was not reported
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The study provided few details on exposure media preparation
	Metric 8:	Consistency of Exposure Administration	Medium	Reporting omissions were unlikely to have a substantial impact on results
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure 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 (0.5% DMSO was appropriate). The study authors had "previously shown that there are no effects onplanarian morphology or behavior at this concentration."
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects, tests were repeated three times
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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Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Dugesia japonica*; regenerating; Adult
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Domain	Metric		Rating	Comments
	Metric 17:	Outcome Assessment Methodology	Medium	The outcome assessment methodology reported the intended outcome of interest with some uncertainty related to some results considered invalid. The study authors reported "LC50 might not be accurate, due to low resolution of lethality data in tested concentrations." Supplementary Table 2. LC50 values in full worms and regenerating tails at day 12.
	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	Low	There was no information in the study to suggest differences among groups in animal attrition or health outcomes. The low resolution may have impacted the LC50 for TBBPA.
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: A concern exists regarding the certainty of the LC50 value due to the low resolution. Additionally, the methodology focused on high throughput screen as opposed to focusing on the specifics of any given study with any of the 87 chemicals assessed.

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Health Outcome:	Behavioral
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HERO ID:	10064285

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity or grade of test substance was not reported
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The study provided few details on exposure media preparation
	Metric 8:	Consistency of Exposure Administration	Low	Reporting omissions were unlikely to have a substantial impact on results
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
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	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration (0.5% DMSO was appropriate). The study authors had ”previously shown that there are no effects onplanarian morphology or behavior at this concentration.”
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	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, tests were repeated three times
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 reported the intended outcome of interest with some uncertainty related to some results considered invalid. Control organisms for the scrunching behavior were inconsistent between replicates (spreadsheet) and error bars/standard deviations were not provided by the study authors making any kind of comparison inconclusive even though it appeared TBBPA had some kind of effect, especially at the highest concentration tested. Additionally, it is not possible to determine any kind of definitive endpoint from this behavior assessment (scrunching). The conclusions would be similar for simulated and unstimulated behaviors reported by the study authors.
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described, while not highlighted, it is assumed that no concentrations had significant effects based on fig.S5
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained

Additional Comments: The behavioral studies reported by the study authors should be interpreted with caution. Control organisms in full and regenerating organisms exhibited effects similar to treatment concentrations of TBBPA. It is unclear which effects would be considered significant effects since error bars/standard error was not reported by the study authors. This applies to resting time, phototaxis, thermotaxis, and scrunching behavior. There does appear to be an effect with speed, but again, there is nothing denoting at which concentrations the effects are significant. While it does appear effects may be occurring as a result of TBBPA, results as reported are inconclusive.

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	Metric 2:	Test Substance Source	Low	Sourced from Sigma Aldrich, but chemical was not analytically verified by the laboratory.
	Metric 3:	Test Substance Purity	High	reported as 99.77% pure.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Authors did not use a true negative control. "Negative" controls contained either acetaminophen or L-ascorbic acid. A positive solvent control (0.5% DMSO) was also used. The authors presented information from previous studies to verify that acetaminophen or L-ascorbic acid did not affect the test organisms.
	Metric 5:	Negative Control Response	High	100% survival in solvent control (0.5% DMSO), no mortality observed in "negative" controls.
	Metric 6:	Randomized Allocation	Low	No mention of randomization in this paper. Author did cite another paper which may have described randomization.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The experimental system and methods for preparation of test media were described in adequate detail and appropriately accounted for the physical-chemical properties of the test substance (solvent used due to the low solubility of the test material). Measured test concentrations were not provided.
	Metric 8:	Consistency of Exposure Administration	High	No inconsistencies were noticed or reported
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured, highest concentration listed as 99.5 uM in supplemental data instead of a nominal value of 100 uM as stated in text
	Metric 10:	Exposure Duration and Frequency	High	Exposure duration (12 days) was adequate to cause mortality for some of the chemicals in this study (mortalities seen in TBBPA, but not TCEP).
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Exposure concentrations seemed adequate to illicit a response (mortality) for most chemicals tested in this study, TBBPA included. This was below the NOAEL for TCEP, but the purpose of the study was to compare the mortality between chemicals within a specific group, so this was not determined to affect the rating of this study.
	Metric 12:	Testing at or Below Solubility Limit	Medium	Solvent (0.5% DMSO) was used to facilitate the dissolution of the test material into the test medium. The two highest concentrations (100 and 10 uM) are above the solubility listed in the final scope (4.1 mg/L).

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Study Citation:	Zhang, S., Ireland, D., Sipes, N. S., Behl, M., Collins, E. S. (2019). Screening for neurotoxic potential of 15 flame retardants using freshwater planarians. Neurotoxicology and Teratology 73:54-66.
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; Worms (e.g., Annelids, Nematodes); *Dugesia japonica*; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5469417

Domain	Metric		Rating	Comments
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Supplemental methods description describes the test organism in more detail (Zhang et al., 2019). Test organisms were described as adults and "Test worms were manually selected to fall within a certain range of sizes and we found full worm length, after automated size measurement, to be 7.3 6 2.3 mm (mean 6 SD), and tail worm length to be 7.3 mm þ/- 2.7 mm (mean 6 SD).
	Metric 14:	Acclimatization and Pretreatment Conditions	High	In supplemental materials, pretreatment procedure was described as: "Animals were starved for at least 5 days before beingused for experiments and their containers were cleaned immediately prior to worm selection for experiments." This was adequate for gut-clearance.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	8 individuals were used for each replicate.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Test conditions were fully described in supplemental methods paper (Zhang et al., 2019): "Wells of a 48- well plate with 200 ml of IO water." The authors noted that in the present study, they updated the feeding procedure, but this was not described clearly, "we fed planarians used in FR screen 2 commercial freeze-dried organic chicken liver (Amazon, Seattle, WA) to better control food quality and thus minimize animal fitness variability." It was unclear when this supplemental feeding occurred during the exposure period or if the lack of feeding could have contributed to additional mortality in some chemicals in the screen.
	Metric 17:	Outcome Assessment Methodology	High	Outcome assessment was adequate to describe the comparative mortality of several flame retardants following exposure to the test material.
	Metric 18:	Consistency of Outcome Assessment	High	Assessment methods seemed consistent across groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. Besides the feeding procedure, there were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure (e.g., infection) that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				

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Study Citation:	Zhang, S., Ireland, D., Sipes, N. S., Behl, M., Collins, E. S. (2019). Screening for neurotoxic potential of 15 flame retardants using freshwater planarians. Neurotoxicology and Teratology 73:54-66.
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; Worms (e.g., Annelids, Nematodes); *Dugesia japonica*; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5469417

Domain	Metric		Rating	Comments
	Metric 21:	Statistical Methods	Low	The authors describe the statistical procedure as: "Statistical significance was determined using either a one-tailed Fisher's exact test for lethality, eye regeneration, phototaxis, and scrunching endpoints; Mann Whitney U test for thermotaxis and unstimulated behavior; or two-tailed t-test for unstimulated behavior (depending on normality of the sample) using a significance level of 0.05. This was adequate to determine if mortality was significant." This analysis and the raw data describing mortality were not presented in this study.
	Metric 22:	Reporting of Data	Low	A large amount of data was presented in the supplemental file (Zhang et al., 2019): https://doi.org/10.1016/j.ntt.2019.03.003. Data were not clear and interpretation was difficult.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes reported.

Additional Comments: This form is for the mortality test for the adult organisms conducted on day 12. Mortality data is located in supplemental 1 xls.

Overall Quality Determination Medium

Study Citation:	Zhang, S., Ireland, D., Sipes, N. S., Behl, M., Collins, E. S. (2019). Screening for neurotoxic potential of 15 flame retardants using freshwater planarians. Neurotoxicology and Teratology 73:54-66.
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; Worms (e.g., Annelids, Nematodes); *Dugesia japonica*; regenerative; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5469417

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Test substance clearly identified with name and CAS #
	Metric 2:	Test Substance Source	Low	Sourced from Sigma Aldrich, but chemical was not analytically verified by the laboratory.
	Metric 3:	Test Substance Purity	High	reported as 99.77% pure
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Authors did not use a true negative control. "Negative" controls contained either acetaminophen or L-ascorbic acid. A positive solvent control (0.5% DMSO) was also used. The authors presented information from previous studies to verify that acetaminophen or L-ascorbic acid did not affect the test organisms.
	Metric 5:	Negative Control Response	High	100% survival in solvent control (0.5% DMSO), no mortality observed in "negative" controls
	Metric 6:	Randomized Allocation	Low	No mention of randomization in this paper. Author did cite another paper which may have described randomization.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The experimental system and methods for preparation of test media were described in adequate detail and appropriately accounted for the physical-chemical properties of the test substance (solvent used due to the low solubility of the test material). Measured test concentrations were not provided.
	Metric 8:	Consistency of Exposure Administration	High	No inconsistencies were noticed or reported
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured, highest concentration listed as 99.5 uM in supplemental data instead of a nominal value of 100 uM as stated in text
	Metric 10:	Exposure Duration and Frequency	High	Exposure duration (12 days) was adequate to cause mortality for some of the chemicals in this study (mortalities seen in TBBPA, but not TCEP).
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Exposure concentrations seemed adequate to illicit a response (mortality) for most chemicals tested in this study, TBBPA included. This was below the NOAEL for TCEP, but the purpose of the study was to compare the mortality between chemicals within a specific group, so this was not determined to affect the rating of this study.
	Metric 12:	Testing at or Below Solubility Limit	Medium	Solvent (0.5% DMSO) was used to facilitate the dissolution of the test material into the test medium. The two highest concentrations (100 and 10 uM) are above the solubility listed in the final scope (4.1 mg/L).
Domain 4: Test Organism				

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Study Citation:	Zhang, S., Ireland, D., Sipes, N. S., Behl, M., Collins, E. S. (2019). Screening for neurotoxic potential of 15 flame retardants using freshwater planarians. Neurotoxicology and Teratology 73:54-66.
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; Worms (e.g., Annelids, Nematodes); *Dugesia japonica*; regenerative; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5469417

Domain	Metric		Rating	Comments
	Metric 13:	Test Organism Characteristics	High	Supplemental methods description describes the test organism in more detail (Zhang et al., 2019). Test organisms were described as adults and "Test worms were manually selected to fall within a certain range of sizes and we found full worm length, after automated size measurement, to be 7.3 6 2.3 mm (mean 6 SD), and tail worm length to be 7.3 mm þ/- 2.7 mm (mean 6 SD).
	Metric 14:	Acclimatization and Pretreatment Conditions	High	In supplemental materials, pretreatment procedure was described as: "Animals were starved for at least 5 days before beingused for experiments and their containers were cleaned immediately prior to worm selection for experiments." This was adequate for gut-clearance.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	8 individuals were used for each replicate.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Test conditions were fully described in supplemental methods paper (Zhang et al., 2019): "Wells of a 48- well plate with 200 ml of IO water." The authors noted that in the present study, they updated the feeding procedure, but this was not described clearly, "we fed planarians used in FR screen 2 commercial freeze-dried organic chicken liver (Amazon, Seattle, WA) to better control food quality and thus minimize animal fitness variability." It was unclear when this supplemental feeding occurred during the exposure period or if the lack of feeding could have contributed to additional mortality in some chemicals in the screen.
	Metric 17:	Outcome Assessment Methodology	High	Outcome assessment was adequate to describe the comparative mortality of several flame retardants following exposure to the test material.
	Metric 18:	Consistency of Outcome Assessment	High	Assessment methods seemed consistent across groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. Besides the feeding procedure, there were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure (e.g., infection) that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				

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Study Citation:	Zhang, S., Ireland, D., Sipes, N. S., Behl, M., Collins, E. S. (2019). Screening for neurotoxic potential of 15 flame retardants using freshwater planarians. Neurotoxicology and Teratology 73:54-66.
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; Worms (e.g., Annelids, Nematodes); *Dugesia japonica*; regenerative; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5469417

Domain	Metric		Rating	Comments
	Metric 21:	Statistical Methods	Low	The authors describe the statistical procedure as: "Statistical significance was determined using either a one-tailed Fisher's exact test for lethality, eye regeneration, phototaxis, and scrunching endpoints; Mann Whitney U test for thermotaxis and unstimulated behavior; or two-tailed t-test for unstimulated behavior (depending on normality of the sample) using a significance level of 0.05. This was adequate to determine if mortality was significant." This analysis and the raw data describing mortality were not presented in this study.
	Metric 22:	Reporting of Data	Low	A large amount of data was presented in the supplemental file (Zhang et al., 2019): https://doi.org/10.1016/j.ntt.2019.03.003. Data were not clear and interpretation was difficult.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes reported.

Additional Comments: This form is for the mortality test for the regenerating adult organisms at dat 12. The data for mortality are located in Supplement 1 xls.

Overall Quality Determination Medium

Study Citation:	Zhang, S., Ireland, D., Sipes, N. S., Behl, M., Collins, E. S. (2019). Screening for neurotoxic potential of 15 flame retardants using freshwater planarians. Neurotoxicology and Teratology 73:54-66.
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; Worms (e.g., Annelids, Nematodes); *Dugesia japonica*; regenerative; Adult
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5469417

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Test substance clearly identified with name and CAS #
	Metric 2:	Test Substance Source	Low	Sourced from Sigma Aldrich, but chemical was not analytically verified by the laboratory.
	Metric 3:	Test Substance Purity	High	reported as 99.77% pure
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Authors did not use a true negative control. ”Negative” controls contained either acetaminophen or L-ascorbic acid. A positive solvent control (0.5% DMSO) was also used. The authors presented information from previous studies to verify that acetaminophen or L-ascorbic acid did not affect the test organisms.
	Metric 5:	Negative Control Response	High	No effects observed in ”negative” or solvent controls
	Metric 6:	Randomized Allocation	Low	No mention of randomization in this paper. Author did cite another paper which may have described randomization.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The experimental system and methods for preparation of test media were described in adequate detail and appropriately accounted for the physical-chemical properties of the test substance (solvent used due to the low solubility of the test material). Measured test concentrations were not provided.
	Metric 8:	Consistency of Exposure Administration	High	No inconsistencies were noticed or reported
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured, highest concentration listed as 99.5 uM in supplemental data instead of a nominal value of 100 uM as stated in text
	Metric 10:	Exposure Duration and Frequency	High	Exposure duration (12 days) was adequate to elicit a response for some of the chemicals in this study.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Exposure concentrations seemed adequate to elicit a response (behavior) for most chemicals tested in this study.
	Metric 12:	Testing at or Below Solubility Limit	Medium	Solvent (0.5% DMSO) was used to facilitate the dissolution of the test material into the test medium. The two highest concentrations (100 and 10 uM) are above the solubility listed in the final scope (4.1 mg/L).
Domain 4: Test Organism				

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Study Citation:	Zhang, S., Ireland, D., Sipes, N. S., Behl, M., Collins, E. S. (2019). Screening for neurotoxic potential of 15 flame retardants using freshwater planarians. Neurotoxicology and Teratology 73:54-66.
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; Worms (e.g., Annelids, Nematodes); *Dugesia japonica*; regenerative; Adult
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5469417

Domain	Metric		Rating	Comments
	Metric 13:	Test Organism Characteristics	High	Supplemental methods description describes the test organism in more detail (Zhang et al., 2019). Test organisms were described as adults and "Test worms were manually selected to fall within a certain range of sizes and we found full worm length, after automated size measurement, to be 7.3 6 2.3 mm (mean 6 SD), and tail worm length to be 7.3 mm þ/- 2.7 mm (mean 6 SD).
	Metric 14:	Acclimatization and Pretreatment Conditions	High	In supplemental materials, pretreatment procedure was described as: "Animals were starved for at least 5 days before beingused for experiments and their containers were cleaned immediately prior to worm selection for experiments." This was adequate for gut-clearance.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	8 individuals were used for each replicate.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Test conditions were fully described in supplemental methods paper (Zhang et al., 2019): "Wells of a 48- well plate with 200 ml of IO water." The authors noted that in the present study, they updated the feeding procedure, but this was not described clearly, "we fed planarians used in FR screen 2 commercial freeze-dried organic chicken liver (Amazon, Seattle, WA) to better control food quality and thus minimize animal fitness variability." It was unclear when this supplemental feeding occurred during the exposure period or if the lack of feeding could have contributed to additional mortality in some chemicals in the screen.
	Metric 17:	Outcome Assessment Methodology	High	Outcome assessment was adequate
	Metric 18:	Consistency of Outcome Assessment	High	Assessment methods seemed consistent across groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. Besides the feeding procedure, there were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure (e.g., infection) that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				

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Study Citation:	Zhang, S., Ireland, D., Sipes, N. S., Behl, M., Collins, E. S. (2019). Screening for neurotoxic potential of 15 flame retardants using freshwater planarians. Neurotoxicology and Teratology 73:54-66.
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; Worms (e.g., Annelids, Nematodes); *Dugesia japonica*; regenerative; Adult
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5469417

Domain	Metric		Rating	Comments
	Metric 21:	Statistical Methods	Low	The authors describe the statistical procedure as: "Statistical significance was determined using either a one-tailed Fisher's exact test for lethality, eye regeneration, phototaxis, and scrunching endpoints; Mann Whitney U test for thermotaxis and unstimulated behavior; or two-tailed t-test for unstimulated behavior (depending on normality of the sample) using a significance level of 0.05. This was adequate to determine if mortality was significant." This analysis and the raw data describing mortality were not presented in this study.
	Metric 22:	Reporting of Data	Low	A large amount of data was presented in the supplemental file (Zhang et al., 2019): https://doi.org/10.1016/j.ntt.2019.03.003. Data were not clear and interpretation was difficult.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes reported.

Additional Comments: This form is for behavior at day 12 for regenerative adults. The data are located in Supplement 1 xls. Behavior data includes assays for: Speed, rest, phototaxsis, thermotaxsis, and scrunching.

Overall Quality Determination Medium

Study Citation:	Zhang, S., Ireland, D., Sipes, N. S., Behl, M., Collins, E. S. (2019). Screening for neurotoxic potential of 15 flame retardants using freshwater planarians. Neurotoxicology and Teratology 73:54-66.
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; Worms (e.g., Annelids, Nematodes); *Dugesia japonica*; Adult
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5469417

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Test substance clearly identified with name and CAS #
	Metric 2:	Test Substance Source	Low	Sourced from Sigma Aldrich, but chemical was not analytically verified by the laboratory.
	Metric 3:	Test Substance Purity	High	reported as 99.77% pure
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Authors did not use a true negative control. ”Negative” controls contained either acetaminophen or L-ascorbic acid. A positive solvent control (0.5% DMSO) was also used. The authors presented information from previous studies to verify that acetaminophen or L-ascorbic acid did not affect the test organisms.
	Metric 5:	Negative Control Response	High	No effects observed in ”negative” or solvent controls
	Metric 6:	Randomized Allocation	Low	No mention of randomization in this paper. Author did cite another paper which may have described randomization.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The experimental system and methods for preparation of test media were described in adequate detail and appropriately accounted for the physical-chemical properties of the test substance (solvent used due to the low solubility of the test material). Measured test concentrations were not provided.
	Metric 8:	Consistency of Exposure Administration	High	No inconsistencies were noticed or reported
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured, highest concentration listed as 99.5 uM in supplemental data instead of a nominal value of 100 uM as stated in text
	Metric 10:	Exposure Duration and Frequency	High	Exposure duration (12 days) was adequate to elicit a response for some of the chemicals in this study.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Exposure concentrations seemed adequate to elicit a response (behavior) for most chemicals tested in this study.
	Metric 12:	Testing at or Below Solubility Limit	High	Solvent (0.5% DMSO) was used to facilitate the dissolution of the test material into the test medium.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Supplemental methods description describes the test organism in more detail (Zhang et al., 2019). Test organisms were described as adults and ”Test worms were manually selected to fall within a certain range of sizes and we found full worm length, after automated size measurement, to be 7.3 6 2.3 mm (mean 6 SD), and tail worm length to be 7.3 mm þ/- 2.7 mm (mean 6 SD).

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Study Citation:	Zhang, S., Ireland, D., Sipes, N. S., Behl, M., Collins, E. S. (2019). Screening for neurotoxic potential of 15 flame retardants using freshwater planarians. Neurotoxicology and Teratology 73:54-66.
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; Worms (e.g., Annelids, Nematodes); *Dugesia japonica*; Adult
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5469417

Domain	Metric		Rating	Comments
	Metric 14:	Acclimatization and Pretreatment Conditions	High	In supplemental materials, pretreatment procedure was described as: ”Animals were starved for at least 5 days before beingused for experiments and their containers were cleaned immediately prior to worm selection for experiments.” This was adequate for gut-clearance.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	8 individuals were used for each replicate.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Test conditions were fully described in supplemental methods paper (Zhang et al., 2019): ”Wells of a 48- well plate with 200 ml of IO water.” The authors noted that in the present study, they updated the feeding procedure, but this was not described clearly, ”we fed planarians used in FR screen 2 commercial freeze-dried organic chicken liver (Amazon, Seattle, WA) to better control food quality and thus minimize animal fitness variability.” It was unclear when this supplemental feeding occurred during the exposure period or if the lack of feeding could have contributed to additional mortality in some chemicals in the screen.
	Metric 17:	Outcome Assessment Methodology	High	Outcome assessment was adequate
	Metric 18:	Consistency of Outcome Assessment	High	Assessment methods seemed consistent across groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. Besides the feeding procedure, there were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure (e.g., infection) that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	The authors describe the statistical procedure as: ”Statistical significance was determined using either a one-tailed Fisher’s exact test for lethality, eye regeneration, phototaxis, and scrunching endpoints; Mann Whitney U test for thermotaxis and unstimulated behavior; or two-tailed t-test for unstimulated behavior (depending on normality of the sample) using a significance level of 0.05. This was adequate to determine if mortality was significant.” This analysis and the raw data describing mortality were not presented in this study.

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Study Citation:	Zhang, S., Ireland, D., Sipes, N. S., Behl, M., Collins, E. S. (2019). Screening for neurotoxic potential of 15 flame retardants using freshwater planarians. Neurotoxicology and Teratology 73:54-66.
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; Worms (e.g., Annelids, Nematodes); *Dugesia japonica*; Adult
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5469417

Domain	Metric		Rating	Comments
	Metric 22:	Reporting of Data	Low	A large amount of data was presented in the supplemental file (Zhang et al., 2019): https://doi.org/10.1016/j.ntt.2019.03.003. Data were not clear and interpretation was difficult.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes reported.

Additional Comments: This form is for behavior at day 12 for adults. The data are located in Supplement 1 xls. Behavior data includes assays for: Speed, rest, phototaxsis, thermotaxsis, and scrunching.

Overall Quality Determination Medium

Study Citation:	Ltd,, W.I. (2006). Tetrabromobisphenol-A (TBBPA): A prolonged sediment toxicity test with Hyalella azteca using spiked sediment.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	7415563

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS# and structure on page 56/104.
	Metric 2:	Test Substance Source	High	The test substance identity was analytically verified by a commercial laboratory. Reported on page 55/104.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.2%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using appropriate concurrent negative control groups.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods. Results are reported as nominal.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via sediment.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health.

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Study Citation:	Ltd,, W.I. (2006). Tetrabromobisphenol-A (TBBPA): A prolonged sediment toxicity test with Hyalella azteca using spiked sediment.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	7415563

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 were no information presented 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: Results for mortality are on page 19/104, table 10 on page 32/103.

Overall Quality Determination High

Study Citation:	Ltd,, W.I. (2006). Tetrabromobisphenol-A (TBBPA): A prolonged sediment toxicity test with Hyalella azteca using spiked sediment.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	7415563

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS# and structure on page 56/104.
	Metric 2:	Test Substance Source	High	The test substance identity was analytically verified by a commercial laboratory. Reported on page 55/104.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.2%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using appropriate concurrent negative control groups.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods. Results are reported as nominal.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via sediment.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups.

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Study Citation:	Ltd,, W.I. (2006). Tetrabromobisphenol-A (TBBPA): A prolonged sediment toxicity test with Hyalella azteca using spiked sediment.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	7415563

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 information presented 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: Results for growth are on page 20/104, table 10 on page 32/103.

Overall Quality Determination High

Study Citation:	Debenest, T., Gagné, F., Petit, A. N., André, C., Kohli, M., Blaise, C. (2010). Ecotoxicity of a brominated flame retardant (tetrabromobisphenol A) and its derivatives to aquatic organisms. Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 152(4):407-412.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; *Hydra attenuata a*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1294788

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%
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Calculation implies that a negative control was used
	Metric 5:	Negative Control Response	Low	The biological response of the negative control groups was not reported.
	Metric 6:	Randomized Allocation	Low	No random allocation was reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
	Metric 8:	Consistency of Exposure Administration	Low	details of exposure administration were not provided
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured or measurements were not reported
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
	Metric 12:	Testing at or Below Solubility Limit	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test animals was not reported
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized
	Metric 15:	Number of Organisms and Replicates per Group	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Reported environmental conditions were not sufficient to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed

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Study Citation:	Debenest, T., Gagné, F., Petit, A. N., André, C., Kohli, M., Blaise, C. (2010). Ecotoxicity of a brominated flame retardant (tetrabromobisphenol A) and its derivatives to aquatic organisms. Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 152(4):407-412.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; *Hydra attenuata a*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1294788

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22:	Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	Low	unexpected outcomes were not reported

Additional Comments: None

Overall Quality Determination Low

Study Citation:	He, Q., Wang, X., Sun, P., Wang, Z., Wang, L. (2015). Acute and chronic toxicity of tetrabromobisphenol A to three aquatic species under different pH conditions. Aquatic Toxicology 164:145-154.
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; Worms (e.g., Annelids, Nematodes); *Limnodrilus hoffmeisteri*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Immobilization
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3350555

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	High	The test substance identity was not analytically verified by the performing laboratory but source was clearly identified.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	Low	The biological response of the negative control groups was not reported
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The experimental 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 but few details were provided
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods
	Metric 10:	Exposure Duration and Frequency	Medium	Exposure duration was shorted than standard
	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	all pretreatment conditions were the same for control and exposed organisms,
	Metric 15:	Number of Organisms and Replicates per Group	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the amount of details provided in the study being reviewed.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Few details provided so difficult to assess

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Study Citation:	He, Q., Wang, X., Sun, P., Wang, Z., Wang, L. (2015). Acute and chronic toxicity of tetrabromobisphenol A to three aquatic species under different pH conditions. Aquatic Toxicology 164:145-154.
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; Worms (e.g., Annelids, Nematodes); *Limnodrilus hoffmeisteri*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Immobilization
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3350555

Domain	Metric		Rating	Comments
	Metric 17:	Outcome Assessment Methodology	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
	Metric 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	pH was varied across experiments but was consistent across treatments
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups within an exposure
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described
	Metric 22:	Reporting of Data	Medium	only resultant EC50s were reported
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcome

Additional Comments: pH effects and mixture results with DMSO also reported

Overall Quality Determination High

Study Citation:	Yang, S. W., Yan, Z. G., Xu, F. F., Wang, S. R., Wu, F. C. (2012). Development of freshwater aquatic life criteria for tetrabromobisphenol A in China. Environmental Pollution 169:59-63.
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; Worms (e.g., Annelids, Nematodes); *Limnodrilus hoffmeisteri*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2562857

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%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using appropriate concurrent negative control groups
	Metric 5:	Negative Control Response	High	Mortality was less than 8 percent
	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	The study provided few details on exposure administration
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and 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 above the solubility limit listed in the Final TBBPA Scope (4.15 mg/L) were included, however an ethanol solvent was used and a solvent control was indicated.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed

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Study Citation:	Yang, S. W., Yan, Z. G., Xu, F. F., Wang, S. R., Wu, F. C. (2012). Development of freshwater aquatic life criteria for tetrabromobisphenol A in China. Environmental Pollution 169:59-63.
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; Worms (e.g., Annelids, Nematodes); *Limnodrilus hoffmeisteri*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2562857

Domain	Metric		Rating	Comments
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the outcome assessment were not reported.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	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
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: None

Overall Quality Determination Medium

Study Citation:	Ltd,, W.I. (2002). Tetrabromobisphenol A: A prolonged sediment toxicity test with Lumbriculus variegatus using spiked sediment with 2% total organic carbon.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, Sediment; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Lumbriculus variegatus*; Adult
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	7769074

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 a commercial laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 98.91%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and 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	N/A	Exposure was via sediment.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions and all pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health.

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Study Citation:	Ltd,, W.I. (2002). Tetrabromobisphenol A: A prolonged sediment toxicity test with Lumbriculus variegatus using spiked sediment with 2% total organic carbon.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, Sediment; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Lumbriculus variegatus*; Adult
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	7769074

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 were no information presented 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: Results for growth are on page 16/104, table 4 on page 21/103.

Overall Quality Determination High

Study Citation:	Ltd,, W.I. (2002). Tetrabromobisphenol A: A prolonged sediment toxicity test with Lumbriculus variegatus using spiked sediment with 2% total organic carbon.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, Sediment; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Lumbriculus variegatus*; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	7769074

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS# and structure.
	Metric 2:	Test Substance Source	High	The test substance identity was analytically verified by a commercial laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 98.91%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and 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	N/A	Exposure was via sediment.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions and all pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	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:	Ltd,, W.I. (2002). Tetrabromobisphenol A: A prolonged sediment toxicity test with Lumbriculus variegatus using spiked sediment with 2% total organic carbon.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, Sediment; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Lumbriculus variegatus*; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	7769074

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 were no information presented 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: Results for mortality are on page 15/104.

Overall Quality Determination High

Study Citation:	Ltd,, W.I. (2002). Tetrabromobisphenol A: A prolonged sediment toxicity test with Lumbriculus variegatus using spiked sediment with 5% total organic carbon.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, Sediment; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Lumbriculus variegatus*; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	7769200

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS# and structure (page 53/104).
	Metric 2:	Test Substance Source	High	The test substance identity was analytically verified by a commercial laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 98.91%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group..
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and 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 justified for a dose response.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via sediment
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions and all pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health.

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Study Citation:	Ltd,, W.I. (2002). Tetrabromobisphenol A: A prolonged sediment toxicity test with Lumbriculus variegatus using spiked sediment with 5% total organic carbon.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, Sediment; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Lumbriculus variegatus*; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	7769200

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 were no information presented 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: Results for mortality are on page 15/104

Overall Quality Determination High

Study Citation:	Ltd,, W.I. (2002). Tetrabromobisphenol A: A prolonged sediment toxicity test with Lumbriculus variegatus using spiked sediment with 5% total organic carbon.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, Sediment; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Lumbriculus variegatus*; Adult
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	7769200

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS# and structure.
	Metric 2:	Test Substance Source	High	The test substance identity was analytically verified by a commercial laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 98.91%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and 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 justified for a dose response.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via sediment.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions and all pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	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:	Ltd,, W.I. (2002). Tetrabromobisphenol A: A prolonged sediment toxicity test with Lumbriculus variegatus using spiked sediment with 5% total organic carbon.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, Sediment; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Lumbriculus variegatus*; Adult
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	7769200

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 were no information presented 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: Results for growth are on page 16/104, table 4 on page 21/104.

Overall Quality Determination High

Study Citation:	Yang, S. W., Yan, Z. G., Xu, F. F., Wang, S. R., Wu, F. C. (2012). Development of freshwater aquatic life criteria for tetrabromobisphenol A in China. Environmental Pollution 169:59-63.
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; *Macrobrachium nipponense*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2562857

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%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using appropriate concurrent negative control groups
	Metric 5:	Negative Control Response	High	Mortality was less than 8 percent
	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	The study provided few details on exposure administration
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and 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 above the solubility limit listed in the Final TBBPA Scope (4.15 mg/L) were included, however an ethanol solvent was used and a solvent control was indicated.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed

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Study Citation:	Yang, S. W., Yan, Z. G., Xu, F. F., Wang, S. R., Wu, F. C. (2012). Development of freshwater aquatic life criteria for tetrabromobisphenol A in China. Environmental Pollution 169:59-63.
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; *Macrobrachium nipponense*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2562857

Domain	Metric		Rating	Comments
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the outcome assessment were not reported.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	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
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: None

Overall Quality Determination Medium

Study Citation:	Souza, C.d., T. T., Castro, G. B., Bernegossi, A. C., Felipe, M. C., Pinheiro, F. R., Colombo-Corbi, V., Girolli, D. A., Gorni, G. R., Corbi, J. J. (2023). Pristina longiseta reproduction test: Chronic exposure to environmental contaminants. Environmental Science and Pollution Research 30(9):23578-23588.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Pristina longiseta*; Adult
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	12812456

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The TBBPA was identified by name only.
	Metric 2:	Test Substance Source	Low	The source of the TBBPA was not reported.
	Metric 3:	Test Substance Purity	Low	The purity was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	The study authors reported the use of a concurrent negative control as well as a solvent control using methanol.
	Metric 5:	Negative Control Response	High	The negative control response and the solvent control response were reported in Figure 2B.
	Metric 6:	Randomized Allocation	Low	It was not reported how the worms were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	A stock solution of 500mg/L TBBPA was prepared in methanol, and dechlorinated tap water was reported to be used as the dilution water in the test, but little else was reported on the preparation of the test substance. The test system was unclear. It is unclear if the test system was static.
	Metric 8:	Consistency of Exposure Administration	Medium	The test was conducted in 60mL of test solution in a glass bottle with 100mL capacity. 10mg of sand was used in each test chamber. Organisms with no apparent reproduction zone were used. It is unclear if the test was static or flow-through.
	Metric 9:	Measurement of Test Substance Concentration	Medium	The TBBPA was quantified using LC-UHPLC-MS/MS. The exposure concentrations were not reported.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration was seven days. This was chosen after monitoring reproduction in 4, 7, and 10 day tests. The seven day duration was determined to be the best duration. This duration was appropriate for the outcome of interest.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	There were five exposure levels. The 7d EC50 value was reported to be <62.5ug/L, which was the lowest test concentration. All the test concentrations observed essentially the same reproductive response. Several lower test concentrations may have allowed for a dose response to be observed and a more accurate EC50 value to be reported.
	Metric 12:	Testing at or Below Solubility Limit	High	The test concentrations were below the water solubility limit.
Domain 4: Test Organism				

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Study Citation:	Souza, C.d., T. T., Castro, G. B., Bernegossi, A. C., Felipe, M. C., Pinheiro, F. R., Colombo-Corbi, V., Girolli, D. A., Gorni, G. R., Corbi, J. J. (2023). Pristina longiseta reproduction test: Chronic exposure to environmental contaminants. Environmental Science and Pollution Research 30(9):23578-23588.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Pristina longiseta*; Adult
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	12812456

Domain	Metric		Rating	Comments
	Metric 13:	Test Organism Characteristics	High	The test organisms were cultured in-house at the Aquatic Ecology Laboratory in Sao Carlos School of Engineering, University of Sao Paulo in Brazil. Worms with no apparent fission zone that were 2-4mm in length were used.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The P. longiseta were cultured under conditions similar to those of the test conditions.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Each test chamber contained six text organisms, and each test concentrations was carried out in triplicate.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	The worms were cultured in dechlorinated test water and kept under constant aeration with a photoperiod of 12L:12D. The culture temperature 25C. A pH of 6.5-7.0 was maintained. Test conditions were essential identical to culture conditions with the addition of the test chemical. The worms were fed 2mg of TetraMin at the beginning of the test.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome(s) of interest–The number of new organisms after a seven day exposure to TBBPA.
	Metric 18:	Consistency of Outcome Assessment	High	The organisms were counted at the end of the seven day study. The offspring were counted from the total number of individuals.
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 adequately and were appropriate for the outcome assessed.
	Metric 22:	Reporting of Data	High	Data for the exposure response, control response, and the solvent control response were reported in Figure 2B. The seven day EC50 value for TBBPA was <62.5ug/L and was reported in Table 1 and in the text.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.

Additional Comments: This evaluation is for the effect of TBBPA on the reproduction of aquatic worm P. longiseta. The number of new worms was counted at the end of a seven day exposure. The 7d EC50 value was reported to be <62.5ug/L.

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Study Citation:	Souza, C.d., T. T., Castro, G. B., Bernegossi, A. C., Felipe, M. C., Pinheiro, F. R., Colombo-Corbi, V., Girolli, D. A., Gorni, G. R., Corbi, J. J. (2023). Pristina longiseta reproduction test: Chronic exposure to environmental contaminants. Environmental Science and Pollution Research 30(9):23578-23588.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Pristina longiseta*; Adult
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	12812456

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

Study Citation:	Debenest, T., Gagné, F., Petit, A. N., André, C., Kohli, M., Blaise, C. (2010). Ecotoxicity of a brominated flame retardant (tetrabromobisphenol A) and its derivatives to aquatic organisms. Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 152(4):407-412.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; *Thamnocephalus platyurus*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1294788

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%
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Calculation implies that a negative control was used
	Metric 5:	Negative Control Response	Low	The biological response of the negative control groups was not reported.
	Metric 6:	Randomized Allocation	Low	No random allocation reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
	Metric 8:	Consistency of Exposure Administration	Low	details of exposure administration were not provided
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured or measurements were not reported
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
	Metric 12:	Testing at or Below Solubility Limit	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test animals was not reported
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized
	Metric 15:	Number of Organisms and Replicates per Group	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Reported environmental conditions were not sufficient to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed

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Study Citation:	Debenest, T., Gagné, F., Petit, A. N., André, C., Kohli, M., Blaise, C. (2010). Ecotoxicity of a brominated flame retardant (tetrabromobisphenol A) and its derivatives to aquatic organisms. Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 152(4):407-412.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; *Thamnocephalus platyurus*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1294788

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22:	Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	Low	unexpected outcomes were not reported

Additional Comments: None

Overall Quality Determination Low

Study Citation:	Debenest, T., Gagné, F., Petit, A. N., Kohli, M., Eullafroy, P., Blaise, C. (2010). Monitoring of a flame retardant (tetrabromobisphenol A) toxicity on different microalgae assessed by flow cytometry. Journal of Environmental Monitoring 12(10):1918-1923.
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; *Chlamydomonas reinhardtii*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5496905

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	"TBBPA was synthesized by the National Water Research Institute(Burlington, Ontario, Canada)." The test substance identity was analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Medium	Few details of exposure administration were reported
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Use of lower concentrations may have been helpful in establishing a clearer dose response
	Metric 12:	Testing at or Below Solubility Limit	Medium	The solvent concentration slightly exceeded an appropriate concentration
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				

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Study Citation:	Debenest, T., Gagné, F., Petit, A. N., Kohli, M., Eullafroy, P., Blaise, C. (2010). Monitoring of a flame retardant (tetrabromobisphenol A) toxicity on different microalgae assessed by flow cytometry. Journal of Environmental Monitoring 12(10):1918-1923.
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; *Chlamydomonas reinhardtii*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5496905

Domain	Metric		Rating	Comments
	Metric 16:	Adequacy of Test Conditions	Medium	Environmental conditions of test system were conducive to maintenance of organism health, few details reported
	Metric 17:	Outcome Assessment Methodology	High	the assessment methodology was sensitive and appropriate for the outcomes of interest.
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	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: viable cell population growth based on chlorophyll autofluorescence

Overall Quality Determination High

Study Citation:	Debenest, T., Gagné, F., Petit, A. N., Kohli, M., Eullafroy, P., Blaise, C. (2010). Monitoring of a flame retardant (tetrabromobisphenol A) toxicity on different microalgae assessed by flow cytometry. Journal of Environmental Monitoring 12(10):1918-1923.
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; *Chlamydomonas reinhardtii*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5496905

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	"TBBPA was synthesized by the National Water Research Institute(Burlington, Ontario, Canada)." The test substance identity was analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Medium	Few details of exposure administration were reported
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Use of lower concentrations may have been helpful in establishing a clearer dose response
	Metric 12:	Testing at or Below Solubility Limit	Medium	The solvent concentration slightly exceeded an appropriate concentration
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Environmental conditions of test system were conducive to maintenance of organism health, few details reported
	Metric 17:	Outcome Assessment Methodology	High	the assessment methodology was sensitive and appropriate for the outcomes of interest.

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Study Citation:	Debenest, T., Gagné, F., Petit, A. N., Kohli, M., Eullafroy, P., Blaise, C. (2010). Monitoring of a flame retardant (tetrabromobisphenol A) toxicity on different microalgae assessed by flow cytometry. Journal of Environmental Monitoring 12(10):1918-1923.
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; *Chlamydomonas reinhardtii*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5496905

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
	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: esterase activity was measured for this data evaluation

Overall Quality Determination High

Study Citation:	Peng, F.,uQ, Ying, G., Yang, B.,in, Liu, Y., Lai, H., Zhou, G., Chen, J.,un, Zhao, J. L. (2014). Biotransformation of the flame retardant tetrabromobisphenol-a (TBBPA) by freshwater microalgae. Environmental Toxicology and Chemistry 33(8):1705-1711.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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; *Chlorella pyrenoidosa*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2523760

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the amount of details provided in the study being reviewed.
	Metric 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 may have a substantial impact on results
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations measurements were not reported
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Exposure groups and spacing were justified for a dose response and adequate for the purpose of the study.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of replicates was not reported.
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:	Peng, F.,uQ, Ying, G., Yang, B.,in, Liu, Y., Lai, H., Zhou, G., Chen, J.,un, Zhao, J. L. (2014). Biotransformation of the flame retardant tetrabromobisphenol-a (TBBPA) by freshwater microalgae. Environmental Toxicology and Chemistry 33(8):1705-1711.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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; *Chlorella pyrenoidosa*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2523760

Domain	Metric		Rating	Comments
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22:	Reporting of Data	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: population growth. TBBPA was not detected or only trace amounts in biotransformation experiment

Overall Quality Determination Medium

Study Citation:	Peng, F.,uQ, Ying, G., Yang, B.,in, Liu, Y., Lai, H., Zhou, G., Chen, J.,un, Zhao, J. L. (2014). Biotransformation of the flame retardant tetrabromobisphenol-a (TBBPA) by freshwater microalgae. Environmental Toxicology and Chemistry 33(8):1705-1711.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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; *Chlorella pyrenoidosa*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Photosynthesis
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2523760

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the amount of details provided in the study being reviewed.
	Metric 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 may have a substantial impact on results
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations measurements were not reported
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Exposure groups and spacing were adequate to address the purpose of the study.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of replicates was not reported.
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:	Peng, F.,uQ, Ying, G., Yang, B.,in, Liu, Y., Lai, H., Zhou, G., Chen, J.,un, Zhao, J. L. (2014). Biotransformation of the flame retardant tetrabromobisphenol-a (TBBPA) by freshwater microalgae. Environmental Toxicology and Chemistry 33(8):1705-1711.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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; *Chlorella pyrenoidosa*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Photosynthesis
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2523760

Domain	Metric		Rating	Comments
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22:	Reporting of Data	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: TBBPA was not detected or only trace amounts in biotransformation experiment

Overall Quality Determination Medium

Study Citation:	Peng, F.,uQ, Ying, G., Yang, B.,in, Liu, Y., Lai, H., Zhou, G., Chen, J.,un, Zhao, J. L. (2014). Biotransformation of the flame retardant tetrabromobisphenol-a (TBBPA) by freshwater microalgae. Environmental Toxicology and Chemistry 33(8):1705-1711.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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; *Chlorella pyrenoidosa*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2523760

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the amount of details provided in the study being reviewed.
	Metric 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 may have a substantial impact on results
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations measurements were not reported
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Exposure groups and spacing were adequate to address the purpose of the study.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of replicates was not reported.
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:	Peng, F.,uQ, Ying, G., Yang, B.,in, Liu, Y., Lai, H., Zhou, G., Chen, J.,un, Zhao, J. L. (2014). Biotransformation of the flame retardant tetrabromobisphenol-a (TBBPA) by freshwater microalgae. Environmental Toxicology and Chemistry 33(8):1705-1711.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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; *Chlorella pyrenoidosa*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2523760

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22:	Reporting of Data	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: This evaluation is for the growth inhibition reported in the supplemental material in Figure S1.

Overall Quality Determination Medium

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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, 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; *Coelastrum sphaericum*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2523760

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the amount of details provided in the study being reviewed.
	Metric 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 may have a substantial impact on results
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations measurements were not reported
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	there is only one exposure concentration
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of replicates was not reported.
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:	Peng, F.,uQ, Ying, G., Yang, B.,in, Liu, Y., Lai, H., Zhou, G., Chen, J.,un, Zhao, J. L. (2014). Biotransformation of the flame retardant tetrabromobisphenol-a (TBBPA) by freshwater microalgae. Environmental Toxicology and Chemistry 33(8):1705-1711.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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; *Coelastrum sphaericum*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2523760

Domain	Metric		Rating	Comments
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22:	Reporting of Data	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: population growth. TBBPA was not detected or only trace amounts in biotransformation experiment

Overall Quality Determination Medium

Study Citation:	Debenest, T., Gagné, F., Petit, A. N., Kohli, M., Eullafroy, P., Blaise, C. (2010). Monitoring of a flame retardant (tetrabromobisphenol A) toxicity on different microalgae assessed by flow cytometry. Journal of Environmental Monitoring 12(10):1918-1923.
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; *Nitzschia palea*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5496905

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	"TBBPA was synthesized by the National Water Research Institute(Burlington, Ontario, Canada)." The test substance identity was analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Medium	Few details of exposure administration were reported
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type."Each species was exposed to 1.8, 4.8, 9.2, 12.9 and 16.5 mmol L-1 of TBBPA for 72 h."
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Use of lower concentrations may have been helpful in establishing a clearer dose response
	Metric 12:	Testing at or Below Solubility Limit	Medium	The solvent concentration slightly exceeded an appropriate concentration
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				

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Study Citation:	Debenest, T., Gagné, F., Petit, A. N., Kohli, M., Eullafroy, P., Blaise, C. (2010). Monitoring of a flame retardant (tetrabromobisphenol A) toxicity on different microalgae assessed by flow cytometry. Journal of Environmental Monitoring 12(10):1918-1923.
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; *Nitzschia palea*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5496905

Domain	Metric		Rating	Comments
	Metric 16:	Adequacy of Test Conditions	Medium	Environmental conditions of test system were conducive to maintenance of organism health, few details reported
	Metric 17:	Outcome Assessment Methodology	High	the assessment methodology was sensitive and appropriate for the outcomes of interest.
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	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: viable cell population growth based on chlorophyll autofluorescence

Overall Quality Determination High

Study Citation:	Debenest, T., Gagné, F., Petit, A. N., Kohli, M., Eullafroy, P., Blaise, C. (2010). Monitoring of a flame retardant (tetrabromobisphenol A) toxicity on different microalgae assessed by flow cytometry. Journal of Environmental Monitoring 12(10):1918-1923.
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; *Nitzschia palea*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5496905

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	”TBBPA was synthesized by the National Water Research Institute(Burlington, Ontario, Canada).” The test substance identity was analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Medium	Few details of exposure administration were reported
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.”Each species was exposed to 1.8, 4.8, 9.2, 12.9 and 16.5 mmol L-1 of TBBPA for 72 h.”
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Use of lower concentrations may have been helpful in establishing a clearer dose response
	Metric 12:	Testing at or Below Solubility Limit	Medium	The solvent concentration slightly exceeded an appropriate concentration
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Environmental conditions of test system were conducive to maintenance of organism health, few details reported

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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:	Vegetation; Non-vascular Plants; *Nitzschia palea*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5496905

Domain	Metric		Rating	Comments
	Metric 17:	Outcome Assessment Methodology	High	the assessment methodology was sensitive and appropriate for the outcomes of interest.
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	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: esterase activity was measured for this data evaluation

Overall Quality Determination High

Study Citation:	Debenest, T., Petit, A. N., Gagné, F., Kohli, M., Nguyen, N., Blaise, C. (2011). Comparative toxicity of a brominated flame retardant (tetrabromobisphenol A) on microalgae with single and multi-species bioassays. Chemosphere 85(1):50-55.
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; *Nitzschia palea*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4509862

Domain		Metric	Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	High	The test substance was synthesized by and analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent solvent control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control groups was reported and was adequate
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Medium	Reporting omissions were unlikely to have a substantial impact on results
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and 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	The solvent concentration was appropriate (0.5%).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17:	Outcome Assessment Methodology	Medium	The outcome assessment methodology reported the intended outcome of interest

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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:	Vegetation; Non-vascular Plants; *Nitzschia palea*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4509862

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	Medium	Data on outcomes unrelated to controlled variables for each study group were not reported because only substantial differences among groups were noted
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not all concentrations were analyzed
	Metric 22:	Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	Medium	Minor uncertainties were identified in how the study characterized unexpected outcomes

Additional Comments: None

Overall Quality Determination Medium

Study Citation:	Debenest, T., Gagné, F., Petit, A. N., André, C., Kohli, M., Blaise, C. (2010). Ecotoxicity of a brominated flame retardant (tetrabromobisphenol A) and its derivatives to aquatic organisms. Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 152(4):407-412.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Non-vascular Plants; *Pseudokirchneriella subcapitata*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1294788

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%
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Calculation implies that a negative control was used
	Metric 5:	Negative Control Response	Low	The biological response of the negative control groups was not reported.
	Metric 6:	Randomized Allocation	Low	No random allocation reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
	Metric 8:	Consistency of Exposure Administration	Low	details of exposure administration were not provided
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured or measurements were not reported
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
	Metric 12:	Testing at or Below Solubility Limit	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test animals was not reported
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized
	Metric 15:	Number of Organisms and Replicates per Group	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Reported environmental conditions were not sufficient to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed

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Taxa, Species, Age:	Vegetation; Non-vascular Plants; *Pseudokirchneriella subcapitata*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1294788

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22:	Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	Low	unexpected outcomes were not reported

Additional Comments: None

Overall Quality Determination Low

Study Citation:	Debenest, T., Gagné, F., Petit, A. N., Kohli, M., Eullafroy, P., Blaise, C. (2010). Monitoring of a flame retardant (tetrabromobisphenol A) toxicity on different microalgae assessed by flow cytometry. Journal of Environmental Monitoring 12(10):1918-1923.
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Taxa, Species, Age:	Vegetation; Non-vascular Plants; *Pseudokirchneriella subcapitata*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5496905

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	"TBBPA was synthesized by the National Water Research Institute(Burlington, Ontario, Canada)." The test substance identity was analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Medium	Few details of exposure administration were reported
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type."Each species was exposed to 1.8, 4.8, 9.2, 12.9 and 16.5 mmol L-1 of TBBPA for 72 h."
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Use of lower concentrations may have been helpful in establishing a clearer dose response
	Metric 12:	Testing at or Below Solubility Limit	Medium	The solvent concentration slightly exceeded an appropriate concentration
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				

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Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
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Domain	Metric		Rating	Comments
	Metric 16:	Adequacy of Test Conditions	Medium	Environmental conditions of test system were conducive to maintenance of organism health, few details reported
	Metric 17:	Outcome Assessment Methodology	High	the assessment methodology was sensitive and appropriate for the outcomes of interest.
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	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: viable cell population growth based on chlorophyll autofluorescence

Overall Quality Determination High

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Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
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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	Low	Purity or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control 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	Few details of exposure administration were reported
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
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Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Environmental conditions of test system were conducive to maintenance of organism health, few details reported
	Metric 17:	Outcome Assessment Methodology	High	the assessment methodology was sensitive and appropriate for the outcomes of interest.

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Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained

Additional Comments: esterase activity was measured for this data evaluation

Overall Quality Determination High

Study Citation:	Debenest, T., Petit, A. N., Gagné, F., Kohli, M., Nguyen, N., Blaise, C. (2011). Comparative toxicity of a brominated flame retardant (tetrabromobisphenol A) on microalgae with single and multi-species bioassays. Chemosphere 85(1):50-55.
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Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4509862

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	High	The test substance was synthesized by and analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent solvent control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control groups was reported and was adequate
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Medium	Reporting omissions were unlikely to have a substantial impact on results
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and 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	The solvent concentration was appropriate (0.5%).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17:	Outcome Assessment Methodology	Medium	The outcome assessment methodology reported the intended outcome of interest

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Study Citation:	Debenest, T., Petit, A. N., Gagné, F., Kohli, M., Nguyen, N., Blaise, C. (2011). Comparative toxicity of a brominated flame retardant (tetrabromobisphenol A) on microalgae with single and multi-species bioassays. Chemosphere 85(1):50-55.
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; *Pseudokirchneriella subcapitata*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4509862

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	Medium	Data on outcomes unrelated to controlled variables for each study group were not reported because only substantial differences among groups were noted
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not all concentrations were analyzed
	Metric 22:	Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	Medium	Minor uncertainties were identified in how the study characterized unexpected outcomes

Additional Comments: None

Overall Quality Determination Medium

Study Citation:	Peng, F.,uQ, Ying, G., Yang, B.,in, Liu, Y., Lai, H., Zhou, G., Chen, J.,un, Zhao, J. L. (2014). Biotransformation of the flame retardant tetrabromobisphenol-a (TBBPA) by freshwater microalgae. Environmental Toxicology and Chemistry 33(8):1705-1711.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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; *Pseudokirchneriella subcapitata*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2523760

Domain		Metric	Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the amount of details provided in the study being reviewed.
	Metric 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 may have a substantial impact on results
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations measurements were not reported
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	there is only one exposure concentration
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of replicates was not reported.
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:	Peng, F.,uQ, Ying, G., Yang, B.,in, Liu, Y., Lai, H., Zhou, G., Chen, J.,un, Zhao, J. L. (2014). Biotransformation of the flame retardant tetrabromobisphenol-a (TBBPA) by freshwater microalgae. Environmental Toxicology and Chemistry 33(8):1705-1711.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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; *Pseudokirchneriella subcapitata*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2523760

Domain	Metric		Rating	Comments
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22:	Reporting of Data	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: population growth. TBBPA was not detected or only trace amounts in biotransformation experiment

Overall Quality Determination Medium

Study Citation:	Sazawa, K., Furuhashi, Y., Hata, N., Taguchi, S., Fukushima, M., Kuramitz, H. (2013). Evaluation of the toxicity of tetrabromobisphenol A and some of its oxidation products using a micro-scale algal growth inhibition test. Toxicological and Environmental Chemistry 95(3):472-482.
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; *Pseudokirchneriella subcapitata*; NIES-35; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5497680

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	baseline values were reported, calculation of EC50 implies use of a control value
	Metric 5:	Negative Control Response	Low	The biological response of the negative control groups was not reported
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Medium	Reporting omissions were unlikely to have a substantial impact on results
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	standard duration was used
	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	Reporting omissions prevented determination of whether exposure concentrations exceeded the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Medium	all pretreatment conditions were the same for control and exposed organisms, but few details were provided
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reported environmental conditions were not sufficient to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest

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Study Citation:	Sazawa, K., Furuhashi, Y., Hata, N., Taguchi, S., Fukushima, M., Kuramitz, H. (2013). Evaluation of the toxicity of tetrabromobisphenol A and some of its oxidation products using a micro-scale algal growth inhibition test. Toxicological and Environmental Chemistry 95(3):472-482.
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; *Pseudokirchneriella subcapitata*; NIES-35; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5497680

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

Additional Comments: humic acid effects also reported

Overall Quality Determination Low

Study Citation:	Peng, F.,uQ, Ying, G., Yang, B.,in, Liu, Y., Lai, H., Zhou, G., Chen, J.,un, Zhao, J. L. (2014). Biotransformation of the flame retardant tetrabromobisphenol-a (TBBPA) by freshwater microalgae. Environmental Toxicology and Chemistry 33(8):1705-1711.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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; *Scenedesmus acuminatus*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2523760

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the amount of details provided in the study being reviewed.
	Metric 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 may have a substantial impact on results
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations measurements were not reported
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	there is only one exposure concentration
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of replicates was not reported.
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:	Peng, F.,uQ, Ying, G., Yang, B.,in, Liu, Y., Lai, H., Zhou, G., Chen, J.,un, Zhao, J. L. (2014). Biotransformation of the flame retardant tetrabromobisphenol-a (TBBPA) by freshwater microalgae. Environmental Toxicology and Chemistry 33(8):1705-1711.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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; *Scenedesmus acuminatus*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2523760

Domain	Metric		Rating	Comments
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22:	Reporting of Data	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: population growth. TBBPA was not detected or only trace amounts in biotransformation experiment

Overall Quality Determination Medium

Study Citation:	Meng, Y., Wang, S., Wang, Z., Ye, N., Fang, H. (2018). Algal toxicity of binary mixtures of zinc oxide nanoparticles and tetrabromobisphenol A: Roles of dissolved organic matters. Environmental Toxicology and Pharmacology 64:78-85.
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; *Scenedesmus obliquus*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5497133

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only. A mixture of zinc nanoparticles was used in combination to TBBPA. Exposure solely to TBBPA did not result in any observed effects. Only one concentration of TBBPA was used.
	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 98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control groups
	Metric 5:	Negative Control Response	High	The biological response of the negative control 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	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
	Metric 8:	Consistency of Exposure Administration	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	only one concentration tested (496 ug/L)
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms,
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of replicates was not reported.
Domain 5: Outcome Assessment				

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Study Citation:	Meng, Y., Wang, S., Wang, Z., Ye, N., Fang, H. (2018). Algal toxicity of binary mixtures of zinc oxide nanoparticles and tetrabromobisphenol A: Roles of dissolved organic matters. Environmental Toxicology and Pharmacology 64:78-85.
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; *Scenedesmus obliquus*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5497133

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	Uninformative	Algal growth was assessed. Only one concentration of TBBPA was evaluated in this study which is not enough to establish a dose-response relationship. According to the study authors, the "the primary objective of this study was to explore the impacts of DOM on the joint toxicity of ZnO NPs and TBBPA at relevant low-exposure concentrations (< 1 mg/L)." Exposure solely to TBBA was evaluated, however, there were no treatment-related effects of TBBPA to the algae. The authors indicate, "As shown in Fig. 2(A), the single TBBPA (496 μg/L), DOM (1 mgC/ L), and DMSO (0.1% v/v) systems had no significant effects on the growth of S. obliquus compared to the control ($p > 0.05$)."
	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 were no differences among groups but few details were provided
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained

Additional Comments: Population growth, mixture with zinc nanoparticles also reported. It is not possible to associate endpoints observed in the study with TBBPA. Only one concentration of TBBPA was evaluated in this study which is not enough to establish a dose-response relationship. According to the study authors, the "the primary objective of this study was to explore the impacts of DOM on the joint toxicity of ZnO NPs and TBBPA at relevant low-exposure concentrations (< 1 mg/L)." Exposure solely to TBBA was evaluated. However, there were no treatment-related effects of TBBPA to the algae. The authors indicate, "As shown in Fig. 2(A), the single TBBPA (496 μg/L), DOM (1 mgC/ L), and DMSO (0.1% v/v) systems had no significant effects on the growth of S. obliquus compared to the control ($p > 0.05$)."

Overall Quality Determination High

Study Citation:	Meng, Y., Wang, S., Wang, Z., Ye, N., Fang, H. (2018). Algal toxicity of binary mixtures of zinc oxide nanoparticles and tetrabromobisphenol A: Roles of dissolved organic matters. Environmental Toxicology and Pharmacology 64:78-85.
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; *Scenedesmus obliquus*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cytotoxicity-Oxidative stress (including redox biology)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5497133

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only. A mixture of zinc nanoparticles was used in combination to TBBPA. Exposure solely to TBBPA did not result in any observed effects. Only one concentration of TBBPA was used.
	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 98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control groups
	Metric 5:	Negative Control Response	High	The biological response of the negative control 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	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
	Metric 8:	Consistency of Exposure Administration	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	only one concentration tested (496 ug/L)
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms,
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of replicates was not reported.
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:	Meng, Y., Wang, S., Wang, Z., Ye, N., Fang, H. (2018). Algal toxicity of binary mixtures of zinc oxide nanoparticles and tetrabromobisphenol A: Roles of dissolved organic matters. Environmental Toxicology and Pharmacology 64:78-85.
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; *Scenedesmus obliquus*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cytotoxicity-Oxidative stress (including redox biology)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5497133

Domain	Metric		Rating	Comments
	Metric 17:	Outcome Assessment Methodology	Uninformative	Figure 4A shows there is no difference between DMSO and TBBPA. Although it appears that the control shows a slight significant difference to TBBPA, "DMSO was used to prepare TBBPA stock solutions." This indicates that it is more appropriate to compare TBBPA to the solvent control than the negative control. Additionally, only one concentration of TBBPA was used making any type of dose/response analysis not possible. Figure 5 further validates a lack of biomarker response from exposure to TBBPA. No difference for membrane permeability was observed.
	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 were no differences among groups but few details were provided
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained

Additional Comments: Biomarkers (mechanistic endpoints) were assessed. ROS, membrane permeability and potential, mixture with zinc nanoparticles also reported. It is not possible to associate endpoints observed in the study with TBBPA. Only one concentration of TBBPA was evaluated in this study which is not enough to establish a dose-response relationship. According to the study authors, the "the primary objective of this study was to explore the impacts of DOM on the joint toxicity of ZnO NPs and TBBPA at relevant low-exposure concentrations ($<$ 1 mg/L)." Exposure solely to TBBA was evaluated. However, there were no treatment-related effects of TBBPA to the algae. The study authors report, "As shown in Fig. 4(A), the individual TBBPA (496 μg/L), DOM (1 mgC/L), and DMSO (0.1% v/v) systems did not significantly change the ROS level in S. obliquus."

Overall Quality Determination Uninformative

Study Citation:	Peng, F.,uQ, Ying, G., Yang, B.,in, Liu, Y., Lai, H., Zhou, G., Chen, J.,un, Zhao, J. L. (2014). Biotransformation of the flame retardant tetrabromobisphenol-a (TBBPA) by freshwater microalgae. Environmental Toxicology and Chemistry 33(8):1705-1711.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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; *Scenedesmus obliquus*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2523760

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the amount of details provided in the study being reviewed.
	Metric 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 may have a substantial impact on results
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations measurements were not reported
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Exposure groups and spacing were justified for a dose response and adequate for the purpose of the study.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of replicates was not reported.
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:	Peng, F.,uQ, Ying, G., Yang, B.,in, Liu, Y., Lai, H., Zhou, G., Chen, J.,un, Zhao, J. L. (2014). Biotransformation of the flame retardant tetrabromobisphenol-a (TBBPA) by freshwater microalgae. Environmental Toxicology and Chemistry 33(8):1705-1711.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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; *Scenedesmus obliquus*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2523760

Domain	Metric		Rating	Comments
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22:	Reporting of Data	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: population growth. TBBPA was not detected or only trace amounts in biotransformation experiment

Overall Quality Determination Medium

Study Citation:	Peng, F.,uQ, Ying, G., Yang, B.,in, Liu, Y., Lai, H., Zhou, G., Chen, J.,un, Zhao, J. L. (2014). Biotransformation of the flame retardant tetrabromobisphenol-a (TBBPA) by freshwater microalgae. Environmental Toxicology and Chemistry 33(8):1705-1711.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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; *Scenedesmus obliquus*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2523760

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the amount of details provided in the study being reviewed.
	Metric 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 may have a substantial impact on results
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations measurements were not reported
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	there is only one exposure concentration
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of replicates was not reported.
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:	Peng, F.,uQ, Ying, G., Yang, B.,in, Liu, Y., Lai, H., Zhou, G., Chen, J.,un, Zhao, J. L. (2014). Biotransformation of the flame retardant tetrabromobisphenol-a (TBBPA) by freshwater microalgae. Environmental Toxicology and Chemistry 33(8):1705-1711.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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; *Scenedesmus obliquus*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2523760

Domain	Metric		Rating	Comments
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22:	Reporting of Data	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: This evaluation is for the growth inhibition reported in the supplemental material in Figure S1.

Overall Quality Determination Medium

Study Citation:	Peng, F.,uQ, Ying, G., Yang, B.,in, Liu, Y., Lai, H., Zhou, G., Chen, J.,un, Zhao, J. L. (2014). Biotransformation of the flame retardant tetrabromobisphenol-a (TBBPA) by freshwater microalgae. Environmental Toxicology and Chemistry 33(8):1705-1711.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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; *Scenedesmus obliquus*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Photosynthesis
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2523760

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the amount of details provided in the study being reviewed.
	Metric 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 may have a substantial impact on results
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations measurements were not reported
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	there is only one exposure concentration
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of replicates was not reported.
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:	Peng, F.,uQ, Ying, G., Yang, B.,in, Liu, Y., Lai, H., Zhou, G., Chen, J.,un, Zhao, J. L. (2014). Biotransformation of the flame retardant tetrabromobisphenol-a (TBBPA) by freshwater microalgae. Environmental Toxicology and Chemistry 33(8):1705-1711.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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; *Scenedesmus obliquus*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Photosynthesis
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2523760

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22:	Reporting of Data	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: TBBPA was not detected or only trace amounts in biotransformation experiment

Overall Quality Determination Medium

Study Citation:	Yang, S. W., Yan, Z. G., Xu, F. F., Wang, S. R., Wu, F. C. (2012). Development of freshwater aquatic life criteria for tetrabromobisphenol A in China. Environmental Pollution 169:59-63.
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; *Scenedesmus quadricanda*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2562857

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%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using appropriate concurrent negative control groups
	Metric 5:	Negative Control Response	High	Mortality was less than 8 percent
	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	The study provided few details on exposure administration
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and 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 above the solubility limit listed in the Final TBBPA Scope (4.15 mg/L) were included, however an ethanol solvent was used and a solvent control was indicated.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed

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Study Citation:	Yang, S. W., Yan, Z. G., Xu, F. F., Wang, S. R., Wu, F. C. (2012). Development of freshwater aquatic life criteria for tetrabromobisphenol A in China. Environmental Pollution 169:59-63.
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; *Scenedesmus quadricanda*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2562857

Domain	Metric		Rating	Comments
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was not clearly reported
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the outcome assessment were not reported.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	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
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: None

Overall Quality Determination Medium

Study Citation:	Peng, F.,uQ, Ying, G., Yang, B.,in, Liu, Y., Lai, H., Zhou, G., Chen, J.,un, Zhao, J. L. (2014). Biotransformation of the flame retardant tetrabromobisphenol-a (TBBPA) by freshwater microalgae. Environmental Toxicology and Chemistry 33(8):1705-1711.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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; *Scenedesmus quadricauda*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2523760

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the amount of details provided in the study being reviewed.
	Metric 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 may have a substantial impact on results
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations measurements were not reported
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	there is only one exposure concentration
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of replicates was not reported.
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:	Peng, F.,uQ, Ying, G., Yang, B.,in, Liu, Y., Lai, H., Zhou, G., Chen, J.,un, Zhao, J. L. (2014). Biotransformation of the flame retardant tetrabromobisphenol-a (TBBPA) by freshwater microalgae. Environmental Toxicology and Chemistry 33(8):1705-1711.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Water, 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; *Scenedesmus quadricauda*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2523760

Domain	Metric		Rating	Comments
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment were limited
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22:	Reporting of Data	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: population growth. TBBPA was not detected or only trace amounts in biotransformation experiment

Overall Quality Determination Medium

Study Citation:	Springborn Laboratories, (1988). Acute toxicity of tetrabromobisphenol A to fathead minnow (Pimephales promelas) under flow-through conditions with attachments and cover letter dated 111788.
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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547583

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 100%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using appropriate concurrent negative control groups.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical methods.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure (96 hours) was reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	The number of exposure groups (5) and spacing of exposure levels were adequate but there were not adequate effects for a dose response.
	Metric 12:	Testing at or Below Solubility Limit	Medium	The solvent concentration was appropriate although some precipitate was noted and test concentrations exceeded solubility. The highest measured concentration (5.6 mg/L) exceeds the solubility stated in the final scope (4.1 mg/L).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	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	Number of organisms (initial cell density of2.3 - 3.8x10^4 cells per ml) and number of replicates (3) was adequate.
Domain 5: Outcome Assessment				

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Study Citation:	Springborn Laboratories, (1988). Acute toxicity of tetrabromobisphenol A to fathead minnow (Pimephales promelas) under flow-through conditions with attachments and cover letter dated 111788.
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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547583

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	Medium	A reported precipitate was noted that may influence results.
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups that could influence the outcome assessment. A deviation in protocol was noted on page 70 that the incubation chamber was out of power for ~13 hours. The minimum temperature at this time was 20C and not beloved to be far from the required 24 +/- 1 C to impact algal growth.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described but no effects were noted.
	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 were reported.

Additional Comments: 96 Hour growth for S. capricornutus results are presented on page 68/85 in the narrative and Figure 1 on page 76/85.

Overall Quality Determination High

Study Citation:	Sun, Y., Guo, H., Yu, H., Wang, X., Wu, J., Xue, Y. (2008). Bioaccumulation and physiological effects of tetrabromobisphenol A in coontail Ceratophyllum demersum L. Chemosphere 70(10):1787-1795.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; *Ceratophyllum demersum*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Oxidative stress (including redox biology)-Photosynthesis
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4181234

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#.
	Metric 2:	Test Substance Source	Low	The source was not reported and the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent solvent control group.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	Test media preparation methods were reported. Stock solution was made in acetone. Test solutions were renewed every other day. The final solvent concentration in the test solution or control was not reported and exposure concentrations were not measured.
	Metric 8:	Consistency of Exposure Administration	Low	Exposures appears to be administered consistently across study groups but only few details were provided. It was reported that eight gram fresh weight of C. demersum was exposed to 0.5 mg/l in kinetic experiment but this detail was not provided for the static experiment in section 2.2.2. Methods for weighing plants were not reported, unclear whether all exposures had the same mass of plants. Reporting omissions are likely to have a substantial impact on results.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure (14 day exposure) was reported and 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. 5 exposure groups and a control was used for the 'static' experiment (14 day exposure) and one exposure group was used for the 'kinetic exepriment'. The latter was conducted to examine more closely the biological response at 1, 2, 4, 7 and 14 d.
	Metric 12:	Testing at or Below Solubility Limit	Medium	The solvent concentration was not reported, but the biological response of the solvent control was acceptable.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	There are minor reservations about the source of test organisms. Plants of C. demersum were obtained from "local unpolluted ponds".

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Study Citation:	Sun, Y., Guo, H., Yu, H., Wang, X., Wu, J., Xue, Y. (2008). Bioaccumulation and physiological effects of tetrabromobisphenol A in coontail Ceratophyllum demersum L. Chemosphere 70(10):1787-1795.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; *Ceratophyllum demersum*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Oxidative stress (including redox biology)-Photosynthesis
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4181234

Domain	Metric		Rating	Comments
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	It was reported that eight gram fresh weight of C. demersum was used for the experiments. There were 3 replicates per concentration.
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 methodologies for mechanistic endpoints (free radicals, lipid peroxidation, chlorophyll content, non-enzymatic antioxidants (reduced glutathione (GSH) and oxidized glutathione (GSSG)) and enzymatic antioxidants (superoxide dismutase (SOD) and peroxidase (POD)) reported the intended outcome of interest. Methods were briefly described and appropriate references were cited for additional details.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure (e.g., infection) that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed. Student's t test was conducted. This is not an appropriate test when there are more than 2 treatment groups.
	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 all the mechanistic data (free radicals, lipid peroxidation, chlorophyll content, non-enzymatic antioxidants (reduced glutathione (GSH) and oxidized glutathione (GSSG)) and enzymatic antioxidants (superoxide dismutase (SOD) and peroxidase (POD)) in the study. Exposure concentrations were not verified and the final solvent concentration in the test solution was not reported. Student's t test was conducted, this test is not appropriate when there are more than 2 treatment groups.

Overall Quality Determination Medium

Study Citation:	Sun, Y., Guo, H., Yu, H., Wang, X., Wu, J., Xue, Y. (2008). Bioaccumulation and physiological effects of tetrabromobisphenol A in coontail Ceratophyllum demersum L. Chemosphere 70(10):1787-1795.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; *Ceratophyllum demersum*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4181234

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#.
	Metric 2:	Test Substance Source	Low	The source was not reported and the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent solvent control group.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported (Section 2.3) and adequate.
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	Test media preparation methods were reported. Stock solution was made in acetone. Test solutions were renewed every other day. The final solvent concentration in the test solution or control was not reported and exposure concentrations were not measured.
	Metric 8:	Consistency of Exposure Administration	Medium	Exposures were administered consistently across study groups but only few details were provided. It was reported that eight gram fresh weight of C. demersum was exposed to 0.5 mg/l in 'kinetic' experiment but this detail was not provided for the 'static' experiment in section 2.2.2. Methods for weighing plants were not reported, unclear whether all exposures had the same mass of plants. Reporting omissions are likely to have a substantial impact on results.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured in test media.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure (14 days) was reported and 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.5 exposure groups and a control was used for the 'static' experiment (14 day exposure) and one exposure group was used for the 'kinetic exepriment'. The latter was conducted to examine more closely the biological response at 1, 2, 4, 7 and 14 d.
	Metric 12:	Testing at or Below Solubility Limit	Medium	The solvent concentration was not reported, but the biological response of the solvent control was acceptable.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	There are minor reservations about the source of test organisms. Plants of C. demersum were obtained from "local unpolluted ponds".
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions.

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Study Citation:	Sun, Y., Guo, H., Yu, H., Wang, X., Wu, J., Xue, Y. (2008). Bioaccumulation and physiological effects of tetrabromobisphenol A in coontail Ceratophyllum demersum L. Chemosphere 70(10):1787-1795.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; *Ceratophyllum demersum*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4181234

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Medium	It was reported that eight gram fresh weight of C. demersum was used for the experiments. There were 3 replicates per concentration.
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	Medium	The outcome assessment methodology reported the intended outcome of interest. The analytical methods for determining TBBPA concentration in plant tissue was reported. The weight of plants harvested from each treatment and the conversion calculations from fresh weight to dry weight was not reported.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups in the 'static' and across time intervals in the 'kinetic' experiments.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately. Regression analysis was done for the the kinetic experiment (Fig 1b; section 3:1) but for the static experiment, it was reported that significant differences were found but statistical results were not provided.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment group (Fig 1). Results for control were qualitatively described in the text (section 2.3).
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: This form is for all bioaccumulation experiments of TBBPA in plant tissue. Exposure concentrations were not verified and the final solvent concentration in the test solution was not reported.

Overall Quality Determination Medium

Study Citation:	Agroscience,, E.E., LLC, (2023). TBBPA: A 7-day static-renewal toxicity test with duckweed (Lemna gibba G3).
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; *Lemna gibba*; G3; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	12155422

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 test substance was from Bromine Compounds Ltd. A certificate of analysis was reported in Appendix 2.
	Metric 3:	Test Substance Purity	High	The purity was reported to be >99.0%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of both a negative control and a solvent control.
	Metric 5:	Negative Control Response	High	The negative control responses were reported in Table 7. The solvent control and the negative control data were pooled in these results because there was no significant difference between the negative and solvent control groups.
	Metric 6:	Randomized Allocation	Medium	It was reported that the plants were indiscriminately assigned and transferred from the culture into the test chambers.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The test chambers were 250mL glass beakers with 100mL of test solution. The preparation of the stock solution and the working stocks was described adequately. The test system was a static renewal system.
	Metric 8:	Consistency of Exposure Administration	Medium	Exposures appeared to be administered consistently across study groups. Test chambers, number of organisms, and test conditions were consistent. Old test concentrations appeared to be much lower than newly made concentrations when analyzed. This creates concern regarding exposure consistency between renewals. Results were reported in terms of both mean measured and nominal concentrations.
	Metric 9:	Measurement of Test Substance Concentration	High	Test concentrations were analyzed using LC/MS/MS on days 0, 3, 5, and 7 (test termination). The analytical method is reported in detail in Appendix 5. The measured concentrations are reported in Table 3.
	Metric 10:	Exposure Duration and Frequency	High	The study duration was for seven days, which is typical for a Lemna spp. test.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	There were five test concentrations with a twofold geometric progression, which is in accordance with EPA test guidelines.
	Metric 12:	Testing at or Below Solubility Limit	High	At the adjusted pH used in the test, it appears that the test concentrations would have been below the solubility limit. DMF was also used at a solvent at an appropriate level with an appropriate solvent control response.
Domain 4: Test Organism				

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Study Citation:	Agroscience,, E.E., LLC, (2023). TBBPA: A 7-day static-renewal toxicity test with duckweed (Lemna gibba G3).
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; *Lemna gibba*; G3; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	12155422

Domain	Metric		Rating	Comments
	Metric 13:	Test Organism Characteristics	High	The duckweed was cultured in house at Eurofins. The original culture was reported to be from the United States Department of Agriculture.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The duckweed was reported to be cultured under the same conditions as the test conditions.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	There were four replicates each consisting of four plants to total 12 fronds. This is appropriate according to EPA test guidelines.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	The culture medium used for both testing and culturing was reported to be 20X AAP medium which is described in Appendix 3. The temperature was reported to be 25C. pH and light intensity were also both reported and appropriate.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcomes of interest. The study authors assessed frond number yield and growth rate as well as biomass yield and growth rate.
	Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. Observations of the plants were made on days 3 and 5, and total number of duckweed plants was determined at test termination.
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 analyses were reported in the ”Statistical Analyses” section of the reference and were appropriate for the study.
	Metric 22:	Reporting of Data	High	Growth outcomes were reported in Table 7 in terms of mean frond number yield, frond number growth rate, biomass yield, and biomass growth rate. Data was presented for all exposure groups and the controls. Table 8 contains EC50 values along with NOECs and LOECs.
	Metric 23:	Explanation of Unexpected Outcomes	Low	The data tables did not provide measures of variability.

Additional Comments: This evaluation was for the toxicity of TBBPA to duckweed Lemna gibba G3. This form is for the development/growth outcomes including frond number yield, frond number growth rate, biomass yield, and biomass growth rate. Chlorosis and necrosis could also be included here.

Overall Quality Determination High

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Study Citation:	Agroscience,, E.E., LLC, (2023). TBBPA: A 7-day static-renewal toxicity test with duckweed (Lemna gibba G3).
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; *Lemna gibba*; G3; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	12155422

Domain	Metric	Rating	Comments

Study Citation:	Agroscience,, E.E., LLC, (2023). TBBPA: A 7-day static-renewal toxicity test with duckweed (Lemna gibba G3).
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; *Lemna gibba*; G3; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	12155422

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 test substance was from Bromine Compounds Ltd. A certificate of analysis was reported in Appendix 2.
	Metric 3:	Test Substance Purity	High	The purity was reported to be >99.0%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of both a negative control and a solvent control.
	Metric 5:	Negative Control Response	High	The negative control responses were reported in Table 6 and were appropriate for the outcome of interest.
	Metric 6:	Randomized Allocation	Medium	It was reported that the plants were indiscriminately assigned and transferred from the culture into the test chambers.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The test chambers were 250mL glass beakers with 100mL of test solution. The preparation of the stock solution and the working stocks was described adequately. The test system was a static renewal system.
	Metric 8:	Consistency of Exposure Administration	Medium	Exposures appeared to be administered consistently across study groups. Test chambers, number of organisms, and test conditions were consistent. Old test concentrations appeared to be much lower than newly made concentrations when analyzed. This creates concern regarding exposure consistency between renewals. Results were reported in terms of both mean measured and nominal concentrations.
	Metric 9:	Measurement of Test Substance Concentration	High	Test concentrations were analyzed using LC/MS/MS on days 0, 3, 5, and 7 (test termination). The analytical method is reported in detail in Appendix 5. The measured concentrations are reported in Table 3.
	Metric 10:	Exposure Duration and Frequency	High	The study duration was for seven days, which is typical for a Lemna spp. test.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	There were five test concentrations with a twofold geometric progression, which is in accordance with EPA test guidelines.
	Metric 12:	Testing at or Below Solubility Limit	High	At the adjusted pH used in the test, it appears that the test concentrations would have been below the solubility limit. DMF was also used at a solvent at an appropriate level with an appropriate solvent control response.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The duckweed was cultured in house at Eurofins. The original culture was reported to be from the United States Department of Agriculture.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The duckweed was reported to be cultured under the same conditions as the test conditions.

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Study Citation:	Agroscience,, E.E., LLC, (2023). TBBPA: A 7-day static-renewal toxicity test with duckweed (Lemna gibba G3).
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; *Lemna gibba*; G3; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	12155422

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Medium	There were four replicates each consisting of four plants to total 12 fronds. This is appropriate according to EPA test guidelines.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	The culture medium used for both testing and culturing was reported to be 20X AAP medium which is described in Appendix 3. The temperature was reported to be 25C. pH and light intensity were also both reported and appropriate.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcomes of interest. Study authors assessed whether plants were dead on days 3, 5, and 7.
	Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups. Observations of the plants were made on days 3 and 5, and total number of duckweed plants was determined at test termination.
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	N/A	Statistical analysis was reported in the ”Statistical Analyses” section of the reference and were appropriate for the study. None of the plants died in any of the treatment groups, so statistical analysis was not necessary for this portion of the study.
	Metric 22:	Reporting of Data	High	Mortality data was reported in Table 6 along with data on chlorosis and necrosis for all treatment groups and controls.
	Metric 23:	Explanation of Unexpected Outcomes	Low	The data tables did not provide measures of variability.

Additional Comments: This evaluation was for the toxicity of TBBPA to duckweed Lemna gibba G3. This form is for the mortality outcome presented in Table 6.

Overall Quality Determination High

Study Citation:	Agroscience,, E.E., LLC, (2023). TBBPA: A 7-day static-renewal toxicity test with duckweed (Lemna gibba G3).
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; *Lemna gibba*; G3; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	12155422

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 test substance was from Bromine Compounds Ltd. A certificate of analysis was reported in Appendix 2.
	Metric 3:	Test Substance Purity	High	The purity was reported to be >99.0%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of a negative control.
	Metric 5:	Negative Control Response	High	The negative control response was reported in Table 1 for the Non-GLP range finding test.
	Metric 6:	Randomized Allocation	Low	The allocation method for the range finding test was not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The experimental system for the range finding test was not reported in detail.
	Metric 8:	Consistency of Exposure Administration	Low	Information regarding the range finding test was limited, so it was difficult to determine consistency of administration.
	Metric 9:	Measurement of Test Substance Concentration	Low	The test concentrations for the nominal test did not appear to be analyzed.
	Metric 10:	Exposure Duration and Frequency	High	The study duration was for seven days, which is typical for a Lemna spp. test.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	There were only three exposure groups, but this was a range finding test.
	Metric 12:	Testing at or Below Solubility Limit	Medium	At the adjusted pH used in the test, it appears that the test concentrations would have been below the solubility limit. DMF was used as a solvent for the definitive test, but it is unclear if it was also used for the range finding test, since there were no solvent control results reported in Table 1.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The duckweed was cultured in house at Eurofins. The original culture was reported to be from the United States Department of Agriculture.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The duckweed was reported to be cultured under the same conditions as the test conditions.
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of test organisms and the number of replicates were not reported for the range finding test.
Domain 5: Outcome Assessment				

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Study Citation:	Agroscience,, E.E., LLC, (2023). TBBPA: A 7-day static-renewal toxicity test with duckweed (Lemna gibba G3).
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; *Lemna gibba*; G3; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	12155422

Domain	Metric		Rating	Comments
	Metric 16:	Adequacy of Test Conditions	Medium	The culture medium used for both testing and culturing was reported to be 20X AAP medium which is described in Appendix 3. Test conditions for the range finding test were not reported, though they are likely to be similar to the definitive test.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcomes of interest, which was the mean frond number after seven days.
	Metric 18:	Consistency of Outcome Assessment	Medium	Details of the outcome assessment were limited for the range finding test. Mean frond number was reported, but it is unclear when observations were made.
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	Low	Statistical methods for the range finding test were not reported. Mean frond numbers were reported in Table 1.
	Metric 22:	Reporting of Data	High	Mean frond number for the range finding test was reported in Table 1 for all treatment groups and the negative control.
	Metric 23:	Explanation of Unexpected Outcomes	High	The variability was reported in Table 1, and no other unexpected outcomes were reported.

Additional Comments: This evaluation was for the toxicity of TBBPA to duckweed Lemna gibba G3. This form is for the development/growth outcome reported from the non-GLP range finding test.

Overall Quality Determination Medium

Study Citation:	Debenest, T., Gagné, F., Petit, A. N., André, C., Kohli, M., Blaise, C. (2010). Ecotoxicity of a brominated flame retardant (tetrabromobisphenol A) and its derivatives to aquatic organisms. Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 152(4):407-412.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; *Spinacia oleracea*; photosynthetic enzyme complex test in vitro; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Cytotoxicity-Photosynthesis
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1294788

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%
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Calculation implies that a negative control was used
	Metric 5:	Negative Control Response	Low	The biological response of the negative control groups was not reported.
	Metric 6:	Randomized Allocation	Low	No random allocation reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
	Metric 8:	Consistency of Exposure Administration	Low	details of exposure administration were not provided
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured or measurements were not reported
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
	Metric 12:	Testing at or Below Solubility Limit	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test animals was not reported
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized
	Metric 15:	Number of Organisms and Replicates per Group	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Reported environmental conditions were not sufficient to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed

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Study Citation:	Debenest, T., Gagné, F., Petit, A. N., André, C., Kohli, M., Blaise, C. (2010). Ecotoxicity of a brominated flame retardant (tetrabromobisphenol A) and its derivatives to aquatic organisms. Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 152(4):407-412.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; *Spinacia oleracea*; photosynthetic enzyme complex test in vitro; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Cytotoxicity-Photosynthesis
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1294788

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	Low	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22:	Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	Low	unexpected outcomes were not reported

Additional Comments: This evaluation form is for the in vitro test conducted using spinach photosynthetic enzyme complexes. Photosynthetic efficiency inhibition was assessed.

Overall Quality Determination Low

Study Citation:	Huang, Q., Chen, Y., Lin, L., Liu, Y., Chi, Y., Lin, Y., Ye, G., Zhu, H., Dong, S. (2017). Different effects of bisphenol a and its halogenated derivatives on the reproduction and development of Oryzias melastigma under environmentally relevant doses. Science of the Total Environment 595:752-758.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5499513

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	A solvent control was used in all control as well as treatment groups. "The DMSO concentration was 0.2% in both control and experimental groups."
	Metric 5:	Negative Control Response	Medium	The biological response (hatchling success) of the negative control group was reported, assuming the value in the first column in Table 1 associated with TCBPA is the control group. The exposure to this group was 0 mg/L.
	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 few details on exposure media preparation or test system
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate, 0.2%
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test animals was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				

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Study Citation:	Huang, Q., Chen, Y., Lin, L., Liu, Y., Chi, Y., Lin, Y., Ye, G., Zhu, H., Dong, S. (2017). Different effects of bisphenol a and its halogenated derivatives on the reproduction and development of Oryzias melastigma under environmentally relevant doses. Science of the Total Environment 595:752-758.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5499513

Domain	Metric		Rating	Comments
	Metric 16:	Adequacy of Test Conditions	Medium	Minimal information was reported. "Oryzias melastigma was maintained in 30% artificial seawater under 28 ± 2 °C with 14 h-light/10 h-dark photoperiod. They were fed with live brine shrimp nauplii twice a day." "They were exposed to BPA, TBBPA, and TCBPA at the serial doses of 0.05, 0.2, 0.8, and 1.6 mg/L. Three replicates (n = 15) were performed for each group. Exposure solutions were renewed daily. The number of the newly hatched fish was counted every day. The embryos were recognized dead if they showed no heartbeat and blood circling under microscope (Wu et al. 2012). The newly hatched larvae without embryonic exposure were also subjected to exposure under the same conditions except that the exposure solutions were renewed every two days. Two exposure doses (0.05 and 0.2 mg/L) were adopted. At 21 dpf, six fish from each replicate were randomly sampled for detection of mRNA levels. The remaining fish were continuously exposed for a long-term period of four months. During the long-term exposure, F1 embryos were collected within a period of ten days."
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described. "The software SPSS 16.0 (SPSS Inc., Chicago, IL, USA)was used for statisticalanalysis. Levene's testwas used to determine the homogeneity ofvariance. One-way analysis of variance (ANOVA) followed by a Tukey's test was performed to determine the significance of data. The threshold for the statistical difference was accepted at * $p<0.05$, ** $p<0.01$, *** $p<0.001$."
	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:	Morality based on hatchling success.			

Overall Quality Determination Medium

Study Citation:	Huang, Q., Chen, Y., Lin, L., Liu, Y., Chi, Y., Lin, Y., Ye, G., Zhu, H., Dong, S. (2017). Different effects of bisphenol a and its halogenated derivatives on the reproduction and development of Oryzias melastigma under environmentally relevant doses. Science of the Total Environment 595:752-758.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5499513

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	A solvent control was used in all control as well as treatment groups. "The DMSO concentration was 0.2% in both control and experimental groups."
	Metric 5:	Negative Control Response	Medium	The biological response of the negative control group was reported
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided few details on exposure media preparation or test system
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate, 0.2%
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test animals was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				

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Study Citation:	Huang, Q., Chen, Y., Lin, L., Liu, Y., Chi, Y., Lin, Y., Ye, G., Zhu, H., Dong, S. (2017). Different effects of bisphenol a and its halogenated derivatives on the reproduction and development of Oryzias melastigma under environmentally relevant doses. Science of the Total Environment 595:752-758.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5499513

Domain	Metric		Rating	Comments
	Metric 16:	Adequacy of Test Conditions	Medium	Minimal information was reported. "Oryzias melastigma was maintained in 30% artificial seawater under 28 ± 2 °C with 14 h-light/10 h-dark photoperiod. They were fed with live brine shrimp nauplii twice a day." "They were exposed to BPA, TBBPA, and TCBPA at the serial doses of 0.05, 0.2, 0.8, and 1.6 mg/L. Three replicates (n = 15) were performed for each group. Exposure solutions were renewed daily. The number of the newly hatched fish was counted every day. The embryos were recognized dead if they showed no heartbeat and blood circling under microscope (Wu et al. 2012). The newly hatched larvae without embryonic exposure were also subjected to exposure under the same conditions except that the exposure solutions were renewed every two days. Two exposure doses (0.05 and 0.2 mg/L) were adopted. At 21 dpf, six fish from each replicate were randomly sampled for detection of mRNA levels. The remaining fish were continuously exposed for a long-term period of four months. During the long-term exposure, F1 embryos were collected within a period of ten days."
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described. "The software SPSS 16.0 (SPSS Inc., Chicago, IL, USA)was used for statisticalanalysis. Levene's testwas used to determine the homogeneity ofvariance. One-way analysis of variance (ANOVA) followed by a Tukey's test was performed to determine the significance of data. The threshold for the statistical difference was accepted at * $p<0.05$, ** $p<0.01$, *** $p<0.001$."
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: None

Overall Quality Determination Medium

Study Citation:	Huang, Q., Chen, Y., Lin, L., Liu, Y., Chi, Y., Lin, Y., Ye, G., Zhu, H., Dong, S. (2017). Different effects of bisphenol a and its halogenated derivatives on the reproduction and development of Oryzias melastigma under environmentally relevant doses. Science of the Total Environment 595:752-758.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5499513

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	A solvent control was used in all control as well as treatment groups. "The DMSO concentration was 0.2% in both control and experimental groups."
	Metric 5:	Negative Control Response	Medium	The biological response of the negative control group was reported
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided few details on exposure media preparation or test system
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate, 0.2%
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test animals was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				

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Study Citation:	Huang, Q., Chen, Y., Lin, L., Liu, Y., Chi, Y., Lin, Y., Ye, G., Zhu, H., Dong, S. (2017). Different effects of bisphenol a and its halogenated derivatives on the reproduction and development of Oryzias melastigma under environmentally relevant doses. Science of the Total Environment 595:752-758.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5499513

Domain	Metric		Rating	Comments
	Metric 16:	Adequacy of Test Conditions	Medium	Minimal information was reported. "Oryzias melastigma was maintained in 30% artificial seawater under 28 ± 2 °C with 14 h-light/10 h-dark photoperiod. They were fed with live brine shrimp nauplii twice a day." "They were exposed to BPA, TBBPA, and TCBPA at the serial doses of 0.05, 0.2, 0.8, and 1.6 mg/L. Three replicates (n = 15) were performed for each group. Exposure solutions were renewed daily. The number of the newly hatched fish was counted every day. The embryos were recognized dead if they showed no heartbeat and blood circling under microscope (Wu et al. 2012). The newly hatched larvae without embryonic exposure were also subjected to exposure under the same conditions except that the exposure solutions were renewed every two days. Two exposure doses (0.05 and 0.2 mg/L) were adopted. At 21 dpf, six fish from each replicate were randomly sampled for detection of mRNA levels. The remaining fish were continuously exposed for a long-term period of four months. During the long-term exposure, F1 embryos were collected within a period of ten days."
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described. "The software SPSS 16.0 (SPSS Inc., Chicago, IL, USA)was used for statisticalanalysis. Levene's testwas used to determine the homogeneity ofvariance. One-way analysis of variance (ANOVA) followed by a Tukey's test was performed to determine the significance of data. The threshold for the statistical difference was accepted at * $p<0.05$, ** $p<0.01$, *** $p<0.001$."
	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: hatching enzymes

Overall Quality Determination Medium

Study Citation:	Huang, Q., Chen, Y., Lin, L., Liu, Y., Chi, Y., Lin, Y., Ye, G., Zhu, H., Dong, S. (2017). Different effects of bisphenol a and its halogenated derivatives on the reproduction and development of Oryzias melastigma under environmentally relevant doses. Science of the Total Environment 595:752-758.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Embryo
Health Outcome:	Cardiovascular
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5499513

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	A solvent control was used in all control as well as treatment groups. "The DMSO concentration was 0.2% in both control and experimental groups."
	Metric 5:	Negative Control Response	Low	The biological response of the 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 few details on exposure media preparation or test system
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate, 0.2%
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test animals was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				

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Study Citation:	Huang, Q., Chen, Y., Lin, L., Liu, Y., Chi, Y., Lin, Y., Ye, G., Zhu, H., Dong, S. (2017). Different effects of bisphenol a and its halogenated derivatives on the reproduction and development of Oryzias melastigma under environmentally relevant doses. Science of the Total Environment 595:752-758.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Embryo
Health Outcome:	Cardiovascular
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5499513

Domain	Metric		Rating	Comments
	Metric 16:	Adequacy of Test Conditions	Medium	Minimal information was reported. "Oryzias melastigma was maintained in 30% artificial seawater under 28 ± 2 °C with 14 h-light/10 h-dark photoperiod. They were fed with live brine shrimp nauplii twice a day." "They were exposed to BPA, TBBPA, and TCBPA at the serial doses of 0.05, 0.2, 0.8, and 1.6 mg/L. Three replicates (n = 15) were performed for each group. Exposure solutions were renewed daily. The number of the newly hatched fish was counted every day. The embryos were recognized dead if they showed no heartbeat and blood circling under microscope (Wu et al. 2012). The newly hatched larvae without embryonic exposure were also subjected to exposure under the same conditions except that the exposure solutions were renewed every two days. Two exposure doses (0.05 and 0.2 mg/L) were adopted. At 21 dpf, six fish from each replicate were randomly sampled for detection of mRNA levels. The remaining fish were continuously exposed for a long-term period of four months. During the long-term exposure, F1 embryos were collected within a period of ten days."
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described. "The software SPSS 16.0 (SPSS Inc., Chicago, IL, USA)was used for statisticalanalysis. Levene's testwas used to determine the homogeneity ofvariance. One-way analysis of variance (ANOVA) followed by a Tukey's test was performed to determine the significance of data. The threshold for the statistical difference was accepted at * $p<0.05$, ** $p<0.01$, *** $p<0.001$."
	Metric 22:	Reporting of Data	Medium	Heart rate results were shown in Figure 1D. Control data was not included.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes and measures of variability were included in the figure.

Additional Comments: This evaluation is for the heart rate endpoint.

Overall Quality Determination Medium

Study Citation:	Huang, Q., Chen, Y., Lin, L., Liu, Y., Chi, Y., Lin, Y., Ye, G., Zhu, H., Dong, S. (2017). Different effects of bisphenol a and its halogenated derivatives on the reproduction and development of Oryzias melastigma under environmentally relevant doses. Science of the Total Environment 595:752-758.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Larvae
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5499513

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	A solvent control was used in all control as well as treatment groups. "The DMSO concentration was 0.2% in both control and experimental groups."
	Metric 5:	Negative Control Response	Medium	The biological response of the negative control group was reported
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided few details on exposure media preparation or test system
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate, 0.2%
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test animals was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				

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Study Citation:	Huang, Q., Chen, Y., Lin, L., Liu, Y., Chi, Y., Lin, Y., Ye, G., Zhu, H., Dong, S. (2017). Different effects of bisphenol a and its halogenated derivatives on the reproduction and development of Oryzias melastigma under environmentally relevant doses. Science of the Total Environment 595:752-758.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Larvae
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5499513

Domain	Metric		Rating	Comments
	Metric 16:	Adequacy of Test Conditions	Medium	Minimal information was reported. "Oryzias melastigma was maintained in 30% artificial seawater under 28 ± 2 °C with 14 h-light/10 h-dark photoperiod. They were fed with live brine shrimp nauplii twice a day." "They were exposed to BPA, TBBPA, and TCBPA at the serial doses of 0.05, 0.2, 0.8, and 1.6 mg/L. Three replicates (n = 15) were performed for each group. Exposure solutions were renewed daily. The number of the newly hatched fish was counted every day. The embryos were recognized dead if they showed no heartbeat and blood circling under microscope (Wu et al. 2012). The newly hatched larvae without embryonic exposure were also subjected to exposure under the same conditions except that the exposure solutions were renewed every two days. Two exposure doses (0.05 and 0.2 mg/L) were adopted. At 21 dpf, six fish from each replicate were randomly sampled for detection of mRNA levels. The remaining fish were continuously exposed for a long-term period of four months. During the long-term exposure, F1 embryos were collected within a period of ten days."
	Metric 17:	Outcome Assessment Methodology	Low	It was not described in the methods how body weight was determined.
	Metric 18:	Consistency of Outcome Assessment	Low	Since there were limited details, it is unclear if methods were consistent across treatment groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described. "The software SPSS 16.0 (SPSS Inc., Chicago, IL, USA)was used for statisticalanalysis. Levene's testwas used to determine the homogeneity ofvariance. One-way analysis of variance (ANOVA) followed by a Tukey's test was performed to determine the significance of data. The threshold for the statistical difference was accepted at * $p<0.05$, ** $p<0.01$, *** $p<0.001$."
	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 body weight determination after a four-month exposure.

Overall Quality Determination Medium

Study Citation:	Huang, Q., Chen, Y., Lin, L., Liu, Y., Chi, Y., Lin, Y., Ye, G., Zhu, H., Dong, S. (2017). Different effects of bisphenol a and its halogenated derivatives on the reproduction and development of Oryzias melastigma under environmentally relevant doses. Science of the Total Environment 595:752-758.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Larvae
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5499513

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	A solvent control was used in all control as well as treatment groups. "The DMSO concentration was 0.2% in both control and experimental groups."
	Metric 5:	Negative Control Response	Medium	The biological response of the negative control group was reported
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided few details on exposure media preparation or test system
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate, 0.2%
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test animals was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				

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Study Citation:	Huang, Q., Chen, Y., Lin, L., Liu, Y., Chi, Y., Lin, Y., Ye, G., Zhu, H., Dong, S. (2017). Different effects of bisphenol a and its halogenated derivatives on the reproduction and development of Oryzias melastigma under environmentally relevant doses. Science of the Total Environment 595:752-758.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Larvae
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5499513

Domain	Metric		Rating	Comments
	Metric 16:	Adequacy of Test Conditions	Medium	Minimal information was reported. "Oryzias melastigma was maintained in 30% artificial seawater under 28 ± 2 °C with 14 h-light/10 h-dark photoperiod. They were fed with live brine shrimp nauplii twice a day." "They were exposed to BPA, TBBPA, and TCBPA at the serial doses of 0.05, 0.2, 0.8, and 1.6 mg/L. Three replicates (n = 15) were performed for each group. Exposure solutions were renewed daily. The number of the newly hatched fish was counted every day. The embryos were recognized dead if they showed no heartbeat and blood circling under microscope (Wu et al. 2012). The newly hatched larvae without embryonic exposure were also subjected to exposure under the same conditions except that the exposure solutions were renewed every two days. Two exposure doses (0.05 and 0.2 mg/L) were adopted. At 21 dpf, six fish from each replicate were randomly sampled for detection of mRNA levels. The remaining fish were continuously exposed for a long-term period of four months. During the long-term exposure, F1 embryos were collected within a period of ten days."
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	N/A	relative expression levels reported. "The relative fold changes of the tested genes were analyzed by the 2- ΔΔCt method. Ribosomal protein L7 (RPL7) was used as the reference gene (Livak and Schmittgen 2001). The relative expression data were presented as heatmap using the MEV software."
	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: "Heatmap of gene expression upon BPA and H-BPAs exposure at various developmental stages in Oryzias melastigma." Larval gene expression was evaluated with ERα, ChgH, and VTG2 genes.

Overall Quality Determination Medium

Study Citation:	Huang, Q., Chen, Y., Lin, L., Liu, Y., Chi, Y., Lin, Y., Ye, G., Zhu, H., Dong, S. (2017). Different effects of bisphenol a and its halogenated derivatives on the reproduction and development of Oryzias melastigma under environmentally relevant doses. Science of the Total Environment 595:752-758.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5499513

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	A solvent control was used in all control as well as treatment groups. "The DMSO concentration was 0.2% in both control and experimental groups."
	Metric 5:	Negative Control Response	Medium	The biological response of the negative control group was reported
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided few details on exposure media preparation or test system
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate, 0.2%
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test animals was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				

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Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5499513

Domain	Metric		Rating	Comments
	Metric 16:	Adequacy of Test Conditions	Low	Minimal information was reported. "Oryzias melastigma was maintained in 30% artificial seawater under 28 ± 2 °C with 14 h-light/10 h-dark photoperiod. They were fed with live brine shrimp nauplii twice a day." "They were exposed to BPA, TBBPA, and TCBPA at the serial doses of 0.05, 0.2, 0.8, and 1.6 mg/L. Three replicates (n = 15) were performed for each group. Exposure solutions were renewed daily. The number of the newly hatched fish was counted every day. The embryos were recognized dead if they showed no heartbeat and blood circling under microscope (Wu et al. 2012). The newly hatched larvae without embryonic exposure were also subjected to exposure under the same conditions except that the exposure solutions were renewed every two days. Two exposure doses (0.05 and 0.2 mg/L) were adopted. At 21 dpf, six fish from each replicate were randomly sampled for detection of mRNA levels. The remaining fish were continuously exposed for a long-term period of four months. During the long-term exposure, F1 embryos were collected within a period of ten days."
	Metric 17:	Outcome Assessment Methodology	Medium	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	N/A	relative expression levels reported. "The relative fold changes of the tested genes were analyzed by the 2- ΔΔCt method. Ribosomal protein L7 (RPL7) was used as the reference gene (Livak and Schmittgen 2001). The relative expression data were presented as heatmap using the MEV software."
	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: "Heatmap of gene expression upon BPA and H-BPAs exposure at various developmental stages in Oryzias melastigma." Embryo gene expression was evaluated with ERα, ChgH, and VTG2 genes.

Overall Quality Determination Medium

Study Citation:	Huang, Q., Chen, Y., Lin, L., Liu, Y., Chi, Y., Lin, Y., Ye, G., Zhu, H., Dong, S. (2017). Different effects of bisphenol a and its halogenated derivatives on the reproduction and development of Oryzias melastigma under environmentally relevant doses. Science of the Total Environment 595:752-758.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5499513

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	A solvent control was used in all control as well as treatment groups. "The DMSO concentration was 0.2% in both control and experimental groups."
	Metric 5:	Negative Control Response	Medium	The biological response of the negative control group was reported
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided few details on exposure media preparation or test system
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate, 0.2%
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test animals was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				

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Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5499513

Domain	Metric		Rating	Comments
	Metric 16:	Adequacy of Test Conditions	Medium	Minimal information was reported. "Oryzias melastigma was maintained in 30% artificial seawater under 28 ± 2 °C with 14 h-light/10 h-dark photoperiod. They were fed with live brine shrimp nauplii twice a day." "They were exposed to BPA, TBBPA, and TCBPA at the serial doses of 0.05, 0.2, 0.8, and 1.6 mg/L. Three replicates (n = 15) were performed for each group. Exposure solutions were renewed daily. The number of the newly hatched fish was counted every day. The embryos were recognized dead if they showed no heartbeat and blood circling under microscope (Wu et al. 2012). The newly hatched larvae without embryonic exposure were also subjected to exposure under the same conditions except that the exposure solutions were renewed every two days. Two exposure doses (0.05 and 0.2 mg/L) were adopted. At 21 dpf, six fish from each replicate were randomly sampled for detection of mRNA levels. The remaining fish were continuously exposed for a long-term period of four months. During the long-term exposure, F1 embryos were collected within a period of ten days."
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described. "The software SPSS 16.0 (SPSS Inc., Chicago, IL, USA)was used for statisticalanalysis. Levene's testwas used to determine the homogeneity ofvariance. One-way analysis of variance (ANOVA) followed by a Tukey's test was performed to determine the significance of data. The threshold for the statistical difference was accepted at * $p<0.05$, ** $p<0.01$, *** $p<0.001$."
	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: "Heatmap of gene expression upon BPA and H-BPAs exposure at various developmental stages in Oryzias melastigma." Embryo gene expression was evaluated with ERα, ChgH, and VTG2 genes.

Overall Quality Determination Medium

Study Citation:	Huang, Q., Chen, Y., Lin, L., Liu, Y., Chi, Y., Lin, Y., Ye, G., Zhu, H., Dong, S. (2017). Different effects of bisphenol a and its halogenated derivatives on the reproduction and development of Oryzias melastigma under environmentally relevant doses. Science of the Total Environment 595:752-758.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Larvae
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5499513

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	A solvent control was used in all control as well as treatment groups. "The DMSO concentration was 0.2% in both control and experimental groups."
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided few details on exposure media preparation or test system
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate, 0.2%
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test animals was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				

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Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Larvae
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5499513

Domain	Metric		Rating	Comments
	Metric 16:	Adequacy of Test Conditions	Medium	Minimal information was reported. "Oryzias melastigma was maintained in 30% artificial seawater under 28 ± 2 °C with 14 h-light/10 h-dark photoperiod. They were fed with live brine shrimp nauplii twice a day." "They were exposed to BPA, TBBPA, and TCBPA at the serial doses of 0.05, 0.2, 0.8, and 1.6 mg/L. Three replicates (n = 15) were performed for each group. Exposure solutions were renewed daily. The number of the newly hatched fish was counted every day. The embryos were recognized dead if they showed no heartbeat and blood circling under microscope (Wu et al. 2012). The newly hatched larvae without embryonic exposure were also subjected to exposure under the same conditions except that the exposure solutions were renewed every two days. Two exposure doses (0.05 and 0.2 mg/L) were adopted. At 21 dpf, six fish from each replicate were randomly sampled for detection of mRNA levels. The remaining fish were continuously exposed for a long-term period of four months. During the long-term exposure, F1 embryos were collected within a period of ten days."
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical analysis methods were described in section 2.7.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group in Table 2.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes and measures of variability were included in Table 2.

Additional Comments: This evaluation is for the assessment of sex organs.

Overall Quality Determination Medium

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Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Larvae
Health Outcome:	Mechanistic-Endocrine toxicity
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5499513

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	A solvent control was used in all control as well as treatment groups. "The DMSO concentration was 0.2% in both control and experimental groups."
	Metric 5:	Negative Control Response	Medium	The biological response of the negative control group was reported
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided few details on exposure media preparation or test system
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate, 0.2%
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test animals was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				

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Duration:	Overall Duration: $>$ 21 days; Exposure Duration: $>$ 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Larvae
Health Outcome:	Mechanistic-Endocrine toxicity
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5499513

Domain	Metric		Rating	Comments
	Metric 16:	Adequacy of Test Conditions	Medium	Minimal information was reported. "Oryzias melastigma was maintained in 30% artificial seawater under 28 $\pm$ 2 °C with 14 h-light/10 h-dark photoperiod. They were fed with live brine shrimp nauplii twice a day." "They were exposed to BPA, TBBPA, and TCBPA at the serial doses of 0.05, 0.2, 0.8, and 1.6 mg/L. Three replicates (n = 15) were performed for each group. Exposure solutions were renewed daily. The number of the newly hatched fish was counted every day. The embryos were recognized dead if they showed no heartbeat and blood circling under microscope (Wu et al. 2012). The newly hatched larvae without embryonic exposure were also subjected to exposure under the same conditions except that the exposure solutions were renewed every two days. Two exposure doses (0.05 and 0.2 mg/L) were adopted. At 21 dpf, six fish from each replicate were randomly sampled for detection of mRNA levels. The remaining fish were continuously exposed for a long-term period of four months. During the long-term exposure, F1 embryos were collected within a period of ten days."
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	The statistical analysis methods were described in section 2.7.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group in Figure 3.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes and measures of variability were shown in Figure 3.

Additional Comments: This evaluation is for the assessment of 17-beta estradiol and testosterone.

Overall Quality Determination Medium

Study Citation:	Gong, W., Zhu, L., Hao, Y. (2016). Lethal and sublethal toxicity comparison of BFRs to three marine planktonic copepods: Effects on survival, metabolism and ingestion. PLoS ONE 11(1):e0147790.
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; *Acartia pacifica*; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3350491

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 >95%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable.
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations. Concentrations of test substance were not measured during the study.
	Metric 8:	Consistency of Exposure Administration	Medium	Reporting omissions are likely to have a substantial impact on results. Unclear whether exposure administration was consistent across study groups and controls. The number of exposure groups and exposure concentrations were not reported.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	The number of exposure groups and spacing of exposure levels were not given in the methods other than "The concentration gradients were set up according to a certain common ratio with the mortality less than 10% in the lowest dose and greater than 90% in the highest." Fig 1 shows the concentrations tested but is not very clear.
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	There were significant concerns regarding the source of test organisms. Copepods were collected locally from Qingdao Li-cun River estuary, Cheng-yang District He-tao shrimp ponds, and Laoshan District Nanyao shrimp ponds. It was reported that adult copepods were used for testing but unclear whether copepods used for testing were of the same size.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.

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Study Citation:	Gong, W., Zhu, L., Hao, Y. (2016). Lethal and sublethal toxicity comparison of BFRs to three marine planktonic copepods: Effects on survival, metabolism and ingestion. PLoS ONE 11(1):e0147790.
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; *Acartia pacifica*; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3350491

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms (15 per test chamber) and replicates (3 replicates per concentration) were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reported environmental conditions were not sufficient to evaluate if adequate and whether differences occurred between control and exposed populations.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical methods for determining an LC50 were not reported.
	Metric 22:	Reporting of Data	High	Mean values for exposure-related findings were presented for each treatment and control group in Figure 1 but not clear.
	Metric 23:	Explanation of Unexpected Outcomes	Low	LC 50 was reported without confidence intervals and insufficient information was provided to determine if excessive variability or unexpected outcomes occurred.

Additional Comments: This evaluation form is relevant to the mortality outcome for Acartia pacifica. The reported 96 h-LC50 is 0.04 mg/L of TBBPA.

Overall Quality Determination Medium

Study Citation:	Gong, W., Zhu, L., Hao, Y. (2016). Lethal and sublethal toxicity comparison of BFRs to three marine planktonic copepods: Effects on survival, metabolism and ingestion. PLoS ONE 11(1):e0147790.
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; *Acartia pacifica*; Adult
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3350491

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 >95%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations. Concentrations of test substance were not measured during the study.
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported (48 hr) and 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 reported as 1.25, 2.5, 5, 10 and 20% of 96h-LC50, and were adequate to address the purpose of the study.
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate ((i.e., no effects on biological responses were observed in the solvent control and no interactions were expected between the solvent and test substance).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	There were significant concerns regarding the source of test organisms. There were significant concerns regarding the source of test organisms. Copepods were collected locally from Qingdao Li-cun River estuary, Cheng-yang District He-tao shrimp ponds, and Laoshan District Nanyao shrimp ponds."A 2000 mL beaker was cultured in advance and the copepods were collected at the same time to assure developments were closely synchronized" No details regarding the duration of culture or copepod size were give.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed groups.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms (30) were suitable for a dose response. The number of replicates (3 per concentration) were reported.

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Study Citation:	Gong, W., Zhu, L., Hao, Y. (2016). Lethal and sublethal toxicity comparison of BFRs to three marine planktonic copepods: Effects on survival, metabolism and ingestion. PLoS ONE 11(1):e0147790.
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; *Acartia pacifica*; Adult
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3350491

Domain	Metric		Rating	Comments
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reported environmental conditions were not sufficient to evaluate if adequate and whether differences occurred between control and exposed populations.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology to determine ingestion and filtration rates were reported in detail.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described.
	Metric 22:	Reporting of Data	High	Mean values for exposure-related findings were presented for each treatment and control group (Figure 3).
	Metric 23:	Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.

Additional Comments: This evaluation form is for the ingestion and filtrations rates of Acartia pacifica following exposure to TBBPA. Ingestion and filtration rates are grouped under behavioral outcome to be consistent with ECOTOX output for this study.

Overall Quality Determination Medium

Study Citation:	Gong, W., Zhu, L., Hao, Y. (2016). Lethal and sublethal toxicity comparison of BFRs to three marine planktonic copepods: Effects on survival, metabolism and ingestion. PLoS ONE 11(1):e0147790.
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; *Acartia pacifica*; Adult
Health Outcome:	Nutritional & Metabolic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3350491

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 >95%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group. However, there was conflicting information regarding the type of control. In the methods section, it was reported that a solvent control was used but in the results section, it was given that untreated seawater was used as the control.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations. Concentrations of test substance were not measured during the study.
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response. Concentrations tested were 1.25, 2.5, 5, 10 and 20% of 96h-LC50 .
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	There were significant concerns regarding the source of test organisms. Copepods were collected locally from Qingdao Li-cun River estuary, Cheng-yang District He-tao shrimp ponds, and Laoshan District Nanyao shrimp ponds.
	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 (30) were suitable for a dose response. The number of replicates (3 per concentration) were reported.
Domain 5: Outcome Assessment				

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Study Citation:	Gong, W., Zhu, L., Hao, Y. (2016). Lethal and sublethal toxicity comparison of BFRs to three marine planktonic copepods: Effects on survival, metabolism and ingestion. PLoS ONE 11(1):e0147790.
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; *Acartia pacifica*; Adult
Health Outcome:	Nutritional & Metabolic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3350491

Domain	Metric		Rating	Comments
	Metric 16:	Adequacy of Test Conditions	Low	Reported environmental conditions were not sufficient to evaluate if adequate and whether differences occurred between control and exposed populations.
	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 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.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described.
	Metric 22:	Reporting of Data	High	Mean values 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 evaluation form is for the oxygen consumption and ammonia excretion rates of Acartia pacifica following exposure to TBBPA.

Overall Quality Determination Medium

Study Citation:	Wollenberger, L., Dinan, L., Breitholtz, M. (2005). Brominated flame retardants: Activities in a crustacean development test and in an ecdysteroid screening assay. Environmental Toxicology and Chemistry 24(2):400-407.
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; *Acartia tonsa*; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2819235

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Medium	Nomenclature (3,3' ,5,5' tetrabromobisphenol A), chemical formula, and molecular weight are reported, no CASRN is reported.
	Metric 2:	Test Substance Source	Low	Source was reported but not analytically verified.
	Metric 3:	Test Substance Purity	Low	Sigma Aldrich was reported as source but no grade or purity was reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Acetone was used as a carrier and solvent controls were used.
	Metric 5:	Negative Control Response	High	Authors reported "no control mortality" in the results section.
	Metric 6:	Randomized Allocation	Low	Random allocation was not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The acetone stock solutions and dilution methods were detailed, but the authors used an ultrasonic bath for preparation and did not detail the duration of the bath. The final concentration of the acetone in treatment concentrations were reported (100 uL/L). The manufacturer source of synthetic seawater (18 ppt) and it's composition was not reported.
	Metric 8:	Consistency of Exposure Administration	High	The treatments were administered consistently. Final solvent (acetone) concentration was reported (100 uL/L).
	Metric 9:	Measurement of Test Substance Concentration	Low	Although some compounds (BDE-28, BDE-99, BDE-100) were tested, TBBPA concentrations were not verified and are reported as nominal throughout the study.
	Metric 10:	Exposure Duration and Frequency	High	A 48 h duration for acute toxicity was an appropriate duration.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Authors reported using 6 treatment concentrations and a control.
	Metric 12:	Testing at or Below Solubility Limit	High	Table 3 (page 4/8) reports the range of the 6 treatment concentrations in the acute bioassay. Concentrations range from 100 to 500 ug/L and are under the 4.1 mg/L solubility reported in the Final Scope.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The source of copepods was not reported. Age was reported but distribution of sex within the bioassay was not reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The culture conditions prior to the bioassay were reported and are the same conditions for the study. They used 12-14 day old copepods for the acute bioassay.

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Study Citation:	Wollenberger, L., Dinan, L., Breitholtz, M. (2005). Brominated flame retardants: Activities in a crustacean development test and in an ecdysteroid screening assay. Environmental Toxicology and Chemistry 24(2):400-407.
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; *Acartia tonsa*; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2819235

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Authors reported using 5 reps per treatment concentration and 5 animals within each rep.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Food source, feeding protocol and temp/light conditions were reported. Water quality parameters past salinity concentration were not reported.
	Metric 17:	Outcome Assessment Methodology	High	Criteria for mortality assessment was defined as immobility/lack of response to physical touch.
	Metric 18:	Consistency of Outcome Assessment	High	Assessments were consistent among treatments and control groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Medium	No differences among treatment and control groups were attributed to environmental conditions, however, environmental conditions past water temp, salinity, and light cycle were not reported.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	Nothing was reported to indicate differences in groups based on animal attrition or health outcomes.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Probit analysis was used for dose-response estimation. (Swedish Environmental Protection Board. 1990. Probit Analysis, Ver 2.3. Stockholm, Sweden.)
	Metric 22:	Reporting of Data	High	Authors did not report results for each concentration, but did present LC10 and LC50 with 95% CIs. No control mortality occurred and is reported in the narrative of the results section on page 4/8.
	Metric 23:	Explanation of Unexpected Outcomes	High	No unexpected outcomes were reported.

Additional Comments: 48-hr Acute toxicity data for TBBPA are presented in table 3 withL range tested, 48-hr LC10 and LC50 (95% CIs).

Overall Quality Determination High

Study Citation:	Wollenberger, L., Dinan, L., Breitholtz, M. (2005). Brominated flame retardants: Activities in a crustacean development test and in an ecdysteroid screening assay. Environmental Toxicology and Chemistry 24(2):400-407.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; *Acartia tonsa*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2819235

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Medium	Nomenclature (3,3' ,5,5' tetrabromobisphenol A), chemical formula, and molecular weight are reported, no CASRN is reported.
	Metric 2:	Test Substance Source	Low	Source was reported but not analytically verified.
	Metric 3:	Test Substance Purity	Low	Sigma Aldrich was reported as source but no grade or purity was reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Acetone was used as a carrier and solvent controls were used.
	Metric 5:	Negative Control Response	High	The control larval developmental rate is presented in table 4 on page 5/8 and uses this rate as the benchmark for developmental rate among the treatment concentrations.
	Metric 6:	Randomized Allocation	Low	Random allocation was not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The acetone stock solutions and dilution methods were detailed, but the authors used an ultrasonic bath for preparation and did not detail the duration of the bath. The final concentration of the acetone in treatment concentrations were reported (100 uL/L). The manufacturer source of synthetic seawater (18 ppt) and it's composition was not reported.For the developmental exposure, the authors reported renewing the test solutions on day 3 of 5.
	Metric 8:	Consistency of Exposure Administration	High	The treatments were administered consistently. Final solvent (acetone) concentration was reported (100 uL/L).
	Metric 9:	Measurement of Test Substance Concentration	Low	TBBPA concentrations were not verified and are reported as nominal throughout the study.
	Metric 10:	Exposure Duration and Frequency	High	Authors used a 5 day exposure period as they indicated is the period where (at 20C) approximately 50% of the organisms reach a copepodite stage.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Authors reported using 6 treatment concentrations and a control (Table 4).
	Metric 12:	Testing at or Below Solubility Limit	High	Table 4 (page 5/8) reports the range of the 6 treatment concentrations in the bioassay. Concentrations range from 2 to 200 ug/L and are under the 4.1 mg/L solubility reported in the Final Scope.
Domain 4: Test Organism				

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Study Citation:	Wollenberger, L., Dinan, L., Breitholtz, M. (2005). Brominated flame retardants: Activities in a crustacean development test and in an ecdysteroid screening assay. Environmental Toxicology and Chemistry 24(2):400-407.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; *Acartia tonsa*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2819235

Domain	Metric		Rating	Comments
	Metric 13:	Test Organism Characteristics	Medium	The source of copepods was not reported. The authors counted eggs within treatment groups and detailed copepod culture in the first part of the materials and methods section.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The culture conditions prior to the bioassay were reported and are the same conditions for the study. They used distributed eggs into treatment and control groups.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Authors reported using 4 reps per treatment concentration and 30-40 animals within each rep. The control group was represented by 8 replicates.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Food source, feeding protocol and temp/light conditions were reported. Water quality parameters past salinity concentration were not reported.
	Metric 17:	Outcome Assessment Methodology	High	Criteria for larval development rate was detailed in the top of the first column on page 3/8.
	Metric 18:	Consistency of Outcome Assessment	High	Assessments were consistent among treatments and control groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Medium	No differences among treatment and control groups were attributed to environmental conditions, however, environmental conditions past water temp, salinity, and light cycle were not reported.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	Nothing was reported to indicate differences in groups based on animal attrition or health outcomes.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Probit analysis was used for dose-response estimation. (Swedish Environmental Protection Board. 1990. Probit Analysis, Ver 2.3. Stockholm, Sweden.)
	Metric 22:	Reporting of Data	High	Treatment group developmental rates compared to control replicates are presented for each concentration on page 5/8 in Table 4. The 5-d EC10 and EC50 for these data are presented in Table 5 with 95% CIs.
	Metric 23:	Explanation of Unexpected Outcomes	High	No unexpected outcomes were reported.

Additional Comments: 5d larval developmental rate EC10 and EC50 (95% CI) are presented in table 5 on page 5/8.

Overall Quality Determination High

Study Citation:	Wollenberger, L., Dinan, L., Breitholtz, M. (2005). Brominated flame retardants: Activities in a crustacean development test and in an ecdysteroid screening assay. Environmental Toxicology and Chemistry 24(2):400-407.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; *Acartia tonsa*; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2819235

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Medium	Nomenclature (3,3' ,5,5' tetrabromobisphenol A), chemical formula, and molecular weight are reported, no CASRN is reported.
	Metric 2:	Test Substance Source	Low	Source was reported but not analytically verified.
	Metric 3:	Test Substance Purity	Low	Sigma Aldrich was reported as source but no grade or purity was reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Acetone was used as a carrier and solvent controls were used.
	Metric 5:	Negative Control Response	Medium	The narrative reports that all chemicals had a egg hatch among controls from 80-90%. Larval survival was >80%. Specific control hatch and 5-day larval survival are not reported.
	Metric 6:	Randomized Allocation	Low	Random allocation was not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The acetone stock solutions and dilution methods were detailed, but the authors used an ultrasonic bath for preparation and did not detail the duration of the bath. The final concentration of the acetone in treatment concentrations were reported (100 uL/L). The manufacturer source of synthetic seawater (18 ppt) and it's composition was not reported.For the developmental exposure (and egg hatch and larval survival counts), the authors reported renewing the test solutions on day 3 of 5.
	Metric 8:	Consistency of Exposure Administration	High	The treatments were administered consistently. Final solvent (acetone) concentration was reported (100 uL/L).
	Metric 9:	Measurement of Test Substance Concentration	Low	TBBPA concentrations were not verified and are reported as nominal throughout the study.
	Metric 10:	Exposure Duration and Frequency	High	Authors used a 5 day exposure period as they indicated is the period where (at 20C) approximately 50% of the organisms reach a copepodite stage. The numbers of eggs to hatch and number of surviving larvae were counted for each treatment concentrations.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Authors reported using 6 treatment concentrations and a control (Table 4).
	Metric 12:	Testing at or Below Solubility Limit	High	Table 4 (page 5/8) reports the range of the 6 treatment concentrations in the acute bioassay. Concentrations range from 2 to 200 ug/L and are under the 4.1 mg/L solubility reported in the Final Scope.
Domain 4: Test Organism				

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Study Citation:	Wollenberger, L., Dinan, L., Breitholtz, M. (2005). Brominated flame retardants: Activities in a crustacean development test and in an ecdysteroid screening assay. Environmental Toxicology and Chemistry 24(2):400-407.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; *Acartia tonsa*; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2819235

Domain	Metric		Rating	Comments
	Metric 13:	Test Organism Characteristics	Medium	The source of copepods was not reported. The authors counted eggs within treatment groups and detailed copepod culture in the first part of the materials and methods section.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The culture conditions prior to the bioassay were reported and are the same conditions for the study. They used distributed eggs into treatment and control groups.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Authors reported using 4 reps per treatment concentration and 30-40 animals within each rep. The control group was represented by 8 replicates.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Food source, feeding protocol and temp/light conditions were reported. Water quality parameters past salinity concentration were not reported.
	Metric 17:	Outcome Assessment Methodology	High	”On day 5, unhatched eggs, nauplii, and copepodites were counted by light microscopy”
	Metric 18:	Consistency of Outcome Assessment	High	Assessments were consistent among treatments and control groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Medium	No differences among treatment and control groups were attributed to environmental conditions, however, environmental conditions past water temp, salinity, and light cycle were not reported.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	Nothing was reported to indicate differences in groups based on animal attrition or health outcomes.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	onw-way ANOVA and Dunnett post hoc test were performed on egg hatch and larval survival.
	Metric 22:	Reporting of Data	High	Egg hatch and larval survival are reported as mean -/+ 95% CI on page 6/8, Figure 1.
	Metric 23:	Explanation of Unexpected Outcomes	High	No unexpected outcomes were reported.

Additional Comments: Egg hatch and larval survival (no significant differences) are presented for TBBPA for the 5-day exposure experiment on larval developmental rate are presented in Figure 1 on page 6/8.

Overall Quality Determination Medium

Study Citation:	Springborn Laboratories, (1989). Acute toxicity of tetrabromobisphenol A to eastern oysters (Crassostrea virginica) under flow-through conditions with cover letters dated 021489 and 021789.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Mollusks; *Crassostrea virginica*; Juvenile
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547102

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	High	The test substance identity was analytically verified by the performing laboratory
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 100% AI
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, no reported mortalities
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies
	Metric 10:	Exposure Duration and Frequency	High	Standard test duration was used
	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	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest

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Study Citation:	Springborn Laboratories, (1989). Acute toxicity of tetrabromobisphenol A to eastern oysters (Crassostrea virginica) under flow-through conditions with cover letters dated 021489 and 021789.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Mollusks; *Crassostrea virginica*; Juvenile
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547102

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	Medium	Two devisions were reported but not considered to impact the study: 1. The protocol states that the diluter will be visually inspected twice daily. During this study, on1y one of the two or more daily visual inspections was documented.2. A deviation in the performance of the diluter system occurredon day 3 of this study. An electrical malfunction interruptedthe flow of toxicant'to the exposure aquaria, however thismalfunction was repaired and the system returned to no:cDal.function after approximately 8 hours.

Additional Comments: None

Overall Quality Determination High

Study Citation:	Springborn Laboratories, (1989). Bioconcentration and elimination of C-residues by eastern oysters (Crassostrea virginica) exposed to tetrabromobisphenol A (final report) with attached studies and letter 081889.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Mollusks; *Crassostrea virginica*; Juvenile
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547108

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 test substance was obtained from Chemsyn, Lenexa, Kansas. It was not reported if the test substance was analytically verified or not.
	Metric 3:	Test Substance Purity	Low	The test substance purity was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	A control was used.
	Metric 5:	Negative Control Response	High	TBBPA concentrations were always below the detectable level during the entire study.
	Metric 6:	Randomized Allocation	Low	It was not reported if test organisms were randomly allocated to test aquaria.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The test system was adequately described.
	Metric 8:	Consistency of Exposure Administration	High	The exposure was administered consistently.
	Metric 9:	Measurement of Test Substance Concentration	High	Test concentrations were measured at various time points throughout the study.
	Metric 10:	Exposure Duration and Frequency	High	The exposure lasted 20 days, followed by a 14-day depuration period. This was adequate to assess bioconcentration.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	There was only one TBBPA test concentration.
	Metric 12:	Testing at or Below Solubility Limit	High	A solvent was used.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described on page 10/39.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Organisms were acclimated prior to test start.
	Metric 15:	Number of Organisms and Replicates per Group	Low	There was no mention of replicates. There were 60 oysters placed in each test aquarium.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Test conditions were adequate.
	Metric 17:	Outcome Assessment Methodology	High	The assessment of bioconcentration was adequately described.

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Study Citation:	Springborn Laboratories, (1989). Bioconcentration and elimination of C-residues by eastern oysters (Crassostrea virginica) exposed to tetrabromobisphenol A (final report) with attached studies and letter 081889.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Mollusks; *Crassostrea virginica*; Juvenile
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547108

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	High	The assessment was carried out consistently.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	Organisms were acclimated prior to test start and test conditions were adequate.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There were no reported differences in organisms among test groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were described in section 3.3.
	Metric 22:	Reporting of Data	High	Data and results were reported for all test groups.
	Metric 23:	Explanation of Unexpected Outcomes	Low	Measures of variability were not included in the figure.

Additional Comments: This evaluation is for the assessment of TBBPA bioconcentration in oysters.

Overall Quality Determination High

Study Citation:	Brixham Laboratory, (2005). TBBPA: Determination of effects on growth of the common mussel Mytilus edulis.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Mollusks; *Mytilus edulis*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6311092

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.2%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using appropriate concurrent negative control groups.
	Metric 5:	Negative Control Response	High	There were no observed mortalities.
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods (HPLC/MS).
	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	There were five treatment concentrations.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were below the water solubility limit (4.1 mg/L) and a solvent was employed to further aid solubility.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health.

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Study Citation:	Brixham Laboratory, (2005). TBBPA: Determination of effects on growth of the common mussel Mytilus edulis.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Mollusks; *Mytilus edulis*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6311092

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	There were no information presented to suggest differences among groups. Mortality observations were made twice per week.
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
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	N/A	There were no mortalities, so statistical analysis of this endpoint was not necessary.
	Metric 22:	Reporting of Data	Medium	Authors reported in the text that there were no mortalities during the test period. No raw data was shown.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: This evaluation is for the assessment of mortality.

Overall Quality Determination High

Study Citation:	Brixham Laboratory, (2005). TBBPA: Determination of effects on growth of the common mussel Mytilus edulis.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Mollusks; *Mytilus edulis*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6311092

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.2%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using appropriate concurrent negative control groups.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods (HPLC/MS).
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type. Data was analyzed at 14, 28, 42, 56, and 70 days in this long term chronic study.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups (5 treatment concentrations) 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 (4.1 mg/L) and a solvent was employed to further aid solubility
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.

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Study Citation:	Brixham Laboratory, (2005). TBBPA: Determination of effects on growth of the common mussel Mytilus edulis.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Mollusks; *Mytilus edulis*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6311092

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	Medium	There were no information presented to suggest differences among 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 on page 14/86.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group. All raw data are presented on Table 3, page 21.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: Growth Rates for Shell length are presented on page 14/86. Growth rates for tissue weights (dry and wet weight) are on page 16/86.

Overall Quality Determination High

Study Citation:	Fabbri, R., Montagna, M., Balbi, T., Raffo, E., Palumbo, F., Canesi, L. (2014). Adaptation of the bivalve embryotoxicity assay for the high throughput screening of emerging contaminants in Mytilus galloprovincialis. Marine Environmental Research 99:1-8.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Mollusks; *Mytilus galloprovincialis*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3981747

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only.
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity 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 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.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	There are minor reservations about the source of test organisms.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	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	Medium	Reported environmental conditions were not sufficient to evaluate if adequate.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.

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Study Citation:	Fabbri, R., Montagna, M., Balbi, T., Raffo, E., Palumbo, F., Canesi, L. (2014). Adaptation of the bivalve embryotoxicity assay for the high throughput screening of emerging contaminants in Mytilus galloprovincialis. Marine Environmental Research 99:1-8.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Mollusks; *Mytilus galloprovincialis*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3981747

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

Additional Comments: None

Overall Quality Determination Medium

Study Citation:	Ji, C., Li, F., Wang, Q., Zhao, J., Sun, Z., Wu, H. (2016). An integrated proteomic and metabolomic study on the gender-specific responses of mussels Mytilus galloprovincialis to tetrabromobisphenol A (TBBPA). Chemosphere 144:527-539.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Mollusks; *Mytilus galloprovincialis*; Adult
Health Outcome:	Respiratory
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5497037

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The test substance was identified by name only (tetrabromobisphenol A, TBBPA). The authors did not report CASRN, structure, or other chemical descriptors.
	Metric 2:	Test Substance Source	Low	The source was not reported and the identify was not analytically verified by the authors.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	A solvent control (final concentration 0.002% DMSO) was used. The authors explained that based on their previous work, there was no significant proteomic and metabolomic difference in mussel samples between seawater control and solvent, so they did not include a seawater control in this study.
	Metric 5:	Negative Control Response	High	The biological outcome was reported and adequate for the control group (section 3.1, Figure 1).
	Metric 6:	Randomized Allocation	Low	The authors did not report random allocation of the control and treatment groups during TBBPA exposure.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The test media preparation methods lacked important details. The authors reported preparing a stock solution containing a solvent used to prepare the working solution. However, the authors did not report if it was a static, semi-static, or flow-through exposure system, or if water changes were made to replace TBBPA during the 30 day exposure period.
	Metric 8:	Consistency of Exposure Administration	High	There was no evidence that exposure was not consistent across groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration (30 d) was reported and appropriate to measure injury to the gill.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	The goal of the study was not to measure a dose-dependent effect. The study included a solvent control and a single TBBPA group (10 ug/L).
	Metric 12:	Testing at or Below Solubility Limit	High	The exposure concentration (10 ug/L) was below the water solubility limit of TBBPA (4.15 mg/L at 25 C, pH 7.56 and 30.5 mg/L at 25 C, pH 7.99). The exposure was held at 25 C in natural seawater, which has a higher pH than freshwater.
Domain 4: Test Organism				

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Study Citation:	Ji, C., Li, F., Wang, Q., Zhao, J., Sun, Z., Wu, H. (2016). An integrated proteomic and metabolomic study on the gender-specific responses of mussels Mytilus galloprovincialis to tetrabromobisphenol A (TBBPA). Chemosphere 144:527-539.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Mollusks; *Mytilus galloprovincialis*; Adult
Health Outcome:	Respiratory
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5497037

Domain	Metric		Rating	Comments
	Metric 13:	Test Organism Characteristics	Medium	There were minor uncertainties about the test organisms: Adult mussels (shell length:5.5-6.0cm, n=30) were collected in an aquaculture farm (coordinates were provided) where the marine water met the first category quality standard according to "Marine water quality standard of the People's Republic of China". However, the authors did not describe the condition/health status of the mussels.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	After transported to the laboratory, the animals were acclimatized in aerated natural seawater (salinity 31 psu) for 7d.
	Metric 15:	Number of Organisms and Replicates per Group	Low	The authors reported 15 animals to ensure at least six female and six male individuals in each group per treatment group (solvent control and 10 ug/L TBBPA groups). However, there was no mention of replicate groups.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Organism housing conditions (aerated natural seawater (salinity 31 psu), kept at 25°C under a photo period of 12h light and 12h dark, and fed with the Chlorella vulgaris at a ration of 2% of tissue per dry weight daily) were appropriate.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology (histological examination, light microscopy and TUNEL-technique) was reported and appropriate for the intended outcome of interest (TBBPA effect on gill morphology (injury to gill)).
	Metric 18:	Consistency of Outcome Assessment	High	Outcome was consistently assessed 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.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	N/A	The authors reported histological examination of gill tissue in male and female mussels. Background comment: Light microscopy and TUNEL-technique results are commonly descriptive observations of images, and statistical analyses are not most of the time not performed.
	Metric 22:	Reporting of Data	High	Results were reported for both groups (solvent control and treatment) (Figure 1, section 3.1).
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

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Study Citation:	Ji, C., Li, F., Wang, Q., Zhao, J., Sun, Z., Wu, H. (2016). An integrated proteomic and metabolomic study on the gender-specific responses of mussels Mytilus galloprovincialis to tetrabromobisphenol A (TBBPA). Chemosphere 144:527-539.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Mollusks; *Mytilus galloprovincialis*; Adult
Health Outcome:	Respiratory
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5497037

Domain	Metric	Rating	Comments
Additional Comments:	The objective of this study was to characterize the differential responses and elucidate the differential toxicological effects of TBBPA in male and female mussels. Specifically, authors examined organ level injury (histology) and molecular outcomes (gene expression, proteomic, and metabolomic responses) in gill. The gonads were simply used to verify sex. This form was used to evaluate data at the organ level, injury to gill (histology and TUNEL technique). Rationale for selecting respiratory health outcome: Mollusks are filter feeders and thus use the gills to capture food, but the gill is the major organ of respiration for non-mammalian aquatic animals.		

Overall Quality Determination Medium

Study Citation:	Ji, C., Li, F., Wang, Q., Zhao, J., Sun, Z., Wu, H. (2016). An integrated proteomic and metabolomic study on the gender-specific responses of mussels Mytilus galloprovincialis to tetrabromobisphenol A (TBBPA). Chemosphere 144:527-539.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Mollusks; *Mytilus galloprovincialis*; Adult
Health Outcome:	Mechanistic-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5497037

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The test substance was identified by name only (tetrabromobisphenol A, TBBPA). The authors did not report CASRN, structure, or other chemical descriptors.
	Metric 2:	Test Substance Source	Low	The source was not reported and the identify was not analytically verified by the authors.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	A solvent control (final concentration 0.002% DMSO) was used. The authors explained that based on their previous work, there was no significant proteomic and metabolomic difference in mussel samples between seawater control and solvent, so they did not include a seawater control in this study.
	Metric 5:	Negative Control Response	High	The biological outcome was reported and adequate for the control group (section 3.1, Figure 1).
	Metric 6:	Randomized Allocation	Low	The authors did not report random allocation of the control and treatment groups during TBBPA exposure.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The test media preparation methods lacked important details. The authors reported preparing a stock solution containing a solvent used to prepare the working solution. However, the authors did not report if it was a static, semi-static, or flow-through exposure system, or if water changes were made to replace TBBPA during the 30 day exposure period.
	Metric 8:	Consistency of Exposure Administration	High	There was no evidence that exposure was not consistent across groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration (30 d) was reported and appropriate to measure injury to the gill.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	The goal of the study was not to measure a dose-dependent effect. The study included a solvent control and a single TBBPA group (10 ug/L).
	Metric 12:	Testing at or Below Solubility Limit	High	The exposure concentration (10 ug/L) was below the water solubility limit of TBBPA (4.15 mg/L at 25 C, pH 7.56 and 30.5 mg/L at 25 C, pH 7.99). The exposure was held at 25 C in natural seawater, which has a higher pH than freshwater.
Domain 4: Test Organism				

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Study Citation:	Ji, C., Li, F., Wang, Q., Zhao, J., Sun, Z., Wu, H. (2016). An integrated proteomic and metabolomic study on the gender-specific responses of mussels Mytilus galloprovincialis to tetrabromobisphenol A (TBBPA). Chemosphere 144:527-539.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Mollusks; *Mytilus galloprovincialis*; Adult
Health Outcome:	Mechanistic-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5497037

Domain	Metric		Rating	Comments
	Metric 13:	Test Organism Characteristics	Medium	There were minor uncertainties about the test organisms: Adult mussels (shelllength:5.5-6.0cm, n=30) were collected in an aquaculture farm (coordinates were provided) where the marine water met the first category quality standard according to "Marine water quality standard of the People's Republic of China". However, the authors did not describe the condition/health status of the mussels.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	After transported to the laboratory, the animals were acclimatized in aerated natural seawater (salinity 31 psu) for 7d.
	Metric 15:	Number of Organisms and Replicates per Group	Low	Mussel gill tissue metabolomic examination: n=6 for each treatment; iTRAQ-based proteomic examination: n=2 (3 pooled in to one) biological replicates for both male and female mussels for each treatment; mRNA examination: unclear how many organisms were used. The authors did not report replicate exposure units; they only reported exposing 15 mussels to ensure at least six female and six male individuals per group.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Organism housing conditions (aerated natural seawater (salinity 31 psu), kept at 25°C under a photo period of 12h light and 12h dark, and fed with the Chlorella vulgaris at a ration of 2% of tissue per dry weight daily) were appropriate.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodologies (NMR spectroscopy, iTRAQ coupled with RPLC-MS/MS, and qRT-PCR) were reported and appropriate for the intended outcomes of interest (sex-specific metabolomic, proteomic, and mRNA changes following exposure to TBBPA).
	Metric 18:	Consistency of Outcome Assessment	High	Outcome was consistently assessed 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.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Metabolomics: coefficient plots, correlation coefficient (Pearson's product-moment correlation coefficient), Q square and R square for model fitting. Proteomics and mRNA: Student's t-test combined with the Benjamini−Hochberg correction.
	Metric 22:	Reporting of Data	High	Results were reported for both groups (solvent control and treatment) (Table 2, Figures 3 & 4, sections 4.3, 4.5, & 4.6).
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

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Study Citation:	Ji, C., Li, F., Wang, Q., Zhao, J., Sun, Z., Wu, H. (2016). An integrated proteomic and metabolomic study on the gender-specific responses of mussels Mytilus galloprovincialis to tetrabromobisphenol A (TBBPA). Chemosphere 144:527-539.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Mollusks; *Mytilus galloprovincialis*; Adult
Health Outcome:	Mechanistic-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5497037

Domain	Metric	Rating	Comments
Additional Comments:	The objective of this study was to characterize the differential responses and elucidate the differential toxicological effects of TBBPA in male and female mussels. Specifically, authors examined organ level injury (histology) and molecular outcomes (gene expression, proteomic, and metabolomic responses) in gill. The gonads were simply used to verify sex. This form was used to evaluate molecular outcomes (metabolomic data, proteomic data, and mRNA data).		

Overall Quality Determination Medium

Study Citation:	Gong, W., Zhu, L., Hao, Y. (2016). Lethal and sublethal toxicity comparison of BFRs to three marine planktonic copepods: Effects on survival, metabolism and ingestion. PLoS ONE 11(1):e0147790.
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; *Oithona similis*; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3350491

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 >95%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable.
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations. Concentrations of test substance were not measured during the study.
	Metric 8:	Consistency of Exposure Administration	Low	Reporting omissions are likely to have a substantial impact on results. Unclear whether exposure administration was consistent across study groups and controls. The number of exposure groups and exposure concentrations were not reported.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	The number of exposure groups and spacing of exposure levels were not given in the methods other than "The concentration gradients were set up according to a certain common ratio with the mortality less than 10% in the lowest dose and greater than 90% in the highest." Fig 1 shows the concentrations tested but is not very clear.
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	There were significant concerns regarding the source and characteristics of test organisms. Copepods were collected locally from Qingdao Li-cun River estuary, Cheng-yang District He-tao shrimp ponds, and Laoshan District Nanyao shrimp ponds. It was reported that adult copepods were used for testing but unclear whether copepods used for testing were of the same size.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.

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Study Citation:	Gong, W., Zhu, L., Hao, Y. (2016). Lethal and sublethal toxicity comparison of BFRs to three marine planktonic copepods: Effects on survival, metabolism and ingestion. PLoS ONE 11(1):e0147790.
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; *Oithona similis*; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3350491

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms (15 per test chamber) and replicates (3 replicates per concentration) were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reported environmental conditions were not sufficient to evaluate if adequate and whether differences occurred between control and exposed populations.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical methods for determining an LC50 were not reported.
	Metric 22:	Reporting of Data	Medium	Mean values for exposure-related findings were presented for each treatment and control group in Figure 1 but not clear.
	Metric 23:	Explanation of Unexpected Outcomes	Low	LC 50 was reported without confidence intervals and insufficient information was provided to determine if excessive variability or unexpected outcomes occurred.

Additional Comments: This evaluation form is relevant to the mortality outcome for Oithona similis. The reported 96 h-LC50 is 0.566 mg/L of TBBPA.

Overall Quality Determination Medium

Study Citation:	Gong, W., Zhu, L., Hao, Y. (2016). Lethal and sublethal toxicity comparison of BFRs to three marine planktonic copepods: Effects on survival, metabolism and ingestion. PLoS ONE 11(1):e0147790.
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; *Oithona similis*; Adult
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3350491

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 >95%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations. Concentrations of test substance were not measured during the study.
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported (48 hr) and 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 reported as 1.25, 2.5, 5, 10 and 20% of 96h-LC50, and were adequate to address the purpose of the study.
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate ((i.e., no effects on biological responses were observed in the solvent control and no interactions were expected between the solvent and test substance).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	There were significant concerns regarding the source of test organisms. There were significant concerns regarding the source of test organisms. Copepods were collected locally from Qingdao Li-cun River estuary, Cheng-yang District He-tao shrimp ponds, and Laoshan District Nanyao shrimp ponds."A 2000 mL beaker was cultured in advance and the copepods were collected at the same time to assure developments were closely synchronized" No details regarding the duration of culture or copepod size were give.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed groups.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms (30) were suitable for a dose response. The number of replicates (3 per concentration) were reported.

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Study Citation:	Gong, W., Zhu, L., Hao, Y. (2016). Lethal and sublethal toxicity comparison of BFRs to three marine planktonic copepods: Effects on survival, metabolism and ingestion. PLoS ONE 11(1):e0147790.
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; *Oithona similis*; Adult
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3350491

Domain	Metric		Rating	Comments
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reported environmental conditions were not sufficient to evaluate if adequate and whether differences occurred between control and exposed populations.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology to determine ingestion and filtration rates were reported in detail.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described.
	Metric 22:	Reporting of Data	High	Mean values for exposure-related findings were presented for each treatment and control group (Figure 3).
	Metric 23:	Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.

Additional Comments: This evaluation form is for the ingestion and filtrations rates of Oithona similis following exposure to TBBPA. Ingestion and filtration rates are grouped under behavioral outcome to be consistent with ECOTOX output for this study.

Overall Quality Determination Medium

Study Citation:	Gong, W., Zhu, L., Hao, Y. (2016). Lethal and sublethal toxicity comparison of BFRs to three marine planktonic copepods: Effects on survival, metabolism and ingestion. PLoS ONE 11(1):e0147790.
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; *Oithona similis*; Adult
Health Outcome:	Nutritional & Metabolic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3350491

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 >95%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations. Concentrations of test substance were not measured during the study.
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were adequate to address the purpose of the study. 5 exposure groups TBBPA (1.25, 2.5, 5, 10 and 20% of 96h-LC50) were used for the metabolism experiment.
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate based on acute toxicity test.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	There were significant concerns regarding the source and characteristics of test organisms. Copepods were collected locally from Qingdao Li-cun River estuary, Cheng-yang District He-tao shrimp ponds, and Laoshan District Nanyao shrimp ponds. It was reported that adult copepods were used for testing but unclear whether copepods used for testing were of the same size.
	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 (30) and replicates (3 per cncentartion) were reported
Domain 5: Outcome Assessment				

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Study Citation:	Gong, W., Zhu, L., Hao, Y. (2016). Lethal and sublethal toxicity comparison of BFRs to three marine planktonic copepods: Effects on survival, metabolism and ingestion. PLoS ONE 11(1):e0147790.
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; *Oithona similis*; Adult
Health Outcome:	Nutritional & Metabolic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3350491

Domain	Metric		Rating	Comments
	Metric 16:	Adequacy of Test Conditions	Low	Reported environmental conditions were not sufficient to evaluate if adequate and whether differences occurred between control and exposed populations.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodologies for oxygen consumption rate and ammonia excretion rates reported the intended outcome of interest.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described
	Metric 22:	Reporting of Data	High	Mean values for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	Medium	Unexpected outcomes were satisfactorily explained.

Additional Comments: This evaluation form is for the oxygen consumption and ammonia excretion rates of Oithona similis following exposure to TBBPA.

Overall Quality Determination Medium

Study Citation:	Anselmo, R., H.M., Koerting, L., Devito, S., Berg, v.d., J., J.H., Dubbeldam, M., Kwadijk, C., Murk, A. J. (2011). Early life developmental effects of marine persistent organic pollutants on the sea urchin Psammechinus miliaris. Ecotoxicology and Environmental Safety 74(8):2182-2192.
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); *Psammechinus miliaris*; Embryo
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1274149

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	Test substance source was identified (AldrichChemical), but identity was not analytically verified.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 97%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control 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	Medium	The experimental system was described in adequate detail but methods for preparation of test media were not
	Metric 8:	Consistency of Exposure Administration	Medium	Exposures were administered consistently across study groups but few details were provided
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured but organism concentrations were
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The test organisms were adequately described but all organisms came from a single mating pair
	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 numbers of test organisms were reported and sufficient to characterize toxicological effects, however only two replicates was reason for downgrade
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Reported environmental conditions were not sufficient to evaluate if adequate

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Study Citation:	Anselmo, R., H.M., Koerting, L., Devito, S., Berg, v.d., J., J.H., Dubbeldam, M., Kwadijk, C., Murk, A. J. (2011). Early life developmental effects of marine persistent organic pollutants on the sea urchin Psammechinus miliaris. Ecotoxicology and Environmental Safety 74(8):2182-2192.
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); *Psammechinus miliaris*; Embryo
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1274149

Domain	Metric		Rating	Comments
	Metric 17:	Outcome Assessment Methodology	Medium	The outcome assessment methodology reported the intended outcome of interest, some aspects of the assessment were suspect
	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	Average weights were determined from a seperate, unexposed group
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	N/A	Statistical methods were only used to establish a relationship to nominal concentrations
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: None

Overall Quality Determination High

Study Citation:	Anselmo, R., H.M., Koerting, L., Devito, S., Berg, v.d., J., J.H., Dubbeldam, M., Kwadijk, C., Murk, A. J. (2011). Early life developmental effects of marine persistent organic pollutants on the sea urchin Psammechinus miliaris. Ecotoxicology and Environmental Safety 74(8):2182-2192.
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); *Psammechinus miliaris*; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1274149

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	Test substance source was identified (AldrichChemical), but identity was not analytically verified.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 97%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control 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	Medium	The experimental system was described in adequate detail but methods for preparation of test media were not
	Metric 8:	Consistency of Exposure Administration	Medium	Exposures were administered consistently across study groups but few details were provided
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and 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 at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The test organisms were adequately described but all organisms came from a single mating pair
	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 numbers of test organisms were reported and sufficient to characterize toxicological effects, however only two replicates was reason for downgrade
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Reported environmental conditions were not sufficient to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups

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Study Citation:	Anselmo, R., H.M., Koerting, L., Devito, S., Berg, v.d., J., J.H., Dubbeldam, M., Kwadijk, C., Murk, A. J. (2011). Early life developmental effects of marine persistent organic pollutants on the sea urchin Psammechinus miliaris. Ecotoxicology and Environmental Safety 74(8):2182-2192.
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); *Psammechinus miliaris*; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1274149

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 evaluation is for the mortality outcome reported in section 3.2.5 in terms of hatching success.

Overall Quality Determination High

Study Citation:	Anselmo, R., H.M., Koerting, L., Devito, S., Berg, v.d., J., J.H., Dubbeldam, M., Kwadijk, C., Murk, A. J. (2011). Early life developmental effects of marine persistent organic pollutants on the sea urchin Psammechinus miliaris. Ecotoxicology and Environmental Safety 74(8):2182-2192.
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); *Psammechinus miliaris*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1274149

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	Test substance source was identified (AldrichChemical), but identity was not analytically verified.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 97%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control 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	Medium	The experimental system was described in adequate detail but methods for preparation of test media were not
	Metric 8:	Consistency of Exposure Administration	Medium	Exposures were administered consistently across study groups but few details were provided
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and 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 at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The test organisms were adequately described but all organisms came from a single mating pair
	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 numbers of test organisms were reported and sufficient to characterize toxicological effects, however only two replicates was reason for downgrade
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Reported environmental conditions were not sufficient to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups

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Study Citation:	Anselmo, R., H.M., Koerting, L., Devito, S., Berg, v.d., J., J.H., Dubbeldam, M., Kwadijk, C., Murk, A. J. (2011). Early life developmental effects of marine persistent organic pollutants on the sea urchin Psammechinus miliaris. Ecotoxicology and Environmental Safety 74(8):2182-2192.
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); *Psammechinus miliaris*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1274149

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: None

Overall Quality Determination High

Study Citation:	Anselmo, R., H.M., Koerting, L., Devito, S., Berg, v.d., J., J.H., Dubbeldam, M., Kwadijk, C., Murk, A. J. (2011). Early life developmental effects of marine persistent organic pollutants on the sea urchin Psammechinus miliaris. Ecotoxicology and Environmental Safety 74(8):2182-2192.
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); *Psammechinus miliaris*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1274149

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	Test substance source was identified (AldrichChemical), but identity was not analytically verified.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 97%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control 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	Medium	The experimental system was described in adequate detail but methods for preparation of test media were not
	Metric 8:	Consistency of Exposure Administration	Medium	Exposures were administered consistently across study groups but few details were provided
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and 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 at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The test organisms were adequately described but all organisms came from a single mating pair
	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 numbers of test organisms were reported and sufficient to characterize toxicological effects, however only two replicates was reason for downgrade
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Reported environmental conditions were not sufficient to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups

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Study Citation:	Anselmo, R., H.M., Koerting, L., Devito, S., Berg, v.d., J., J.H., Dubbeldam, M., Kwadijk, C., Murk, A. J. (2011). Early life developmental effects of marine persistent organic pollutants on the sea urchin Psammechinus miliaris. Ecotoxicology and Environmental Safety 74(8):2182-2192.
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); *Psammechinus miliaris*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1274149

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: None

Overall Quality Determination High

Study Citation:	Gong, W., Zhu, L., Hao, Y. (2016). Lethal and sublethal toxicity comparison of BFRs to three marine planktonic copepods: Effects on survival, metabolism and ingestion. PLoS ONE 11(1):e0147790.
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; *Pseudodiaptomus inopinus*; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3350491

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 >95%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable.
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations. Concentrations of test substance were not measured during the study.
	Metric 8:	Consistency of Exposure Administration	Low	Reporting omissions are likely to have a substantial impact on results. Unclear whether exposure administration was consistent across study groups and controls. The number of exposure groups and exposure concentrations were not reported.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	The number of exposure groups and spacing of exposure levels were not given in the methods other than "The concentration gradients were set up according to a certain common ratio with the mortality less than 10% in the lowest dose and greater than 90% in the highest." Fig 1 shows the concentrations tested but is not very clear.
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	There were significant concerns regarding the source of test organisms. Copepods were collected locally from Qingdao Li-cun River estuary, Cheng-yang District He-tao shrimp ponds, and Laoshan District Nanyao shrimp ponds. It was reported that adult copepods were used for testing but unclear whether copepods used for testing were of the same size and in the same stage.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms.

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Study Citation:	Gong, W., Zhu, L., Hao, Y. (2016). Lethal and sublethal toxicity comparison of BFRs to three marine planktonic copepods: Effects on survival, metabolism and ingestion. PLoS ONE 11(1):e0147790.
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; *Pseudodiaptomus inopinus*; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3350491

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms (15 per test chamber) and replicates (3 replicates per concentration) were reported and sufficient to characterize toxicological effects.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reported environmental conditions were not sufficient to evaluate if adequate and whether differences occurred between control and exposed populations.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical methods for determining an LC50 were not reported.
	Metric 22:	Reporting of Data	Medium	Mean values for exposure-related findings were presented for each treatment and control group in Figure 1 but not clear.
	Metric 23:	Explanation of Unexpected Outcomes	Low	LC 50 was reported without confidence intervals and insufficient information was provided to determine if excessive variability or unexpected outcomes occurred.

Additional Comments: This evaluation form is relevant to the mortality outcome for Pseudodiaptomus inopinus. The reported 96 h-LC50 is 0.257 mg/L of TBBPA.

Overall Quality Determination Medium

Study Citation:	Gong, W., Zhu, L., Hao, Y. (2016). Lethal and sublethal toxicity comparison of BFRs to three marine planktonic copepods: Effects on survival, metabolism and ingestion. PLoS ONE 11(1):e0147790.
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; *Pseudodiaptomus inopinus*; Adult
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3350491

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 >95%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations. Concentrations of test substance were not measured during the study.
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported (48 hr) and 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 reported as 1.25, 2.5, 5, 10 and 20% of 96h-LC50, and were adequate to address the purpose of the study.
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate ((i.e., no effects on biological responses were observed in the solvent control and no interactions were expected between the solvent and test substance).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	There were significant concerns regarding the source of test organisms. There were significant concerns regarding the source of test organisms. Copepods were collected locally from Qingdao Li-cun River estuary, Cheng-yang District He-tao shrimp ponds, and Laoshan District Nanyao shrimp ponds."A 2000 mL beaker was cultured in advance and the copepods were collected at the same time to assure developments were closely synchronized" No details regarding the duration of culture or copepod size were give.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed groups.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms (30) were suitable for a dose response. The number of replicates (3 per concentration) were reported.

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Study Citation:	Gong, W., Zhu, L., Hao, Y. (2016). Lethal and sublethal toxicity comparison of BFRs to three marine planktonic copepods: Effects on survival, metabolism and ingestion. PLoS ONE 11(1):e0147790.
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; *Pseudodiaptomus inopinus*; Adult
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3350491

Domain	Metric		Rating	Comments
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reported environmental conditions were not sufficient to evaluate if adequate and whether differences occurred between control and exposed populations.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology to determine ingestion and filtration rates were reported in detail.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described.
	Metric 22:	Reporting of Data	High	Mean values for exposure-related findings were presented for each treatment and control group (Figure 3).
	Metric 23:	Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.

Additional Comments: This evaluation form is for the ingestion and filtrations rates of Pseudodiaptomus inopinus following exposure to TBBPA. Ingestion and filtration rates are grouped under behavioral outcome to be consistent with ECOTOX output for this study.

Overall Quality Determination Medium

Study Citation:	Gong, W., Zhu, L., Hao, Y. (2016). Lethal and sublethal toxicity comparison of BFRs to three marine planktonic copepods: Effects on survival, metabolism and ingestion. PLoS ONE 11(1):e0147790.
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; *Pseudodiaptomus inopinus*; Adult
Health Outcome:	Nutritional & Metabolic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3350491

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 >95%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations. Concentrations of test substance were not measured during the study.
	Metric 8:	Consistency of Exposure Administration	High	Details of exposure administration were reported and exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported (48 hr) and 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 reported as 1.25, 2.5, 5, 10 and 20% of 96h-LC50, and were adequate to address the purpose of the study.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	There were significant concerns regarding the source of test organisms. There were significant concerns regarding the source of test organisms. Copepods were collected locally from Qingdao Li-cun River estuary, Cheng-yang District He-tao shrimp ponds, and Laoshan District Nanyao shrimp ponds."A 2000 mL beaker was cultured in advance and the copepods were collected at the same time to assure developments were closely synchronized" No details regarding the duration of culture or copepod size were give.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed groups.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms (30) were suitable for a dose response. The number of replicates (3 per concentration) were reported.
Domain 5: Outcome Assessment				

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Study Citation:	Gong, W., Zhu, L., Hao, Y. (2016). Lethal and sublethal toxicity comparison of BFRs to three marine planktonic copepods: Effects on survival, metabolism and ingestion. PLoS ONE 11(1):e0147790.
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; *Pseudodiaptomus inopinus*; Adult
Health Outcome:	Nutritional & Metabolic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3350491

Domain	Metric		Rating	Comments
	Metric 16:	Adequacy of Test Conditions	Low	Reported environmental conditions were not sufficient to evaluate if adequate and whether differences occurred between control and exposed populations.
	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 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
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described.
	Metric 22:	Reporting of Data	High	Mean values for exposure-related findings were presented for each treatment and control group (Figure 2).
	Metric 23:	Explanation of Unexpected Outcomes	High	Unexpected outcomes were satisfactorily explained.

Additional Comments: This evaluation form is for the oxygen consumption and ammonia excretion rates of Pseudodiaptomus inopinus following exposure to TBBPA.

Overall Quality Determination Medium

Study Citation:	Gong, W.,j, Zhu, L.,y, Jiang, T.,t, Han, C. (2017). The occurrence and spatial-temporal distribution of tetrabromobisphenol A in the coastal intertidal zone of Qingdao in China, with a focus on toxicity assessment by biological monitoring. Chemosphere 185:462-467.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; *Pseudodiaptomus inopinus.*; Larvae
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5519030

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 >98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control groups
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system was described in adequate detail, methods for preparation of test media were minimal
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	There were minor limitations regarding the number of exposure groups but spacing of exposure levels was adequate
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The test organisms were adequately described and were obtained from a long standing culture
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms,
	Metric 15:	Number of Organisms and Replicates per Group	Medium	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	Environmental conditions were not sufficiently reported to evaluate if adequate

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Study Citation:	Gong, W.,j, Zhu, L.,y, Jiang, T.,t, Han, C. (2017). The occurrence and spatial-temporal distribution of tetrabromobisphenol A in the coastal intertidal zone of Qingdao in China, with a focus on toxicity assessment by biological monitoring. Chemosphere 185:462-467.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; *Pseudodiaptomus inopinus.*; Larvae
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5519030

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

Additional Comments: Multi generational test, development rate and maturation time

Overall Quality Determination Medium

Study Citation:	Gong, W.,j, Zhu, L.,y, Jiang, T.,t, Han, C. (2017). The occurrence and spatial-temporal distribution of tetrabromobisphenol A in the coastal intertidal zone of Qingdao in China, with a focus on toxicity assessment by biological monitoring. Chemosphere 185:462-467.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; *Pseudodiaptomus inopinus.*; Larvae
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5519030

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 >98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control groups
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system was described in adequate detail, methods for preparation of test media were minimal
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	There were minor limitations regarding the number of exposure groups but spacing of exposure levels was adequate
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The test organisms were adequately described and were obtained from a long standing culture
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms,
	Metric 15:	Number of Organisms and Replicates per Group	Medium	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	Environmental conditions were not sufficiently reported to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	Medium	The outcome assessment methodology reported the intended outcome of interest but few details were provided

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Study Citation:	Gong, W.,j, Zhu, L.,y, Jiang, T.,t, Han, C. (2017). The occurrence and spatial-temporal distribution of tetrabromobisphenol A in the coastal intertidal zone of Qingdao in China, with a focus on toxicity assessment by biological monitoring. Chemosphere 185:462-467.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Arthropods; *Pseudodiaptomus inopinus.*; Larvae
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5519030

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

Additional Comments: Multi generational test, nauplii produced and hatch rate

Overall Quality Determination Medium

Study Citation:	Jiang, S., Miao, J., Wang, X., Liu, P., Pan, L. (2019). Inhibition of growth in juvenile manila clam Ruditapes philippinarum: Potential adverse outcome pathway of TBBPA. Chemosphere 224:588-596.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Mollusks; *Ruditapes phillipinarum*; Juvenile
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5635811

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The chemical substance was identified by name only: Tetrabromobisphenol A (TBBPA). No CASRN, structure, or other chemical descriptors were provided.
	Metric 2:	Test Substance Source	Low	Test substance source was reported (Sigma Chemical Co.Ltd., UK) but the identity was not chemically verified.
	Metric 3:	Test Substance Purity	High	Chemical purity was 99.3%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	In the acute toxicity test, the authors reported using a solvent control (0.001% dimethyl sulfoxide, DMSO).
	Metric 5:	Negative Control Response	Medium	The authors reported in the text that the survival of the control group was nearly 100% after 96 h. However, they did not show the actual mortality/survival data per treatment group including the control group.
	Metric 6:	Randomized Allocation	Low	The authors did not report random allocation.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The test media preparation was not described in detail (e.g., stock preparation was not reported), but the authors did report using a solvent to dissolve TBBPA (DMSO, final concentration of 0.001% DMSO).
	Metric 8:	Consistency of Exposure Administration	High	There is no evidence to suggest that exposure administration was not administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	The authors reported measuring TBBPA in solution and organism tissues using HPLC (details in the supplementary material, text S1). However, there are no results reported on their measured TBBPA concentrations and all data was reported using nominal concentrations.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration (96 h) was reported and appropriate for the study type (acute toxicity study to derive an 96h-LC50 value).
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Acute toxicity study: The number of exposure groups were appropriate (control and five concentrations of nominal TBBPA (500, 1000, 2000, 4000, 8000 ug/L)). The authors also indicated that these concentrations were selected after a range-finding test.

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Study Citation:	Jiang, S., Miao, J., Wang, X., Liu, P., Pan, L. (2019). Inhibition of growth in juvenile manila clam Ruditapes philippinarum: Potential adverse outcome pathway of TBBPA. Chemosphere 224:588-596.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Mollusks; *Ruditapes phillipinarum*; Juvenile
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5635811

Domain	Metric		Rating	Comments
	Metric 12:	Testing at or Below Solubility Limit	High	A solvent was used to dissolve TBBPA. Considerations: The TBBPA water solubility limit is 4.15 mg/L at 25 C & pH 7.56 and 30.5 mg/L at 25 C & pH 7.99. This experiment was conducted with a marine species and the pH was 8.2 in the seawater used as reported by authors. Moreover, the experiment was run at 18 C (not 25 C), and solubility decrease as temperature decreases. The highest concentration (8000 ug/L) was potentially above the solubility limit. Ultimately, the authors used 0.001% DMSO as the solvent to bring TBBPA into solution.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The organisms were well described (Ruditapes philippinarum, five-month-old juveniles) and were obtained from a reliable source (laboratory bred and cultured stocks from Red Island clam culture center in Qingdao).
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Organisms were acclimated for 7 days and pretreatment culture conditions were described, appropriate, and the same for all treatment groups.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Acute toxicity test: 6 treatment groups, 4 replicate beakers per group, and 10 clams per beaker.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Organism maintenance was described and appropriate (section 2.1.1).
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology (counts of number of living and dead clams at 24 h, 48 h, 72 h and 96 h; foot movement within 5 min was classified as alive) was reported and appropriate for the outcome of interest (acute toxicity to derive 96h-LC50).
	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.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	The nominal TBBPA concentrations were used for the calculationof LC50s using a Probit model with SPSS 17.0 software.
	Metric 22:	Reporting of Data	Low	The authors did not report mortality/survival for each treatment group. They only reported the mortality findings in one sentence in the text of the results (section 3.1): The 96-h LC50 was 7400 ug/L with a 95% confidence interval of 5700-11300 ug/L. This LC50 value is above the solubility limit of TBBPA.

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Study Citation:	Jiang, S., Miao, J., Wang, X., Liu, P., Pan, L. (2019). Inhibition of growth in juvenile manila clam Ruditapes philippinarum: Potential adverse outcome pathway of TBBPA. Chemosphere 224:588-596.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Mollusks; *Ruditapes phillipinarum*; Juvenile
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5635811

Domain	Metric	Rating	Comments
	Metric 23: Explanation of Unexpected Outcomes	Low	The authors reported that the 96-h LC50 value of 7400 ug/L (95% CI of 5700-11300 ug/L) was above the solubility limit of TBBPA. They also indicated that nominal TBBPA concentrations were used for the calculation of the LC50. However, it is unclear why they used the nominal concentrations to report all data if they already measured TBBPA. It is also unclear if the LC50 value is above the water solubility limit in this experiment given that it was performed in seawater with a of pH 8.2 and at 18 C. Temperature and pH both affect solubility of TBBPA.

Additional Comments: This study investigated TBBPA effects on growth impairment in the marine clam Ruditapes phillipinarum measured by shell size, filtration rate, tissue thyroid hormone (TH) contents, and gene expression levels of insulin-like peptides and TH synthesis-related enzymes. The objective of the present study was to establish if linkage could be drawn between molecular events and apical endpoints (growth impairment). To be consistent with the ECOTOX output, filtration rate was tagged as a behavioral (feeding behavior) effect. This form was used to evaluate the acute toxicity data (derivation of 96h-LC50).

Overall Quality Determination High

Study Citation:	Jiang, S., Miao, J., Wang, X., Liu, P., Pan, L. (2019). Inhibition of growth in juvenile manila clam Ruditapes philippinarum: Potential adverse outcome pathway of TBBPA. Chemosphere 224:588-596.
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; *Ruditapes phillipinarum*; Juvenile
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5635811

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The chemical substance was identified by name only: Tetrabromobisphenol A (TBBPA). No CASRN, structure, or other chemical descriptors were provided.
	Metric 2:	Test Substance Source	Low	Test substance source was reported (Sigma Chemical Co.Ltd., UK) but the identity was not chemically verified.
	Metric 3:	Test Substance Purity	High	Chemical purity was 99.3%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Filtration rate test: The authors reported using a control (0 ug/L)(section 2.2).
	Metric 5:	Negative Control Response	High	The biological response of the control (filtration rate) was reported and adequate (Figure 1, section 3.2).
	Metric 6:	Randomized Allocation	Low	The authors did not report random allocation.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The test media preparation was not described in detail (e.g., stock preparation was not reported), but the authors did report using a solvent to dissolve TBBPA (DMSO, final concentration of 0.001% DMSO).
	Metric 8:	Consistency of Exposure Administration	High	There is no evidence to suggest that exposure administration was not administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	The authors reported measuring TBBPA in solution and organism tissues using HPLC (details in the supplementary material, text S1). However, there are no results reported on their measured TBBPA concentrations and all data was reported using nominal concentrations.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration (14 d) was reported and appropriate for the study type (clam filtration rate).
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Filtration rate test: The number of exposure groups were appropriate (0, 62.5, and 500 ug/L). The authors selected these concentration based on an acute toxicity test to derive the 96h-LC50 value.
	Metric 12:	Testing at or Below Solubility Limit	High	Filtration rate test: All concentrations were below the water solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The organisms were well described (Ruditapes philippinarum, five-month-old juveniles) and were obtained from a reliable source (laboratory bred and cultured stocks from Red Island clam culture center in Qingdao).

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Study Citation:	Jiang, S., Miao, J., Wang, X., Liu, P., Pan, L. (2019). Inhibition of growth in juvenile manila clam Ruditapes philippinarum: Potential adverse outcome pathway of TBBPA. Chemosphere 224:588-596.
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; *Ruditapes phillipinarum*; Juvenile
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5635811

Domain	Metric		Rating	Comments
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Organisms were acclimated for at least 7 days (7 day acclimatization prior to acute test, and this chronic test was ran after acute test) and pretreatment culture conditions were described, appropriate, and the same for all treatment groups.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Filtration rate test: 3 treatment groups, 4 replicate aquaria with 100 individuals; 10 individuals per aquarium were transferred to beakers to measure filtration rate on days 0, 1, 3, 7 and 14.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Organism maintenance was described and appropriate.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology (measurement of the loss of neutral red dye particles from the water column) was reported and appropriate for the outcome of interest (TBBPA impact on filtration rate).
	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.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical differences were analyzed by ANOVA followed by Tukey'smultiple comparison test.
	Metric 22:	Reporting of Data	High	Test findings were reported for all treatment groups (Figure 1).
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This study investigated TBBPA effects on growth impairment in the marine clam Ruditapes phillipinarum measured by shell size, filtration rate, tissue thyroid hormone (TH) contents, and gene expression levels of insulin-like peptides and TH synthesis-related enzymes. The objective of the present study was to establish if linkage could be drawn between molecular events and apical endpoints (growth impairment). To be consistent with the ECOTOX output, filtration rate was tagged as a behavioral (feeding behavior) effect. This form was used to evaluate the effects of TBBPA on filtration rate.

Overall Quality Determination High

Study Citation:	Jiang, S., Miao, J., Wang, X., Liu, P., Pan, L. (2019). Inhibition of growth in juvenile manila clam Ruditapes philippinarum: Potential adverse outcome pathway of TBBPA. Chemosphere 224:588-596.
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; *Ruditapes phillipinarum*; Juvenile
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5635811

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The chemical substance was identified by name only: Tetrabromobisphenol A (TBBPA). No CASRN, structure, or other chemical descriptors were provided.
	Metric 2:	Test Substance Source	Low	Test substance source was reported (Sigma Chemical Co.Ltd., UK) but the identity was not chemically verified.
	Metric 3:	Test Substance Purity	High	Chemical purity was 99.3%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	The authors reported using a control (0 ug/L).
	Metric 5:	Negative Control Response	High	The biological response of the control (concentration of T4 & T3 and gene expression) was reported and adequate (Table 3, Figure 2, sections 3.3 & 3.4).
	Metric 6:	Randomized Allocation	Low	The authors did not report random allocation.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The test media preparation was not described in detail (e.g., stock preparation was not reported), but the authors did report using a solvent to dissolve TBBPA (DMSO, final concentration of 0.001% DMSO).
	Metric 8:	Consistency of Exposure Administration	High	There is no evidence to suggest that exposure administration was not administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	The authors reported measuring TBBPA in solution and organism tissues using HPLC (details in the supplementary material, text S1). However, there are no results reported on their measured TBBPA concentrations and all data was reported using nominal concentrations.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration (14 d) was reported and appropriate for the study type (thyroid concentrations and gene expression changes).
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups were appropriate (0, 62.5, and 500 ug/L). The authors selected these concentration based on an acute toxicity test to derive the 96h-LC50 value.
	Metric 12:	Testing at or Below Solubility Limit	High	All concentrations were below the water solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The organisms were well described (Ruditapes philippinarum, five-month-old juveniles) and were obtained from a reliable source (laboratory bred and cultured stocks from Red Island clam culture center in Qingdao).

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Study Citation:	Jiang, S., Miao, J., Wang, X., Liu, P., Pan, L. (2019). Inhibition of growth in juvenile manila clam Ruditapes philippinarum: Potential adverse outcome pathway of TBBPA. Chemosphere 224:588-596.
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; *Ruditapes phillipinarum*; Juvenile
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5635811

Domain	Metric		Rating	Comments
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Organisms were acclimated for at least 7 days (7 day acclimatization prior to acute test, and this chronic test was ran after acute test) and pretreatment culture conditions were described, appropriate, and the same for all treatment groups.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	3 treatment groups, 4 replicate aquaria, 100 individuals per aquarium; 10 individuals per aquarium were used to measure thyroid and 10 for gene expression on days 0, 1, 3, 7 and 14.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Organism maintenance was described and appropriate.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology (T3 & T4 enzyme-linked immunosorbent assays and qPCR) was reported and appropriate for the outcome of interest (TBBPA impact on thyroid concentration and gene expression changes).
	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.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical differences were analyzed by ANOVA followed by Tukey'smultiple comparison test.
	Metric 22:	Reporting of Data	High	Test findings were reported for all treatment groups (Table 3, Figure 2).
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This study investigated TBBPA effects on growth impairment in the marine clam Ruditapes phillipinarum measured by shell size, filtration rate, tissue thyroid hormone (TH) contents, and gene expression levels of insulin-like peptides and TH synthesis-related enzymes. The objective of the present study was to establish if linkage could be drawn between molecular events and apical endpoints (growth impairment). To be consistent with the ECOTOX output filtration rate was tagged as a behavioral (feeding behavior) effect. This form was used to evaluate the effects of TBBPA on tissue thyroid hormone (TH) contents, and gene expression levels of insulin-like peptides and TH synthesis-related enzymes.

Overall Quality Determination High

Study Citation:	Jiang, S., Miao, J., Wang, X., Liu, P., Pan, L. (2019). Inhibition of growth in juvenile manila clam Ruditapes philippinarum: Potential adverse outcome pathway of TBBPA. Chemosphere 224:588-596.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Mollusks; *Ruditapes phillipinarum*; Juvenile
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5635811

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The chemical substance was identified by name only: Tetrabromobisphenol A (TBBPA). No CASRN, structure, or other chemical descriptors were provided.
	Metric 2:	Test Substance Source	Low	Test substance source was reported (Sigma Chemical Co.Ltd., UK) but the identity was not chemically verified.
	Metric 3:	Test Substance Purity	High	Chemical purity was 99.3%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Chronic toxicity test (shell growth): The authors reported using a control (0 ug/L)(section 2.1.2).
	Metric 5:	Negative Control Response	High	The biological response of the control (shell height and length) was reported and adequate (Table 2, section 3.1).
	Metric 6:	Randomized Allocation	Low	The authors did not report random allocation.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The test media preparation was not described in detail (e.g., stock preparation was not reported), but the authors did report using a solvent to dissolve TBBPA (DMSO, final concentration of 0.001% DMSO).
	Metric 8:	Consistency of Exposure Administration	High	There is no evidence to suggest that exposure administration was not administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	The authors reported measuring TBBPA in solution and organism tissues using HPLC (details in the supplementary material, text S1). However, there are no results reported on their measured TBBPA concentrations and all data was reported using nominal concentrations.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration (28 d) was reported and appropriate for the study type (clam shell growth).
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Chronic toxicity test (shell growth): The number of exposure groups were appropriate (0, 62.5, 125, 250, 500 and 1000 ug/L). The authors selected these concentration based on an acute toxicity test to derive the 96h-LC50 value.
	Metric 12:	Testing at or Below Solubility Limit	High	Chronic toxicity test (shell growth): All concentrations were below the water solubility limit.
Domain 4: Test Organism				

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Study Citation:	Jiang, S., Miao, J., Wang, X., Liu, P., Pan, L. (2019). Inhibition of growth in juvenile manila clam Ruditapes philippinarum: Potential adverse outcome pathway of TBBPA. Chemosphere 224:588-596.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (marine); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Mollusks; *Ruditapes phillipinarum*; Juvenile
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5635811

Domain	Metric		Rating	Comments
	Metric 13:	Test Organism Characteristics	High	The organisms were well described (Ruditapes philippinarum, five-month-old juveniles) and were obtained from a reliable source (laboratory bred and cultured stocks from Red Island clam culture center in Qingdao).
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Organisms were acclimated for at least 7 days (7 day acclimatization prior to acute test, and this chronic test was ran after acute test) and pretreatment culture conditions were described, appropriate, and the same for all treatment groups.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Chronic toxicity test (shell growth): 6 treatment groups, 4 replicate beakers, and 20 individuals per beaker.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Organism maintenance was described and appropriate.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology (measurement of shell length and shell height; increases of shell height and shell length compared to the initial measurements) was reported and appropriate for the outcome of interest (TBBPA impact on shell growth).
	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.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical differences were analyzed by ANOVA followed by Tukey'smultiple comparison test.
	Metric 22:	Reporting of Data	High	Test findings were reported for all treatment groups (Table 2).
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This study investigated TBBPA effects on growth impairment in the marine clam Ruditapes phillipinarum measured by shell size, filtration rate, tissue thyroid hormone (TH) contents, and gene expression levels of insulin-like peptides and TH synthesis-related enzymes. The objective of the present study was to establish if linkage could be drawn between molecular events and apical endpoints (growth impairment). To be consistent with the ECOTOX output, filtration rate was tagged as a behavioral (feeding behavior) effect. This form was used to evaluate the chronic toxicity test to determine TBBPA effects on clam shell growth.

Overall Quality Determination High

Study Citation:	Walsh, G. E., Yoder, M. J., Mclaughlin, L. L., Lores, E. M. (1987). Responses of marine unicellular algae to brominated organic compounds in six growth media. Ecotoxicology and Environmental Safety 14(3):215-222.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (marine); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Non-vascular Plants; *Skeletonema costatum*; Greville; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1927837

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 reported but the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Negative and solvent controls were used (algal growth of controls reported in Table 5) but unclear whether these were tested concurrently.
	Metric 5:	Negative Control Response	Medium	The biological responses of the negative control groups were reported (Table 5) but unclear whether the controls were tested concurrently with the treatment groups.
	Metric 6:	Randomized Allocation	Low	Algal study, randomized allocation not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	Experiments were conducted in Delong culture flask capped with stainless steel closures. Test media preparation methods were not described in detail. Solvent concentration used was not reported.
	Metric 8:	Consistency of Exposure Administration	Low	Reporting omissions (exposure concentrations, solvent concentration, concurrent use of controls, etc.) are likely to have a substantial impact on results.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were measured but not reported.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported (72 hr) and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Uninformative	No information is provided on the number of exposure groups and spacing of exposure levels.
	Metric 12:	Testing at or Below Solubility Limit	Low	The solvent concentration in itself was not reported but inferred appropriate ((i.e., no effects on biological responses were observed in the solvent control and no interactions were expected between the solvent and test substance). However, no information was provided on the number of exposure groups, so it is difficult to determine if the solvent concentration used was appropriate to dissolve all the exposure concentrations.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	S. costatum was obtained from the culture collection of the University of Rhode Island. It was not stated whether the inoculum culture was in the exponential growth phase, which is recommended for algal toxicity assays (OECD 201).

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Study Citation:	Walsh, G. E., Yoder, M. J., Mclaughlin, L. L., Lores, E. M. (1987). Responses of marine unicellular algae to brominated organic compounds in six growth media. Ecotoxicology and Environmental Safety 14(3):215-222.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (marine); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Non-vascular Plants; *Skeletonema costatum*; Greville; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1927837

Domain	Metric		Rating	Comments
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized and whether pretreatment conditions were the same for control and exposed groups.
	Metric 15:	Number of Organisms and Replicates per Group	Low	The initial biomass used in the test cultures was not provided. It was stated that each test was replicated.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reporting of environmental conditions and biomass loading was not sufficient to evaluate if adequate and whether differences occurred between control and exposed populations.
	Metric 17:	Outcome Assessment Methodology	Medium	The outcome assessment methodology reported the intended outcome of interest. Cell counts were done using a hemacytometer after 72 hours of exposure in treatment groups. However, it was unclear whether cell counts in controls were done at the same time as the treatment groups.
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment of treatment groups and controls were limited.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. In the discussion section, the authors report that pH of the growth media may have played a role in the observed toxicity but pH was only measured before the test and not during or after the test.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure (e.g., infection) that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately. No statistical test was conducted to see whether algal density in treatment groups were significantly different from controls. Methods for EC50 determination was from an in press report.
	Metric 22:	Reporting of Data	Low	Algal density data for exposure-related findings were not shown for each treatment and control group. EC 50 values were provided for each growth media test without confidence intervals (Table 3a).
	Metric 23:	Explanation of Unexpected Outcomes	Low	Insufficient information was provided to determine if excessive variability or unexpected outcomes occurred.

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Study Citation:	Walsh, G. E., Yoder, M. J., Mclaughlin, L. L., Lores, E. M. (1987). Responses of marine unicellular algae to brominated organic compounds in six growth media. Ecotoxicology and Environmental Safety 14(3):215-222.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (marine); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Non-vascular Plants; *Skeletonema costatum*; Greville; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1927837

Domain	Metric	Rating	Comments
Additional Comments:	The number of exposure groups, exposure concentrations, solvent concentration used and algal population density data for treatment groups and controls were not provided. EC 50 values were calculated using an unpublished method and presented without confidence intervals.		

Overall Quality Determination — Uninformative

Study Citation:	Walsh, G. E., Yoder, M. J., Mclaughlin, L. L., Lores, E. M. (1987). Responses of marine unicellular algae to brominated organic compounds in six growth media. Ecotoxicology and Environmental Safety 14(3):215-222.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (marine); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Non-vascular Plants; *Skeletonema costatum*; Greville; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1927837

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 reported but the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Negative and solvent controls were used (algal growth of controls reported in Table 5) but unclear whether these were tested concurrently.
	Metric 5:	Negative Control Response	Medium	The biological responses of the negative control groups were reported (Table 5) but it was unclear whether the controls were tested concurrently with the treatment groups.
	Metric 6:	Randomized Allocation	Low	Algal study, randomized allocation not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	Experiments were conducted in Delong culture flask capped with stainless steel closures. Test media preparation methods were not described in detail. Solvent concentration used was not reported.
	Metric 8:	Consistency of Exposure Administration	Low	Reporting omissions (exposure concentrations, solvent concentration, concurrent use of controls, etc.) were likely to have a substantial impact on results.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were measured but not reported.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported (72 hr) and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Uninformative	No information is provided on the number of exposure groups and spacing of exposure levels.
	Metric 12:	Testing at or Below Solubility Limit	Low	The solvent concentration in itself was not reported but inferred appropriate ((i.e., no effects on biological responses were observed in the solvent control and no interactions were expected between the solvent and test substance). However, no information was provided on the number of exposure groups, so it is difficult to determine if the solvent concentration used was appropriate to dissolve all the exposure concentrations.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	S. costatum was obtained from the culture collection of the University of Rhode Island. It was not stated whether the inoculum culture was in the exponential growth phase, which is recommended for algal toxicity assays (OECD 201).
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized and whether pretreatment conditions were the same for control and exposed groups.

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Study Citation:	Walsh, G. E., Yoder, M. J., Mclaughlin, L. L., Lores, E. M. (1987). Responses of marine unicellular algae to brominated organic compounds in six growth media. Ecotoxicology and Environmental Safety 14(3):215-222.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (marine); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Non-vascular Plants; *Skeletonema costatum*; Greville; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1927837

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Low	The initial biomass used in the test cultures was not provided. It was stated that each test was replicated.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reporting of environmental conditions and biomass loading was not sufficient to evaluate if adequate and whether differences occurred between control and exposed populations.
	Metric 17:	Outcome Assessment Methodology	Medium	The outcome assessment methodology reported the intended outcome of interest. Cell counts were done using a hemacytometer after 72 hours of exposure in treatment groups. However, it was unclear whether cell counts in controls were done at the same time as the treatment groups.
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment of treatment groups and controls were limited.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. In the discussion section, the authors report that pH of the growth media may have played a role in the observed toxicity but pH was only measured before the test and not during or after the test.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure (e.g., infection) that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately. No statistical test was conducted to see whether algal density in treatment groups were significantly different from controls. Methods for EC50 determination was from an in press report.
	Metric 22:	Reporting of Data	Low	Algal density data for exposure-related findings were not shown for each treatment and control group. EC50 values were provided for each growth media test without confidence intervals (Table 3a).
	Metric 23:	Explanation of Unexpected Outcomes	Low	Insufficient information was provided to determine if excessive variability or unexpected outcomes occurred.

Additional Comments: The number of exposure groups, exposure concentrations, solvent concentration used and algal population density data for treatment groups and controls were not provided. EC50 values were calculated using an unpublished method and presented without confidence intervals.

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Study Citation:	Walsh, G. E., Yoder, M. J., Mclaughlin, L. L., Lores, E. M. (1987). Responses of marine unicellular algae to brominated organic compounds in six growth media. Ecotoxicology and Environmental Safety 14(3):215-222.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (marine); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Non-vascular Plants; *Skeletonema costatum*; Greville; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1927837

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

Study Citation:	Walsh, G. E., Yoder, M. J., Mclaughlin, L. L., Lores, E. M. (1987). Responses of marine unicellular algae to brominated organic compounds in six growth media. Ecotoxicology and Environmental Safety 14(3):215-222.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (marine); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Non-vascular Plants; *Skeletonema costatum*; Greville; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1927837

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 reported but the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Negative and solvent controls were used (algal growth of controls reported in Table 5) but unclear whether these were tested concurrently.
	Metric 5:	Negative Control Response	Medium	The biological responses of the negative control groups were reported (Table 5), but it was unclear whether the controls were tested concurrently with the treatment groups.
	Metric 6:	Randomized Allocation	Low	Algal study, randomized allocation not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	Experiments were conducted in Delong culture flask capped with stainless steel closures. Test media preparation methods were not described in detail. Solvent concentration used was not reported.
	Metric 8:	Consistency of Exposure Administration	Low	Reporting omissions (exposure concentrations, solvent concentration, concurrent use of controls, etc.) are likely to have a substantial impact on results.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were measured but not reported.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported (72 hr) and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Uninformative	No information is provided on the number of exposure groups and spacing of exposure levels.
	Metric 12:	Testing at or Below Solubility Limit	Low	The solvent concentration in itself was not reported but inferred appropriate ((i.e., no effects on biological responses were observed in the solvent control and no interactions were expected between the solvent and test substance). However, no information was provided on the number of exposure groups, so it is difficult to determine if the solvent concentration used was appropriate to dissolve all the exposure concentrations.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	S. costatum was obtained from the culture collection of the University of Rhode Island. It was not stated whether the inoculum culture was in the exponential growth phase, which is recommended for algal toxicity assays (OECD 201).
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized and whether pretreatment conditions were the same for control and exposed groups.

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Study Citation:	Walsh, G. E., Yoder, M. J., Mclaughlin, L. L., Lores, E. M. (1987). Responses of marine unicellular algae to brominated organic compounds in six growth media. Ecotoxicology and Environmental Safety 14(3):215-222.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (marine); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Non-vascular Plants; *Skeletonema costatum*; Greville; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1927837

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Low	The initial biomass used in the test cultures was not provided. It was stated that each test was replicated.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reporting of environmental conditions and biomass loading was not sufficient to evaluate if adequate and whether differences occurred between control and exposed populations.
	Metric 17:	Outcome Assessment Methodology	Medium	The outcome assessment methodology reported the intended outcome of interest. Cell counts were done using a hemacytometer after 72 hours of exposure in treatment groups. However, it was unclear whether cell counts in controls were done at the same time as the treatment groups.
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment of treatment groups and controls were limited.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. In the discussion section, the authors report that pH of the growth media may have played a role in the observed toxicity but pH was only measured before the test and not during or after the test.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure (e.g., infection) that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately. No statistical test was conducted to see whether algal density in treatment groups were significantly different from controls. Methods for EC50 determination was from an in press report.
	Metric 22:	Reporting of Data	Low	Algal density data for exposure-related findings were not shown for each treatment and control group. EC 50 values were provided for each growth media test without confidence intervals (Table 3a).
	Metric 23:	Explanation of Unexpected Outcomes	Low	Insufficient information was provided to determine if excessive variability or unexpected outcomes occurred.

Additional Comments: The number of exposure groups, exposure concentrations, solvent concentration used and algal population density data for treatment groups and controls were not provided. EC 50 values were calculated using an unpublished method and presented without confidence intervals.

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Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (marine); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Non-vascular Plants; *Skeletonema costatum*; Greville; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1927837

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

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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 reported but the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Negative and solvent controls were used (algal growth of controls reported in Table 5) but unclear whether these were tested concurrently.
	Metric 5:	Negative Control Response	Medium	The biological responses of the negative control groups were reported (Table 5) but unclear whether the controls were tested concurrently with the treatment groups.
	Metric 6:	Randomized Allocation	Low	Algal study, randomized allocation not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	Experiments were conducted in Delong culture flask capped with stainless steel closures. Test media preparation methods were not described in detail. Solvent concentration used was not reported.
	Metric 8:	Consistency of Exposure Administration	Low	Reporting omissions (exposure concentrations, solvent concentration, concurrent use of controls, etc.) are likely to have a substantial impact on results.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were measured but not reported.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported (72 hr) and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Uninformative	No information is provided on the number of exposure groups and spacing of exposure levels.
	Metric 12:	Testing at or Below Solubility Limit	Low	The solvent concentration in itself was not reported but inferred appropriate ((i.e., no effects on biological responses were observed in the solvent control and no interactions were expected between the solvent and test substance). However, no information was provided on the number of exposure groups, so it is difficult to determine if the solvent concentration used was appropriate to dissolve all the exposure concentrations.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	S. costatum was obtained from the culture collection of the University of Rhode Island. It was not stated whether the inoculum culture was in the exponential growth phase, which is recommended for algal toxicity assays (OECD 201).
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized and whether pretreatment conditions were the same for control and exposed groups.

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Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
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Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Low	The initial biomass used in the test cultures was not provided. It was stated that each test was replicated.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reporting of environmental conditions and biomass loading was not sufficient to evaluate if adequate and whether differences occurred between control and exposed populations.
	Metric 17:	Outcome Assessment Methodology	Medium	The outcome assessment methodology reported the intended outcome of interest. Cell counts were done using a hemacytometer after 72 hours of exposure in treatment groups. However, it was unclear whether cell counts in controls were done at the same time as the treatment groups.
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment of treatment groups and controls were limited.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. In the discussion section, the authors report that pH of the growth media may have played a role in the observed toxicity but pH was only measured before the test and not during or after the test.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure (e.g., infection) that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately. No statistical test was conducted to see whether algal density in treatment groups were significantly different from controls. Methods for EC50 determination was from an in press report.
	Metric 22:	Reporting of Data	Low	Algal density data for exposure-related findings were not shown for each treatment and control group. EC 50 values were provided for each growth media test without confidence intervals (Table 3a).
	Metric 23:	Explanation of Unexpected Outcomes	Low	Insufficient information was provided to determine if excessive variability or unexpected outcomes occurred.

Additional Comments: The number of exposure groups, exposure concentrations, solvent concentration used and algal population density data for treatment groups and controls were not provided. EC50 values were calculated using an unpublished method and presented without confidence intervals.

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

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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 reported but the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Negative and solvent controls were used (algal growth of controls reported in Table 5) but unclear whether these were tested concurrently.
	Metric 5:	Negative Control Response	Medium	The biological responses of the negative control groups were reported (Table 5) but unclear whether the controls were tested concurrently with the treatment groups.
	Metric 6:	Randomized Allocation	Low	Algal study, randomized allocation not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	Experiments were conducted in Delong culture flask capped with stainless steel closures. Test media preparation methods were not described in detail. Solvent concentration used was not reported.
	Metric 8:	Consistency of Exposure Administration	Low	Reporting omissions (exposure concentrations, solvent concentration, concurrent use of controls, etc.) are likely to have a substantial impact on results.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were measured but not reported.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported (72 hr) and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Uninformative	No information is provided on the number of exposure groups and spacing of exposure levels.
	Metric 12:	Testing at or Below Solubility Limit	Low	The solvent concentration in itself was not reported but inferred appropriate ((i.e., no effects on biological responses were observed in the solvent control and no interactions were expected between the solvent and test substance). However, no information was provided on the number of exposure groups, so it is difficult to determine if the solvent concentration used was appropriate to dissolve all the exposure concentrations.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	S. costatum was obtained from the culture collection of the University of Rhode Island. It was not stated whether the inoculum culture was in the exponential growth phase, which is recommended for algal toxicity assays (OECD 201).
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized and whether pretreatment conditions were the same for control and exposed groups.

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Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Low	The initial biomass used in the test cultures was not provided. It was stated that each test was replicated.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reporting of environmental conditions and biomass loading was not sufficient to evaluate if adequate and whether differences occurred between control and exposed populations.
	Metric 17:	Outcome Assessment Methodology	Medium	The outcome assessment methodology reported the intended outcome of interest. Cell counts were done using a hemacytometer after 72 hours of exposure in treatment groups. However, it was unclear whether cell counts in controls were done at the same time as the treatment groups.
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment of treatment groups and controls were limited.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. In the discussion section, the authors report that pH of the growth media may have played a role in the observed toxicity but pH was only measured before the test and not during or after the test.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure (e.g., infection) that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately. No statistical test was conducted to see whether algal density in treatment groups were significantly different from controls. Methods for EC50 determination was from an in press report.
	Metric 22:	Reporting of Data	Low	Algal density data for exposure-related findings were not shown for each treatment and control group. EC 50 values were provided for each growth media test without confidence intervals (Table 3a).
	Metric 23:	Explanation of Unexpected Outcomes	Low	Insufficient information was provided to determine if excessive variability or unexpected outcomes occurred.

Additional Comments: The number of exposure groups, exposure concentrations, solvent concentration used and algal population density data for treatment groups and controls were not provided. EC 50 values were calculated using an unpublished method and presented without confidence intervals.

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

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Taxa, Species, Age:	Vegetation; Non-vascular Plants; *Thalassiosira pseudonana*; Hasle and Heindal; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
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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 reported but the test substance identity was not analytically verified by the performing laboratory.
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Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Negative and solvent controls were used (algal growth of controls reported in Table 5) but unclear whether these were tested concurrently.
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	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported (72 hr) and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Uninformative	No information is provided on the number of exposure groups and spacing of exposure levels.
	Metric 12:	Testing at or Below Solubility Limit	Low	The solvent concentration in itself was not reported but inferred appropriate ((i.e., no effects on biological responses were observed in the solvent control and no interactions were expected between the solvent and test substance). However, no information was provided on the number of exposure groups, so it is difficult to determine if the solvent concentration used was appropriate to dissolve all the exposure concentrations.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	T. pseudonana was obtained from the culture collection of the Woods Hole Oceanographic Institution. It was not stated whether the inoculum culture was in the exponential growth phase, which is recommended for algal toxicity assays (OECD 201).

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Domain	Metric		Rating	Comments
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	Metric 15:	Number of Organisms and Replicates per Group	Low	The initial biomass used in the test cultures was not provided. It was stated that each test was replicated.
Domain 5: Outcome Assessment				
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	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment of treatment groups and controls were limited.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. In the discussion section, the authors report that pH of the growth media may have played a role in the observed toxicity but pH was only measured before the test and not during or after the test.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure (e.g., infection) that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately. No statistical test was conducted to see whether algal density in treatment groups were significantly different from controls. Methods for EC50 determination was from an in press report.
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Domain	Metric	Rating	Comments
Additional Comments:	The number of exposure groups, exposure concentrations, solvent concentration used and algal population density data for treatment groups and controls were not provided. EC50 values were calculated using an unpublished method and presented without confidence intervals.		

Overall Quality Determination — Uninformative

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Taxa, Species, Age:	Vegetation; Non-vascular Plants; *Thalassiosira pseudonana*; Hasle and Heindal; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1927837

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

Study Citation:	Walsh, G. E., Yoder, M. J., Mclaughlin, L. L., Lores, E. M. (1987). Responses of marine unicellular algae to brominated organic compounds in six growth media. Ecotoxicology and Environmental Safety 14(3):215-222.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (marine); Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Non-vascular Plants; *Thalassiosira pseudonana*; Hasle and Heindal; Not Applicable (e.g., fungi or algae studies) or Not Reported
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Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1927837

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 reported but the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Negative and solvent controls were used (algal growth of controls reported in Table 5) but unclear whether these were tested concurrently.
	Metric 5:	Negative Control Response	Medium	The biological responses of the negative control groups were reported (Table 5), but it was unclear whether the controls were tested concurrently with the treatment groups.
	Metric 6:	Randomized Allocation	Low	Algal study, randomized allocation not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	Experiments were conducted in Delong culture flask capped with stainless steel closures. Test media preparation methods were not described in detail. Solvent concentration used was not reported.
	Metric 8:	Consistency of Exposure Administration	Low	Reporting omissions (exposure concentrations, solvent concentration, concurrent use of controls, etc.) were likely to have a substantial impact on results.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were measured but not reported.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported (72 hr) and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Uninformative	No information is provided on the number of exposure groups and spacing of exposure levels.
	Metric 12:	Testing at or Below Solubility Limit	Low	The solvent concentration in itself was not reported but inferred appropriate ((i.e., no effects on biological responses were observed in the solvent control and no interactions were expected between the solvent and test substance). However, no information was provided on the number of exposure groups, so it is difficult to determine if the solvent concentration used was appropriate to dissolve all the exposure concentrations.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	T. pseudonana was obtained from the culture collection of the Woods Hole Oceanographic Institution. It was not stated whether the inoculum culture was in the exponential growth phase, which is recommended for algal toxicity assays (OECD 201).
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized and whether pretreatment conditions were the same for control and exposed groups.

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Taxa, Species, Age:	Vegetation; Non-vascular Plants; *Thalassiosira pseudonana*; Hasle and Heindal; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1927837

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Low	The initial biomass used in the test cultures was not provided. It was stated that each test was replicated.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reporting of environmental conditions and biomass loading was not sufficient to evaluate if adequate and whether differences occurred between control and exposed populations.
	Metric 17:	Outcome Assessment Methodology	Medium	The outcome assessment methodology reported the intended outcome of interest. Cell counts were done using a hemacytometer after 72 hours of exposure in treatment groups. However, it was unclear whether cell counts in controls were done at the same time as the treatment groups.
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment of treatment groups and controls were limited.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. In the discussion section, the authors report that pH of the growth media may have played a role in the observed toxicity but pH was only measured before the test and not during or after the test.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure (e.g., infection) that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately. No statistical test was conducted to see whether algal density in treatment groups were significantly different from controls. Methods for EC50 determination was from an in press report.
	Metric 22:	Reporting of Data	Low	Algal density data for exposure-related findings were not shown for each treatment and control group. EC50 values were provided for each growth media test without confidence intervals (Table 3a).
	Metric 23:	Explanation of Unexpected Outcomes	Low	Insufficient information was provided to determine if excessive variability or unexpected outcomes occurred.

Additional Comments: The number of exposure groups, exposure concentrations, solvent concentration used and algal population density data for treatment groups and controls were not provided. EC50 values were calculated using an unpublished method and presented without confidence intervals.

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

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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 reported but the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Clean and solvent controls were used (algal growth of controls reported in Table 5), but it was unclear whether these were tested concurrently.
	Metric 5:	Negative Control Response	Medium	The biological responses of the negative control groups were reported (Table 5), but it was unclear whether the controls were tested concurrently with the treatment groups.
	Metric 6:	Randomized Allocation	Low	Algal study, randomized allocation not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	Experiments were conducted in Delong culture flask capped with stainless steel closures. Test media preparation methods were not described in detail. Solvent concentration used was not reported.
	Metric 8:	Consistency of Exposure Administration	Low	Reporting omissions (exposure concentrations, solvent concentration, concurrent use of controls, etc.) were likely to have a substantial impact on results.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were measured but not reported.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported (72 hr) and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Uninformative	No information is provided on the number of exposure groups and spacing of exposure levels.
	Metric 12:	Testing at or Below Solubility Limit	Low	The solvent concentration in itself was not reported but inferred appropriate ((i.e., no effects on biological responses were observed in the solvent control and no interactions were expected between the solvent and test substance). However, no information was provided on the number of exposure groups, so it is difficult to determine if the solvent concentration used was appropriate to dissolve all the exposure concentrations.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	T. pseudonana was obtained from the culture collection of the Woods Hole Oceanographic Institution. It was not stated whether the inoculum culture was in the exponential growth phase, which is recommended for algal toxicity assays (OECD 201).
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized and whether pretreatment conditions were the same for control and exposed groups.

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Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1927837

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Low	The initial biomass used in the test cultures was not provided. It was stated that each test was replicated.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reporting of environmental conditions and biomass loading was not sufficient to evaluate if adequate and whether differences occurred between control and exposed populations.
	Metric 17:	Outcome Assessment Methodology	Medium	The outcome assessment methodology reported the intended outcome of interest. Cell counts were done using a hemacytometer after 72 hours of exposure in treatment groups. However, it was unclear whether cell counts in controls were done at the same time as the treatment groups.
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment of treatment groups and controls were limited.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. In the discussion section, the authors report that pH of the growth media may have played a role in the observed toxicity but pH was only measured before the test and not during or after the test.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure (e.g., infection) that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately. No statistical test was conducted to see whether algal density in treatment groups were significantly different from controls. Methods for EC50 determination was from an in press report.
	Metric 22:	Reporting of Data	Low	Algal density data for exposure-related findings were not shown for each treatment and control group. EC50 values were provided for each growth media test without confidence intervals (Table 3a).
	Metric 23:	Explanation of Unexpected Outcomes	Low	Insufficient information was provided to determine if excessive variability or unexpected outcomes occurred.

Additional Comments: The number of exposure groups, exposure concentrations, solvent concentration used and algal population density data for treatment groups and controls were not provided. EC50 values were calculated using an unpublished method and presented without confidence intervals.

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

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Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
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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 reported but the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Clean and solvent controls were used (algal growth of controls reported in Table 5), but it was unclear whether these were tested concurrently.
	Metric 5:	Negative Control Response	Medium	The biological responses of the negative control groups were reported (Table 5) but unclear whether the controls were tested concurrently with the treatment groups.
	Metric 6:	Randomized Allocation	Low	Algal study, randomized allocation not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	Experiments were conducted in Delong culture flask capped with stainless steel closures. Test media preparation methods were not described in detail. Solvent concentration used was not reported.
	Metric 8:	Consistency of Exposure Administration	Low	Reporting omissions (exposure concentrations, solvent concentration, concurrent use of controls, etc.) were likely to have a substantial impact on results.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were measured but not reported.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported (72 hr) and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Uninformative	No information is provided on the number of exposure groups and spacing of exposure levels.
	Metric 12:	Testing at or Below Solubility Limit	Low	The solvent concentration in itself was not reported but inferred appropriate ((i.e., no effects on biological responses were observed in the solvent control and no interactions were expected between the solvent and test substance). However, no information was provided on the number of exposure groups, so it is difficult to determine if the solvent concentration used was appropriate to dissolve all the exposure concentrations.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	T. pseudonana was obtained from the culture collection of the Woods Hole Oceanographic Institution. It was not stated whether the inoculum culture was in the exponential growth phase, which is recommended for algal toxicity assays (OECD 201).
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized and whether pretreatment conditions were the same for control and exposed groups.

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Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Low	The initial biomass used in the test cultures was not provided. It was stated that each test was replicated.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reporting of environmental conditions and biomass loading was not sufficient to evaluate if adequate and whether differences occurred between control and exposed populations.
	Metric 17:	Outcome Assessment Methodology	Medium	The outcome assessment methodology reported the intended outcome of interest. Cell counts were done using a hemacytometer after 72 hours of exposure in treatment groups. However, it was unclear whether cell counts in controls were done at the same time as the treatment groups.
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment of treatment groups and controls were limited.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups. In the discussion section, the authors report that pH of the growth media may have played a role in the observed toxicity but pH was only measured before the test and not during or after the test.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure (e.g., infection) that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately. No statistical test was conducted to see whether algal density in treatment groups were significantly different from controls. Methods for EC50 determination was from an in press report.
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Overall Quality Determination		**Uninformative**	

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	Metric 12:	Testing at or Below Solubility Limit	Low	The solvent concentration in itself was not reported but inferred appropriate ((i.e., no effects on biological responses were observed in the solvent control and no interactions were expected between the solvent and test substance). However, no information was provided on the number of exposure groups, so it is difficult to determine if the solvent concentration used was appropriate to dissolve all the exposure concentrations.
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Domain	Metric	Rating	Comments
Overall Quality Determination		**Uninformative**	

Study Citation:	Ye, G., Chen, Y., Wang, H. O., Ye, T., Lin, Y., Huang, Q., Chi, Y., Dong, S. (2016). Metabolomics approach reveals metabolic disorders and potential biomarkers associated with the developmental toxicity of tetrabromobisphenol A and tetrachlorobisphenol A. Scientific Reports 6:35257.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555779

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only, and no CASRN or other structural descriptors were reported.
	Metric 2:	Test Substance Source	Low	The source of the test substance was reported (Tokyo Chemical Industry, Tokyo, Japan), but the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity was reported as 97%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Metabolomics (F0 exposed, F1 not exposed): Authors reported using a concurrent control (methods, p. 9). However, the authors did not explicitly indicate if the control was a solvent control (0.2% DMSO) or if the control was in reality a negative control.
	Metric 5:	Negative Control Response	High	Metabolomics (F0 exposed, F1 not exposed): The biological response of the negative control group was reported and reasonable given the clustering pattern of the control group.
	Metric 6:	Randomized Allocation	Low	Researchers did not report if organisms were randomly allocated to exposure groups of the metabolomics assessment.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	Metabolomics (F0 exposed, F1 not exposed): The type of experimental system and test media preparation methods were not described in detail and was hard to know if the authors used the same system as for the developmental toxicity endpoints.
	Metric 8:	Consistency of Exposure Administration	Low	Metabolomics (F0 exposed, F1 not exposed): The study provided few details on exposure administration.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	Medium	Metabolomics (F0 exposed, F1 not exposed): The duration of exposure (2 to 6 dpf = 4 days) was reported and appropriate for assessing metabolomic changes.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	The number of exposure groups and spacing was reported (0, 50, and 200 μ g/L TBBPA). It was unclear what the rationale for the spacing of the groups was.
	Metric 12:	Testing at or Below Solubility Limit	High	The exposure concentrations were 0, 50, and 200 μ g/L TBBPA; below the water solubility limit (4.15 mg/L at 25 C, pH 7.56). In addition, authors used a solvent (0.2% DMSO), and the exposure was conducted in artificial seawater (3%), so, under these conditions solubility of TBBPA was expected to be higher than 4.15 mg/L.

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Study Citation:	Ye, G., Chen, Y., Wang, H. O., Ye, T., Lin, Y., Huang, Q., Chi, Y., Dong, S. (2016). Metabolomics approach reveals metabolic disorders and potential biomarkers associated with the developmental toxicity of tetrabromobisphenol A and tetrachlorobisphenol A. Scientific Reports 6:35257.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555779

Domain	Metric		Rating	Comments
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The test organisms (marine medaka, Oryzias melastigma, embryos) were adequately described, but it was unclear if embryos were from an established laboratory culture or from another source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	There was no evidence to suggest that pretreatment conditions were different across groups.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The authors reported in the 'Sample Preparation' methods (p. 9) that twelve offspring embryos in each replicate were used for metabolomic analysis. However, a different number of organisms was used for various specific metabolic assessments: Figures 1, 2, 3, and 4a showed n = 4; Figure 4c and d showed n = 12; Figure 5c showed n = 8.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology (metabolomics based on gas chromatography-mass spectrometry) addressed the intended outcome (molecular metabolic disorder and nervous system impacts of TBBPA).
	Metric 18:	Consistency of Outcome Assessment	High	Metabolomics (F0 exposed, F1 not exposed): Details regarding the execution of the study protocol for outcome assessment were reported.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	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 to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Metabolomics (F0 exposed, F1 not exposed): Statistical methods were described (non-parametric test, two-tailed Mann-Whitney U test).
	Metric 22:	Reporting of Data	High	Metabolomics (F0 exposed, F1 not exposed): Data for exposure-related findings were presented for each treatment and control group (Figures 1-4 and Figures S3-S10).
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: The goal of the study was to investigate the effects of TBBPA on hatching, development (morphology), and global metabolism and nervous system assessed by a metabolomics analysis in marine medaka (Oryzias melastigma) embryos. This forms was used to evaluate metabolomics of F0 exposed to TBBPA & F1 not exposed to TBBPA. It was unclear how long the whole study was because the authors referred to F0 and F1 but did not indicate if the F1 was generated when F0 reached sexual maturity (~4 months post hatched) or later.

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Study Citation:	Ye, G., Chen, Y., Wang, H. O., Ye, T., Lin, Y., Huang, Q., Chi, Y., Dong, S. (2016). Metabolomics approach reveals metabolic disorders and potential biomarkers associated with the developmental toxicity of tetrabromobisphenol A and tetrachlorobisphenol A. Scientific Reports 6:35257.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555779

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

Study Citation:	Ye, G., Chen, Y., Wang, H. O., Ye, T., Lin, Y., Huang, Q., Chi, Y., Dong, S. (2016). Metabolomics approach reveals metabolic disorders and potential biomarkers associated with the developmental toxicity of tetrabromobisphenol A and tetrachlorobisphenol A. Scientific Reports 6:35257.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Embryo
Health Outcome:	Cardiovascular
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555779

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only, and no CASRN or other structural descriptors were reported.
	Metric 2:	Test Substance Source	Low	The source of the test substance was reported (Tokyo Chemical Industry, Tokyo, Japan), but the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity was reported as 97%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Heart rate: Authors reported using a concurrent control (Figure 4b), but details of the use of a control group were not offered in the exposure methodology section for this outcome (last paragraph of page 8). In addition, because details were not offered, the authors did not explicitly indicate if the control was a solvent control (DMSO) or if the control was in reality a negative control.
	Metric 5:	Negative Control Response	High	Heart rate: The biological response of the negative control group was reported and adequate (Figure 4b).
	Metric 6:	Randomized Allocation	Low	Heart rate: Authors reported that embryos were randomly selected and cultured in 90-mm Petri dishes with 20-mL artificial seawater (page 8).
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	Heart rate: Authors reported preparing TBBPA in DMSO to prepare the stock solution and that the final DMSO concentration in artificial seawater was 0.2%. They also reported that 20-mL artificial seawater was added to 90-mm Petri dishes for embryos or 150-mL artificial seawater to 300-mL glass containers for larvae and that culture liquid was refreshed daily. However, the authors did not offer a rationale for the use of 0.2% DMSO. For example, OECD Test 236 Fish Embryo Acute Toxicity (FET) Test or OCSPP 850.1075 recommends that the final concentration of a solvent should not exceed 100 µl/L or 0.01%.
	Metric 8:	Consistency of Exposure Administration	Low	The was no evidence to suggest that the treatment concentrations were consistently administered across groups. But it was unclear if the control group received DMSO alone.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	Heart rate: The duration of exposure (from 2 to 10 dpf = 8 day exposure) was reported in the methods (p. 8) and appropriate for assessing heart rate impacts.

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Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Embryo
Health Outcome:	Cardiovascular
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555779

Domain	Metric		Rating	Comments
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Heart rate: The number of exposure groups and spacing was reported for a dose response (0, 50, 200 and 800 μ g/L TBBPA). However, the spacing factor was 4 and exceeded recommended spacing factors of test guidelines for the embryonic or aquatic toxicity studies (e.g., OECD Test 236 or OCSPP 850.1000).
	Metric 12:	Testing at or Below Solubility Limit	High	The exposure concentrations were 0, 50, 200 and 800 μ g/L TBBPA; below the water solubility limit. In addition, authors used a solvent (0.2% DMSO), and the exposure was conducted in artificial seawater (3%), so, under these conditions solubility of TBBPA was expected to be higher than 4.15 mg/L at 25 C, pH 7.56.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The test organisms (marine medaka, Oryzias melastigma, embryos) were adequately described, but it was unclear if embryos were from an established laboratory culture or from another source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Heart rate: Authors reported that synchronized fertilized embryos were obtained within 2 hours after the initiation of photophase, and fertilization was confirmed using a dissecting microscope. Embryos were exposed to TBBPA from 2 to 10 dpf (days post fertilization). There is no evidence to indicate that embryos were not treated under the same pretreatment conditions.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Heart rate: In the methods, the authors reported that the numbers of test organisms was 35 embryos and that the exposure was three replicates. However, Figure 4b indicated that number of embryos per group was 67–80.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Heart rate: 35 embryos were cultured in 90-mm Petri dishes with 20-mL artificial seawater and newly hatched larvae were transferred to a 300-mL glass break added with 150-mL artificial seawater. The culture liquid was refreshed daily. Marine medaka embryos hatch around 10 dpf, so exogenous feeding had not started and external feeding was unnecessary. However, it was unclear if the embryos and larvae were kept at the same temperature and salinity conditions as the adults (cultured in 28 $\pm$ 1 °C, 3% salinity).
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology (heart beat counts of embryos for 60 s) addressed the intended outcome (heart rate impacts). Figure 4b indicated that number of embryos assessed was 67–80.
	Metric 18:	Consistency of Outcome Assessment	High	Heart rate: There was no evidence to indicate that the outcome assessment methodology was not employed consistently across groups.
Domain 6: Confounding / Variable Control				

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Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Embryo
Health Outcome:	Cardiovascular
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555779

Domain	Metric		Rating	Comments
	Metric 19:	Confounding Variables in Test Design and Procedures	Medium	Heart rate: There were no reported differences among the study groups in environmental conditions (e.g., number of organisms per experimental unit), however, authors did not clarify other factors (e.g., temperature) that could influence the outcome assessment.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Heart rate: Statistical methods were described (nonparametric test, two-tailed Mann-Whitney U test).
	Metric 22:	Reporting of Data	Medium	Heart rate: Data were presented for 0, 50, and 200 ug/L (Figure 4b). It was unclear why the 800 ug/L group was not assessed.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: The goal of the study was to investigate the effects of TBBPA on hatching, development (morphology), and global metabolism and nervous system assessed by a metabolomics analysis in marine medaka (Oryzias melastigma) embryos. This forms was used to evaluate heart rate.

Overall Quality Determination Medium

Study Citation:	Ye, G., Chen, Y., Wang, H. O., Ye, T., Lin, Y., Huang, Q., Chi, Y., Dong, S. (2016). Metabolomics approach reveals metabolic disorders and potential biomarkers associated with the developmental toxicity of tetrabromobisphenol A and tetrachlorobisphenol A. Scientific Reports 6:35257.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555779

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only, and no CASRN or other structural descriptors were reported.
	Metric 2:	Test Substance Source	Low	The source of the test substance was reported (Tokyo Chemical Industry, Tokyo, Japan), but the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity was reported as 97%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Metabolomics (F0 not exposed, F1 exposed): Authors reported using a concurrent control (methods, p. 9). However, the authors did not explicitly indicate if the control was a solvent control (0.2% DMSO) or if the control was in reality a negative control.
	Metric 5:	Negative Control Response	High	Metabolomics (F0 not exposed, F1 exposed): The biological response of the negative control group was reported and reasonable given the clustering pattern of the control group.
	Metric 6:	Randomized Allocation	Low	Researchers did not report if organisms were randomly allocated to exposure groups of the metabolomics assessment.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	Metabolomics (F0 not exposed, F1 exposed): The type of experimental system and test media preparation methods were not described in detail and was hard to know if the authors used the same system as for the developmental toxicity endpoints.
	Metric 8:	Consistency of Exposure Administration	Low	Metabolomics (F0 not exposed, F1 exposed): The study provided few details on exposure administration.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	Medium	Metabolomics (F0 not exposed, F1 exposed): The duration of exposure (2 to 6 dpf = 4 days) was reported and appropriate for assessing metabolomic changes.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	The number of exposure groups and spacing was reported (0, 50, and 200 μ g/L TBBPA). It was unclear what the rationale for the spacing of the groups was.
	Metric 12:	Testing at or Below Solubility Limit	High	The exposure concentrations were 0, 50, and 200 μ g/L TBBPA; below the water solubility limit (4.15 mg/L at 25 C, pH 7.56). In addition, authors used a solvent (0.2% DMSO), and the exposure was conducted in artificial seawater (3%), so, under these conditions solubility of TBBPA was expected to be higher than 4.15 mg/L.
Domain 4: Test Organism				

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Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555779

Domain	Metric		Rating	Comments
	Metric 13:	Test Organism Characteristics	Medium	The test organisms (marine medaka, Oryzias melastigma, embryos) were adequately described, but it was unclear if embryos were from an established laboratory culture or from another source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	There was no evidence to suggest that pretreatment conditions were different across groups.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The authors reported in the 'Sample Preparation' methods (p. 9) that twelve offspring embryos in each replicate were used for metabolomic analysis. However, a different number of organisms was used for various specific metabolic assessments: Figures 1, 2, 3, and 4a showed n = 4; Figure 4c and d showed n = 12; Figure 5c showed n = 8.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology (metabolomics based on gas chromatography-mass spectrometry) addressed the intended outcome (molecular metabolic disorder and nervous system impacts of TBBPA).
	Metric 18:	Consistency of Outcome Assessment	High	Metabolomics (F0 not exposed, F1 exposed): Details regarding the execution of the study protocol for outcome assessment were reported.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	High	There was no information to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Metabolomics (F0 not exposed, F1 exposed): Statistical methods were described (non-parametric test, two-tailed Mann-Whitney U test).
	Metric 22:	Reporting of Data	High	Metabolomics (F0 not exposed, F1 exposed): Data for exposure-related findings were presented for each treatment and control group (Figures 1-4 and Figures S3-S10).
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: The goal of the study was to investigate the effects of TBBPA on hatching, development (morphology), and global metabolism and nervous system assessed by a metabolomics analysis in marine medaka (Oryzias melastigma) embryos. This forms was used to evaluate metabolomics of F0 not exposed to TBBPA & F1 exposed to TBBPA. It was unclear how long the whole study was because the authors referred to F0 and F1 but did not indicate if the F1 was generated when F0 reached sexual maturity (~4 months post hatched) or later.

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Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555779

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

Study Citation:	Ye, G., Chen, Y., Wang, H. O., Ye, T., Lin, Y., Huang, Q., Chi, Y., Dong, S. (2016). Metabolomics approach reveals metabolic disorders and potential biomarkers associated with the developmental toxicity of tetrabromobisphenol A and tetrachlorobisphenol A. Scientific Reports 6:35257.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555779

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only, and no CASRN or other structural descriptors were reported.
	Metric 2:	Test Substance Source	Low	The source of the test substance was reported (Tokyo Chemical Industry, Tokyo, Japan), but the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity was reported as 97%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Developmental toxicology/morphological changes: Authors reported using a concurrent control (Figure 1S b & c), but details of the use of a control group were not offered in the exposure methodology section for this outcome (last paragraph of page 8). In addition, because details were not offered, the authors did not explicitly indicate if the control was a solvent control (DMSO) or if the control was in reality a negative control.
	Metric 5:	Negative Control Response	High	Developmental toxicology/morphological changes: The biological response of the negative control group was reported and adequate (Figure 1S b & c).
	Metric 6:	Randomized Allocation	Medium	Developmental toxicology/morphological changes: Authors reported that embryos were randomly selected andcultured in 90-mm Petri dishes with 20-mL artificial seawater (page 8).
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	Developmental toxicology/morphological changes: Authors reported preparing TBBPA in DMSO to prepare the stock solution and that the final DMSO concentration in artificial seawater was 0.2%. They also reported that 20-mL artificial seawater was added to 90-mm Petri dishes for embryos or 150-mL artificial seawater to 300-mL glass containers for larvae and that culture liquid was refreshed daily. However, the authors did not offer a rationale for the use of 0.2% DMSO. For example, OECD Test 236 Fish Embryo Acute Toxicity (FET) Test or OCSPP 850.1075 recommends that the final concentration of a solvent should not exceed 100 µl/L or 0.01%.
	Metric 8:	Consistency of Exposure Administration	Low	The was no evidence to suggest that the treatment concentrations were consistently administered across groups. But it was unclear if the control group received DMSO alone.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	Developmental toxicology/morphological changes: The duration of exposure (from 2 to 10 dpf = 8 day exposure) was reported in the methods (p. 8) and appropriate for assessing development and morphological changes.

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Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
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Domain	Metric		Rating	Comments
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Developmental toxicology/morphological changes: The number of exposure groups and spacing was reported for a dose response (0, 50, 200 and 800 μ g/L TBBPA). However, the spacing factor was 4 and exceeded recommended spacing factors of test guidelines for the embryonic or aquatic toxicity studies (e.g., OECD Test 236 or OCSPP 850.1000).
	Metric 12:	Testing at or Below Solubility Limit	High	The exposure concentrations were 0, 50, 200 and 800 μ g/L TBBPA; below the water solubility limit. In addition, authors used a solvent (0.2% DMSO), and the exposure was conducted in artificial seawater (3%), so, under these conditions solubility of TBBPA was expected to be higher than 4.15 mg/L at 25 C, pH 7.56.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The test organisms (marine medaka, Oryzias melastigma, embryos) were adequately described, but it was unclear if embryos were from an established laboratory culture or from another source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Developmental toxicology/morphological changes: Authors reported that synchronized fertilized embryos were obtained within 2 hours after the initiation of photophase, and fertilization was confirmed using a dissecting microscope. Embryos were exposed to TBBPA from 2 to 10 dpf (days post fertilization). There is no evidence to indicate that embryos were not treated under the same pretreatment conditions.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Developmental toxicology/morphological changes: The numbers of test organisms (35 embryos) and replicates (three) were reported and sufficient.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Developmental toxicology/morphological changes: 35 embryos were cultured in 90-mm Petri dishes with 20-mL artificial seawater and newly hatched larvae were transferred to a 300-mL glass break added with 150-mL artificial seawater. The culture liquid was refreshed daily. Marine medaka embryos hatch around 10 dpf, so exogenous feeding had not started and external feeding was unnecessary. However, it was unclear if the embryos and larvae were kept at the same temperature and salinity conditions as the adults (cultured in 28 $\pm$ 1 °C, 3% salinity).
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology (microscopic observation of pericardial and abdominal edemas, marked with arrows in Figure 1S c) addressed the intended outcome (morphological changes).
	Metric 18:	Consistency of Outcome Assessment	High	Developmental toxicology/morphological changes: There was no evidence to indicate that the outcome assessment methodology was not employed consistently across groups.
Domain 6: Confounding / Variable Control				

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Study Citation:	Ye, G., Chen, Y., Wang, H. O., Ye, T., Lin, Y., Huang, Q., Chi, Y., Dong, S. (2016). Metabolomics approach reveals metabolic disorders and potential biomarkers associated with the developmental toxicity of tetrabromobisphenol A and tetrachlorobisphenol A. Scientific Reports 6:35257.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555779

Domain	Metric		Rating	Comments
	Metric 19:	Confounding Variables in Test Design and Procedures	Medium	Developmental toxicology/morphological changes: There were no reported differences among the study groups in environmental conditions (e.g., number of organisms per experimental unit), however, authors did not clarify other factors (e.g., temperature) that could influence the outcome assessment.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Uninformative	Developmental toxicology/morphological changes: Statistical analysis was not conducted for developmental endpoints. Authors only provided representative photographs of an embryo per treatment group (Figure 1S b, c). Moreover, incidence of morphological changes (i.e., pericardial and abdominal edemas) across treatment groups was not reported.
	Metric 22:	Reporting of Data	Low	Developmental toxicology/morphological changes: Authors only provided representative photographs of an embryo per treatment group (Figure 1S c). Moreover, incidence of morphological changes (i.e., pericardial and abdominal edemas) across treatment groups was not reported.
	Metric 23:	Explanation of Unexpected Outcomes	Low	Developmental toxicology/morphological changes: The study did not report any measures of variability or quantify results.

Additional Comments: The goal of the study was to investigate the effects of TBBPA on hatching, development (morphology), and global metabolism and nervous system assessed by a metabolomics analysis in marine medaka (Oryzias melastigma) embryos. This forms was used to evaluate developmental toxicology/morphological changes.

Overall Quality Determination — Uninformative

Study Citation:	Ye, G., Chen, Y., Wang, H. O., Ye, T., Lin, Y., Huang, Q., Chi, Y., Dong, S. (2016). Metabolomics approach reveals metabolic disorders and potential biomarkers associated with the developmental toxicity of tetrabromobisphenol A and tetrachlorobisphenol A. Scientific Reports 6:35257.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555779

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only, and no CASRN or other structural descriptors were reported.
	Metric 2:	Test Substance Source	Low	The source of the test substance was reported (Tokyo Chemical Industry, Tokyo, Japan), but the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity was reported as 97%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Hatchability as surrogate for mortality: Authors reported using a concurrent control (Figure 1Sd), but details of the use of a control group were not offered in the exposure methodology section for this outcome (last paragraph of page 8). In addition, because details were not offered, the authors did not explicitly indicate if the control was a solvent control (DMSO) or if the control was in reality a negative control.
	Metric 5:	Negative Control Response	High	Hatchability as surrogate for mortality: The biological response (percent hatching) of the solvent control group was reported and appropriate (87% hatchability, Figure 1Sd). For example, OECD Test 236 Fish Embryo Acute Toxicity (FET) Test which is a 96 hour test recommends >80% hatchability in the control group.
	Metric 6:	Randomized Allocation	Medium	Authors reported that embryos were randomly selected andcultured in 90-mm Petri dishes with 20-mL artificial seawater (page 8).
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	Hatchability as surrogate for mortality: Authors reported preparing TBBPA in DMSO to prepare the stock solution and that the final DMSO concentration in artificial seawater was 0.2%. They also reported that 20-mL artificial seawater was added to 90-mm Petri dishes for embryos or 150-mL artificial seawater to 300-mL glass containers for larvae and that culture liquid was refreshed daily. However, the authors did not offer a rationale for the use of 0.2% DMSO. For example, OECD Test 236 Fish Embryo Acute Toxicity (FET) Test recommends that the final concentration of a solvent should not exceed 100 µl/L or 0.01%.
	Metric 8:	Consistency of Exposure Administration	Low	The was no evidence to suggest that the treatment concentrations were consistently administered across groups. But it was unclear if the control group received DMSO alone.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	Hatchability as surrogate for mortality: The duration of exposure (from 2 to 10 dpf = 8 day exposure) was reported in the methods (p. 8) and appropriate for assessing hatchability and developmental endpoints

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Study Citation:	Ye, G., Chen, Y., Wang, H. O., Ye, T., Lin, Y., Huang, Q., Chi, Y., Dong, S. (2016). Metabolomics approach reveals metabolic disorders and potential biomarkers associated with the developmental toxicity of tetrabromobisphenol A and tetrachlorobisphenol A. Scientific Reports 6:35257.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555779

Domain	Metric		Rating	Comments
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	Hatchability as surrogate for mortality: The number of exposure groups and spacing was reported for a dose response (0, 50, 200 and 800 μ g/L TBBPA). However, the spacing factor was 4 and exceeded recommended spacing factors of test guidelines for the embryonic or aquatic toxicity studies (e.g., OECD Test 236 or OCSPP 850.1000).
	Metric 12:	Testing at or Below Solubility Limit	High	The exposure concentrations were 0, 50, 200 and 800 μ g/L TBBPA; below the water solubility limit. In addition, authors used a solvent (0.2% DMSO), and the exposure was conducted in artificial seawater (3%), so, under these conditions solubility of TBBPA was expected to be higher than 4.15 mg/L at 25 C, pH 7.56.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	The test organisms (marine medaka, Oryzias melastigma, embryos) were adequately described, but it was unclear if embryos were from an established laboratory culture or from another source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Hatchability as surrogate for mortality: Authors reported that synchronized fertilized embryos were obtained within 2 hours afterthe initiation of photophase, and fertilization was confirmed using a dissecting microscope. Embryos were exposed to TBBPA from 2 to 10 dpf (days post fertilization). There is no evidence to indicate that embryos were not treated under the same pretreatment conditions.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Hatchability as surrogate for mortality: The numbers of test organisms (35 embryos) and replicates (three) were reported and sufficient.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Hatchability as surrogate for mortality: 35 embryos were cultured in 90-mm Petri dishes with 20-mL artificial seawater and newly hatched larvae were transferred to a 300-mL glass break added with 150-mL artificial seawater. The culture liquid was refreshed daily. Marine medaka embryos hatch around 10 dpf, so exogenous feeding had not started and external feeding was unnecessary. However, it was unclear if the embryos and larvae were kept at the same temperature and salinity conditions as the adults (cultured in 28 $\pm$ 1 °C, 3% salinity).
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology (count of percent hatched embryos) addressed the intended outcome (hatchability as surrogate for mortality).
	Metric 18:	Consistency of Outcome Assessment	High	Hatchability as surrogate for mortality: There was not evidence to indicate that the outcome assessment methodology was not employed consistently across groups.
Domain 6: Confounding / Variable Control				

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Study Citation:	Ye, G., Chen, Y., Wang, H. O., Ye, T., Lin, Y., Huang, Q., Chi, Y., Dong, S. (2016). Metabolomics approach reveals metabolic disorders and potential biomarkers associated with the developmental toxicity of tetrabromobisphenol A and tetrachlorobisphenol A. Scientific Reports 6:35257.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Aquatic (freshwater), Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Oryzias melastigma*; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555779

Domain	Metric		Rating	Comments
	Metric 19:	Confounding Variables in Test Design and Procedures	Medium	Hatchability as surrogate for mortality: There were no reported differences among the study groups in environmental conditions (e.g., number of organisms per experimental unit), however, authors did not clarify other factors (e.g., temperature) that could influence the outcome assessment.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Hatchability as surrogate for mortality: Statistical methods were described. A non-parametric test (two-tailed Mann-Whitney U test) was performed using MATLAB 7.11 (MathWorks, USA).
	Metric 22:	Reporting of Data	High	Hatchability as surrogate for mortality: Data were presented for each treatment and control group in Supplementary Information (Figure S 1).
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: The goal of the study was to investigate the effects of TBBPA on hatching, development (morphology), and global metabolism and nervous system assessed by a metabolomics analysis in marine medaka (Oryzias melastigma) embryos. This forms was used to evaluate hatch and hatchability which is in this case was a surrogate for mortality (i.e., exposure was to embryos and authors reported number/percent of hatched embryos inferring that those embryos that did not hatch died).

Overall Quality Determination Medium

Study Citation:	Kuiper, R. V., Cantón, R. F., Leonards, P. E., Jenssen, B. M., Dubbeldam, M., Wester, P. W., Berg, v.d., M., Vos, J. G., Vethaak, A. D. (2007). Long-term exposure of European flounder (Platichthys flesus) to the flame-retardants tetrabromobisphenol A (TBBPA) and hexabromocyclododecane (HBCD). Ecotoxicology and Environmental Safety 67(3):349-360.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Platichthys flesus*; Juvenile
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Oxidative stress (including redox biology)-Endocrine toxicity
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1412802

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Identified by acronym, nomenclature, and analytically verified.
	Metric 2:	Test Substance Source	High	Obtained through Bromine Science and Environmental Forum (BSEF), and was analytically verified.
	Metric 3:	Test Substance Purity	High	Purity = 99.17%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Negative controls used
	Metric 5:	Negative Control Response	High	Control group responses were adequate.
	Metric 6:	Randomized Allocation	Low	Randomization of organism allocation to test groups were not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	Environmental system was well described and accounted for p-chem properties.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were consistent across groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Nominal concentrations used with continuous renewal of test chemical.
	Metric 10:	Exposure Duration and Frequency	High	Exposure duration appropriate with regard to species lifespan.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Number of exposure groups and spacing appopriate.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were above solubility limit. Additionally, the test water temperature was raised to 21 +/- 1 C to improve solubility.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Test organisms were well described.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Fish were acclimatized by raising the temperature of the stock aquariums to 18 C two weeks prior to exposure.
	Metric 15:	Number of Organisms and Replicates per Group	Low	n=10 per exposure group. Test group replicates were not reported.
Domain 5: Outcome Assessment				

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Study Citation:	Kuiper, R. V., Cantón, R. F., Leonards, P. E., Jenssen, B. M., Dubbeldam, M., Wester, P. W., Berg, v.d., M., Vos, J. G., Vethaak, A. D. (2007). Long-term exposure of European flounder (Platichthys flesus) to the flame-retardants tetrabromobisphenol A (TBBPA) and hexabromocyclododecane (HBCD). Ecotoxicology and Environmental Safety 67(3):349-360.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Platichthys flesus*; Juvenile
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Oxidative stress (including redox biology)-Endocrine toxicity
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1412802

Domain	Metric		Rating	Comments
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions were acceptable.
	Metric 17:	Outcome Assessment Methodology	High	Intended outcomes were reported.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	No confounding variables among groups were reported.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	no differences among groups were reported.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical approach well described.
	Metric 22:	Reporting of Data	High	All exposure related findings were reported.
	Metric 23:	Explanation of Unexpected Outcomes	High	No unexpected outcomes were reported.

Additional Comments: This form represents mechanistic aspects of the work outside of the ADME work. These outputs include: VTG (Biomarkers), Endo Tox (Thyroid hormones, aromatase), oxidative stress (EROD).

Overall Quality Determination High

Study Citation:	Kuiper, R. V., Cantón, R. F., Leonards, P. E., Jenssen, B. M., Dubbeldam, M., Wester, P. W., Berg, v.d., M., Vos, J. G., Vethaak, A. D. (2007). Long-term exposure of European flounder (Platichthys flesus) to the flame-retardants tetrabromobisphenol A (TBBPA) and hexabromocyclododecane (HBCD). Ecotoxicology and Environmental Safety 67(3):349-360.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Platichthys flesus*; Juvenile
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1412802

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Identified by acronym, nomenclature, and analytically verified.
	Metric 2:	Test Substance Source	High	Obtained through Bromine Science and Environmental Forum (BSEF), and was analytically verified.
	Metric 3:	Test Substance Purity	High	Purity = 99.17%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Negative controls used.
	Metric 5:	Negative Control Response	High	Control group responses were adequate.
	Metric 6:	Randomized Allocation	Low	Randomization of organism allocation to test groups were not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	Environmental system was well described and accounted for p-chem properties.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were consistent across groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Nominal concentrations used with continuous renewal of test chemical.
	Metric 10:	Exposure Duration and Frequency	High	Exposure duration appropriate with regard to species lifespan.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Number of exposure groups and spacing appopriate.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were above solubility limit. Additionally, the test water temperature was raised to 21 +/- 1 C to improve solubility.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Test organisms were well described.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Fish were acclimatized by raising the temperature of the stock aquariums to 18 C two weeks prior to exposure.
	Metric 15:	Number of Organisms and Replicates per Group	Low	n=10 per exposure group. Test group replicates were not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions were acceptable.
	Metric 17:	Outcome Assessment Methodology	High	Intended outcomes were reported.

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Study Citation:	Kuiper, R. V., Cantón, R. F., Leonards, P. E., Jenssen, B. M., Dubbeldam, M., Wester, P. W., Berg, v.d., M., Vos, J. G., Vethaak, A. D. (2007). Long-term exposure of European flounder (Platichthys flesus) to the flame-retardants tetrabromobisphenol A (TBBPA) and hexabromocyclododecane (HBCD). Ecotoxicology and Environmental Safety 67(3):349-360.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Platichthys flesus*; Juvenile
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1412802

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	No confounding variables among groups were reported.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	no differences among groups were reported.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical approach well described.
	Metric 22:	Reporting of Data	High	All exposure related findings were reported.
	Metric 23:	Explanation of Unexpected Outcomes	High	No unexpected outcomes were reported.

Additional Comments: This form represents ADME data provided Table 1, Figure 2 and 3.

Overall Quality Determination High

Study Citation:	Kuiper, R. V., Cantón, R. F., Leonards, P. E., Jenssen, B. M., Dubbeldam, M., Wester, P. W., Berg, v.d., M., Vos, J. G., Vethaak, A. D. (2007). Long-term exposure of European flounder (Platichthys flesus) to the flame-retardants tetrabromobisphenol A (TBBPA) and hexabromocyclododecane (HBCD). Ecotoxicology and Environmental Safety 67(3):349-360.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Platichthys flesus*; Juvenile
Health Outcome:	Hepatic/Liver
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1412802

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Identified by acronym, nomenclature, and analytically verified.
	Metric 2:	Test Substance Source	High	Obtained through Bromine Science and Environmental Forum (BSEF), and was analytically verified.
	Metric 3:	Test Substance Purity	High	Purity = 99.17%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Negative controls used
	Metric 5:	Negative Control Response	High	Control group responses were adequate.
	Metric 6:	Randomized Allocation	Low	Randomization of organism allocation to test groups were not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	Environmental system was well described and accounted for p-chem properties.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were consistent across groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Nominal concentrations used with continuous renewal of test chemical.
	Metric 10:	Exposure Duration and Frequency	High	Exposure duration appropriate with regard to species lifespan.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Number of exposure groups and spacing appopriate.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were above solubility limit. Additionally, the test water temperature was raised to 21 +/- 1 C to improve solubility.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Test organisms were well described.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Fish were acclimatized by raising the temperature of the stock aquariums to 18 C two weeks prior to exposure.
	Metric 15:	Number of Organisms and Replicates per Group	Low	n=10 per exposure group. Test group replicates were not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions were acceptable.
	Metric 17:	Outcome Assessment Methodology	High	Intended outcomes were reported.

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Study Citation:	Kuiper, R. V., Cantón, R. F., Leonards, P. E., Jenssen, B. M., Dubbeldam, M., Wester, P. W., Berg, v.d., M., Vos, J. G., Vethaak, A. D. (2007). Long-term exposure of European flounder (Platichthys flesus) to the flame-retardants tetrabromobisphenol A (TBBPA) and hexabromocyclododecane (HBCD). Ecotoxicology and Environmental Safety 67(3):349-360.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Platichthys flesus*; Juvenile
Health Outcome:	Hepatic/Liver
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1412802

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	No confounding variables among groups were reported.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	no differences among groups were reported.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical approach well described.
	Metric 22:	Reporting of Data	High	All exposure related findings were reported.
	Metric 23:	Explanation of Unexpected Outcomes	High	No unexpected outcomes were reported.

Additional Comments: This form represents the histology examinations represented in results section 3.7. "There were no dose-related histopathological changes in internal organs including liver, spleen, kidney, gonad and thyroid gland in fish exposed to TBBPA or HBCD. The numbers of macrophage centers in liver, spleen and kidney varied greatly." This evaluation is for the liver reported under hepatic/liver outcome.

Overall Quality Determination High

Study Citation:	Kuiper, R. V., Cantón, R. F., Leonards, P. E., Jenssen, B. M., Dubbeldam, M., Wester, P. W., Berg, v.d., M., Vos, J. G., Vethaak, A. D. (2007). Long-term exposure of European flounder (Platichthys flesus) to the flame-retardants tetrabromobisphenol A (TBBPA) and hexabromocyclododecane (HBCD). Ecotoxicology and Environmental Safety 67(3):349-360.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Platichthys flesus*; Juvenile
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1412802

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Identified by acronym, nomenclature, and analytically verified.
	Metric 2:	Test Substance Source	High	Obtained through Bromine Science and Environmental Forum (BSEF), and was analytically verified.
	Metric 3:	Test Substance Purity	High	Purity = 99.17%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Negative controls used.
	Metric 5:	Negative Control Response	High	Control group responses were adequate.
	Metric 6:	Randomized Allocation	Low	Randomization of organism allocation to test groups were not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	Environmental system was well described and accounted for p-chem properties.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were consistent across groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Nominal concentrations used with continuous renewal of test chemical.
	Metric 10:	Exposure Duration and Frequency	High	Exposure duration appropriate with regard to species lifespan.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Number of exposure groups and spacing appopriate.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were above solubility limit. Additionally, the test water temperature was raised to 21 +/- 1 C to improve solubility.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Test organisms were well described.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Fish were acclimatized by raising the temperature of the stock aquariums to 18 C two weeks prior to exposure.
	Metric 15:	Number of Organisms and Replicates per Group	Low	n=10 per exposure group. Test group replicates were not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions were acceptable.
	Metric 17:	Outcome Assessment Methodology	High	Intended outcomes were reported.

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Study Citation:	Kuiper, R. V., Cantón, R. F., Leonards, P. E., Jenssen, B. M., Dubbeldam, M., Wester, P. W., Berg, v.d., M., Vos, J. G., Vethaak, A. D. (2007). Long-term exposure of European flounder (Platichthys flesus) to the flame-retardants tetrabromobisphenol A (TBBPA) and hexabromocyclododecane (HBCD). Ecotoxicology and Environmental Safety 67(3):349-360.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Platichthys flesus*; Juvenile
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1412802

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	No confounding variables among groups were reported.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	no differences among groups were reported.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical approach well described.
	Metric 22:	Reporting of Data	High	All exposure related findings were reported.
	Metric 23:	Explanation of Unexpected Outcomes	High	No unexpected outcomes were reported.

Additional Comments: This form represents Growth and morphological outcomes from exposure. These include growth, morphology, and organ weight metrics reported in section 3.2 ”gross Observations”.

Overall Quality Determination High

Study Citation:	Kuiper, R. V., Cantón, R. F., Leonards, P. E., Jenssen, B. M., Dubbeldam, M., Wester, P. W., Berg, v.d., M., Vos, J. G., Vethaak, A. D. (2007). Long-term exposure of European flounder (Platichthys flesus) to the flame-retardants tetrabromobisphenol A (TBBPA) and hexabromocyclododecane (HBCD). Ecotoxicology and Environmental Safety 67(3):349-360.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Platichthys flesus*; Juvenile
Health Outcome:	Renal/Kidney
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1412802

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Identified by acronym, nomenclature, and analytically verified.
	Metric 2:	Test Substance Source	High	Obtained through Bromine Science and Environmental Forum (BSEF), and was analytically verified.
	Metric 3:	Test Substance Purity	High	Purity = 99.17%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Negative controls used
	Metric 5:	Negative Control Response	High	Control group responses were adequate.
	Metric 6:	Randomized Allocation	Low	Randomization of organism allocation to test groups were not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	Environmental system was well described and accounted for p-chem properties.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were consistent across groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Nominal concentrations used with continuous renewal of test chemical.
	Metric 10:	Exposure Duration and Frequency	High	Exposure duration appropriate with regard to species lifespan.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Number of exposure groups and spacing appopriate.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were above solubility limit. Additionally, the test water temperature was raised to 21 +/- 1 C to improve solubility.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Test organisms were well described.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Fish were acclimatized by raising the temperature of the stock aquariums to 18 C two weeks prior to exposure.
	Metric 15:	Number of Organisms and Replicates per Group	Low	n=10 per exposure group. Test group replicates were not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions were acceptable.
	Metric 17:	Outcome Assessment Methodology	High	Intended outcomes were reported.

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Study Citation:	Kuiper, R. V., Cantón, R. F., Leonards, P. E., Jenssen, B. M., Dubbeldam, M., Wester, P. W., Berg, v.d., M., Vos, J. G., Vethaak, A. D. (2007). Long-term exposure of European flounder (Platichthys flesus) to the flame-retardants tetrabromobisphenol A (TBBPA) and hexabromocyclododecane (HBCD). Ecotoxicology and Environmental Safety 67(3):349-360.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Platichthys flesus*; Juvenile
Health Outcome:	Renal/Kidney
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1412802

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	No confounding variables among groups were reported.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	no differences among groups were reported.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical approach well described.
	Metric 22:	Reporting of Data	High	All exposure related findings were reported.
	Metric 23:	Explanation of Unexpected Outcomes	High	No unexpected outcomes were reported.

Additional Comments: This form represents the histology examinations represented in results section 3.7. "There were no dose-related histopathological changes in internal organs including liver, spleen, kidney, gonad and thyroid gland in fish exposed to TBBPA or HBCD. The numbers of macrophage centers in liver, spleen and kidney varied greatly." This evaluation is for the kidney under the renal/kidney outcome.

Overall Quality Determination High

Study Citation:	Kuiper, R. V., Cantón, R. F., Leonards, P. E., Jenssen, B. M., Dubbeldam, M., Wester, P. W., Berg, v.d., M., Vos, J. G., Vethaak, A. D. (2007). Long-term exposure of European flounder (Platichthys flesus) to the flame-retardants tetrabromobisphenol A (TBBPA) and hexabromocyclododecane (HBCD). Ecotoxicology and Environmental Safety 67(3):349-360.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Platichthys flesus*; Juvenile
Health Outcome:	Endocrine
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1412802

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Identified by acronym, nomenclature, and analytically verified.
	Metric 2:	Test Substance Source	High	Obtained through Bromine Science and Environmental Forum (BSEF), and was analytically verified.
	Metric 3:	Test Substance Purity	High	Purity = 99.17%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Negative controls used
	Metric 5:	Negative Control Response	High	Control group responses were adequate.
	Metric 6:	Randomized Allocation	Low	Randomization of organism allocation to test groups were not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	Environmental system was well described and accounted for p-chem properties.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were consistent across groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Nominal concentrations used with continuous renewal of test chemical.
	Metric 10:	Exposure Duration and Frequency	High	Exposure duration appropriate with regard to species lifespan.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Number of exposure groups and spacing appopriate.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were above solubility limit. Additionally, the test water temperature was raised to 21 +/- 1 C to improve solubility.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Test organisms were well described.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Fish were acclimatized by raising the temperature of the stock aquariums to 18 C two weeks prior to exposure.
	Metric 15:	Number of Organisms and Replicates per Group	Low	n=10 per exposure group. Test group replicates were not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions were acceptable.
	Metric 17:	Outcome Assessment Methodology	High	Intended outcomes were reported.

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Study Citation:	Kuiper, R. V., Cantón, R. F., Leonards, P. E., Jenssen, B. M., Dubbeldam, M., Wester, P. W., Berg, v.d., M., Vos, J. G., Vethaak, A. D. (2007). Long-term exposure of European flounder (Platichthys flesus) to the flame-retardants tetrabromobisphenol A (TBBPA) and hexabromocyclododecane (HBCD). Ecotoxicology and Environmental Safety 67(3):349-360.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Platichthys flesus*; Juvenile
Health Outcome:	Endocrine
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1412802

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	No confounding variables among groups were reported.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	no differences among groups were reported.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical approach well described.
	Metric 22:	Reporting of Data	High	All exposure related findings were reported.
	Metric 23:	Explanation of Unexpected Outcomes	High	No unexpected outcomes were reported.

Additional Comments: This form represents the histology examinations represented in results section 3.7. "There were no dose-related histopathological changes in internal organs including liver, spleen, kidney, gonad and thyroid gland in fish exposed to TBBPA or HBCD. The numbers of macrophage centers in liver, spleen and kidney varied greatly." This evaluation is for the thyroid under the endocrine outcome.

Overall Quality Determination High

Study Citation:	Kuiper, R. V., Cantón, R. F., Leonards, P. E., Jenssen, B. M., Dubbeldam, M., Wester, P. W., Berg, v.d., M., Vos, J. G., Vethaak, A. D. (2007). Long-term exposure of European flounder (Platichthys flesus) to the flame-retardants tetrabromobisphenol A (TBBPA) and hexabromocyclododecane (HBCD). Ecotoxicology and Environmental Safety 67(3):349-360.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Platichthys flesus*; Juvenile
Health Outcome:	Immune/Hematological
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1412802

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Identified by acronym, nomenclature, and analytically verified.
	Metric 2:	Test Substance Source	High	Obtained through Bromine Science and Environmental Forum (BSEF), and was analytically verified.
	Metric 3:	Test Substance Purity	High	Purity = 99.17%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Negative controls used
	Metric 5:	Negative Control Response	High	Control group responses were adequate.
	Metric 6:	Randomized Allocation	Low	Randomization of organism allocation to test groups were not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	Environmental system was well described and accounted for p-chem properties.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were consistent across groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Nominal concentrations used with continuous renewal of test chemical.
	Metric 10:	Exposure Duration and Frequency	High	Exposure duration appropriate with regard to species lifespan.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Number of exposure groups and spacing appopriate.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were above solubility limit. Additionally, the test water temperature was raised to 21 +/- 1 C to improve solubility.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Test organisms were well described.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Fish were acclimatized by raising the temperature of the stock aquariums to 18 C two weeks prior to exposure.
	Metric 15:	Number of Organisms and Replicates per Group	Low	n=10 per exposure group. Test group replicates were not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions were acceptable.
	Metric 17:	Outcome Assessment Methodology	High	Intended outcomes were reported.

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Study Citation:	Kuiper, R. V., Cantón, R. F., Leonards, P. E., Jenssen, B. M., Dubbeldam, M., Wester, P. W., Berg, v.d., M., Vos, J. G., Vethaak, A. D. (2007). Long-term exposure of European flounder (Platichthys flesus) to the flame-retardants tetrabromobisphenol A (TBBPA) and hexabromocyclododecane (HBCD). Ecotoxicology and Environmental Safety 67(3):349-360.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Platichthys flesus*; Juvenile
Health Outcome:	Immune/Hematological
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1412802

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	No confounding variables among groups were reported.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	no differences among groups were reported.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical approach well described.
	Metric 22:	Reporting of Data	High	All exposure related findings were reported.
	Metric 23:	Explanation of Unexpected Outcomes	High	No unexpected outcomes were reported.

Additional Comments: This form represents the histology examinations represented in results section 3.7. "There were no dose-related histopathological changes in internal organs including liver, spleen, kidney, gonad and thyroid gland in fish exposed to TBBPA or HBCD. The numbers of macrophage centers in liver, spleen and kidney varied greatly." This evaluation is for the spleen under the immune/hematological outcome.

Overall Quality Determination High

Study Citation:	Kuiper, R. V., Cantón, R. F., Leonards, P. E., Jenssen, B. M., Dubbeldam, M., Wester, P. W., Berg, v.d., M., Vos, J. G., Vethaak, A. D. (2007). Long-term exposure of European flounder (Platichthys flesus) to the flame-retardants tetrabromobisphenol A (TBBPA) and hexabromocyclododecane (HBCD). Ecotoxicology and Environmental Safety 67(3):349-360.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Platichthys flesus*; Juvenile
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1412802

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Identified by acronym, nomenclature, and analytically verified.
	Metric 2:	Test Substance Source	High	Obtained through Bromine Science and Environmental Forum (BSEF), and was analytically verified.
	Metric 3:	Test Substance Purity	High	Purity = 99.17%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Negative controls used
	Metric 5:	Negative Control Response	High	Control group responses were adequate.
	Metric 6:	Randomized Allocation	Low	Randomization of organism allocation to test groups were not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	Environmental system was well described and accounted for p-chem properties.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were consistent across groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Nominal concentrations used with continuous renewal of test chemical.
	Metric 10:	Exposure Duration and Frequency	High	Exposure duration appropriate with regard to species lifespan.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Number of exposure groups and spacing appopriate.
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were above solubility limit. Additionally, the test water temperature was raised to 21 +/- 1 C to improve solubility.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Test organisms were well described.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Fish were acclimatized by raising the temperature of the stock aquariums to 18 C two weeks prior to exposure.
	Metric 15:	Number of Organisms and Replicates per Group	Low	n=10 per exposure group. Test group replicates were not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions were acceptable.
	Metric 17:	Outcome Assessment Methodology	High	Intended outcomes were reported.

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Study Citation:	Kuiper, R. V., Cantón, R. F., Leonards, P. E., Jenssen, B. M., Dubbeldam, M., Wester, P. W., Berg, v.d., M., Vos, J. G., Vethaak, A. D. (2007). Long-term exposure of European flounder (Platichthys flesus) to the flame-retardants tetrabromobisphenol A (TBBPA) and hexabromocyclododecane (HBCD). Ecotoxicology and Environmental Safety 67(3):349-360.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Aquatic (brackish); Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Fish; *Platichthys flesus*; Juvenile
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1412802

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	No confounding variables among groups were reported.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	no differences among groups were reported.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical approach well described.
	Metric 22:	Reporting of Data	High	All exposure related findings were reported.
	Metric 23:	Explanation of Unexpected Outcomes	High	No unexpected outcomes were reported.

Additional Comments: This form represents the histology examinations represented in results section 3.7. "There were no dose-related histopathological changes in internal organs including liver, spleen, kidney, gonad and thyroid gland in fish exposed to TBBPA or HBCD. The numbers of macrophage centers in liver, spleen and kidney varied greatly." This evaluation is for the gonads under the reproduction/teratogenic outcome.

Overall Quality Determination High

Study Citation:	Goodman, L. R., Cripe, G. M., Moody, P. H., Halsell, D. G. (1988). Acute toxicity of malathion, tetrabromobisphenol-a, and tirbutyltin chloride to mysids (mysidopsis bahia) of three ages. Bulletin of Environmental Contamination and Toxicology 41(5):746.
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; *Mysidopsis bahia*; Juvenile
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5757963

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	No CAS or structure/formula presented, compound referred to by name ”tetrabromobisphenol- A [2,2-bis(3,5- dibromo-4-hydroxy phenyl) propane]”
	Metric 2:	Test Substance Source	Low	Great lakes chemical company was listed as the source and reported LOT # 051450. The lab did perform in house testing of compound.
	Metric 3:	Test Substance Purity	Low	Purity from the manufacturer was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	The study used both a solvent control and non solvent control. The solvent control was 90% triethylene glycol 10 % acetone at a final concentration of 96 ul/L.
	Metric 5:	Negative Control Response	High	Control survival was reported in the results section as >94%.
	Metric 6:	Randomized Allocation	Low	Distribution was described as ”equal” but not random.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	Treatment concentrations were administered with a flow through system at a rate of 13L/hour. test media preparation was well described on the top of page 3/8.
	Metric 8:	Consistency of Exposure Administration	High	Exposure groups and the three age groups of juveniles were carried out the same.
	Metric 9:	Measurement of Test Substance Concentration	Medium	The concentrations in seawater were analyzed but only recovery at three concentrations (535, 445, and 84 ug/L) were reported.
	Metric 10:	Exposure Duration and Frequency	High	The 96 hour duration for acute toxicity were appropriate.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	The number and range of concentrations was not described.
	Metric 12:	Testing at or Below Solubility Limit	Low	Authors did not provide the range of concentrations tested so it is not possible to determine if the solubility was exceeded.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	Authors reported the test organisms as lab reared. Age was described as it served as an objective in the acute tests.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Each age group was reared in similar conditions as per the first paragraph of the materials and methods (Page 2/8). ”Juveniles were reared on the same food and at approximately the same salinity, temperature, and photoperiod as used during testing.”
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Authors reported that each age and treatment concentration was represented by 20 mysids.

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Study Citation:	Goodman, L. R., Cripe, G. M., Moody, P. H., Halsell, D. G. (1988). Acute toxicity of malathion, tetrabromobisphenol-a, and tirbutyltin chloride to mysids (mysidopsis bahia) of three ages. Bulletin of Environmental Contamination and Toxicology 41(5):746.
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; *Mysidopsis bahia*; Juvenile
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5757963

Domain	Metric		Rating	Comments
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Test conditions are reported on page 3/8 in table 1.
	Metric 17:	Outcome Assessment Methodology	Medium	The authors did not provide specific criteria for mortality (response, appearance etc.).
	Metric 18:	Consistency of Outcome Assessment	High	Outcome assessment appeared to be consistent among treatment and control groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	No reported differences in environmental conditions were reported.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	No information was provided to suggest differences in animal attrition or health outcomes unrelated to exposure were present to influence outcome assessment.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	LC50 estimation methods included moving average (Harris 1959) and binomial tests (Siegel 1965) with the probit method used for graphical determination of LC50 and 95% CIs.
	Metric 22:	Reporting of Data	High	Data on 1 day old LC50 and 95% CI were presented.
	Metric 23:	Explanation of Unexpected Outcomes	High	No unexpected outcomes were reported.

Additional Comments: This form is for 1 day old acute tox test.Ninety-six hour LC50 values, based on mean measured exposure concentrations was reported by the study authors. The range of acute test concentrations used, however, was not reported.

Overall Quality Determination Medium

Study Citation:	Goodman, L. R., Cripe, G. M., Moody, P. H., Halsell, D. G. (1988). Acute toxicity of malathion, tetrabromobisphenol-a, and tirbutyltin chloride to mysids (mysidopsis bahia) of three ages. Bulletin of Environmental Contamination and Toxicology 41(5):746.
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; *Mysidopsis bahia*; Juvenile
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5757963

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	No CAS or structure/formula presented, compound referred to by name ”tetrabromobisphenol- A [2,2-bis(3,5- dibromo-4-hydroxy phenyl) propane]”
	Metric 2:	Test Substance Source	Low	Great lakes chemical company was listed as the source and reported LOT # 051450. The lab did perform in house testing of compound.
	Metric 3:	Test Substance Purity	Low	Purity from the manufacturer was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	The study used both a solvent control and non solvent control. The solvent control was 90% triethylene glycol 10 % acetone at a final concentration of 96 ul/L.
	Metric 5:	Negative Control Response	High	Control survival was reported in the results section as >94%.
	Metric 6:	Randomized Allocation	Low	Distribution was described as ”equal” but not random.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	Treatment concentrations were administered with a flow through system at a rate of 13L/hour. test media preparation was well described on the top of page 3/8.
	Metric 8:	Consistency of Exposure Administration	High	Exposure groups and the three age groups of juveniles were carried out the same.
	Metric 9:	Measurement of Test Substance Concentration	Medium	The concentrations in seawater were analyzed but only recovery at three concentrations (535, 445, and 84 ug/L) were reported.
	Metric 10:	Exposure Duration and Frequency	High	The 96 hour duration for acute toxicity were appropriate.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	The number and range of concentrations was not described.
	Metric 12:	Testing at or Below Solubility Limit	Low	Authors did not provide the range of concentrations tested so it is not possible to determine if the solubility was exceeded.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	Authors reported the test organisms as lab reared. Age was described as it served as an objective in the acute tests.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Each age group was reared in similar conditions as per the first paragraph of the materials and methods (Page 2/8). ”Juveniles were reared on the same food and at approximately the same salinity, temperature, and photoperiod as used during testing.”
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Authors reported that each age and treatment concentration was represented by 20 mysids.
Domain 5: Outcome Assessment				

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Study Citation:	Goodman, L. R., Cripe, G. M., Moody, P. H., Halsell, D. G. (1988). Acute toxicity of malathion, tetrabromobisphenol-a, and tirbutyltin chloride to mysids (mysidopsis bahia) of three ages. Bulletin of Environmental Contamination and Toxicology 41(5):746.
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; *Mysidopsis bahia*; Juvenile
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5757963

Domain	Metric		Rating	Comments
	Metric 16:	Adequacy of Test Conditions	High	Test conditions are reported on page 3/8 in table 1.
	Metric 17:	Outcome Assessment Methodology	Medium	The authors did not provide specific criteria for mortality (response, appearance etc.).
	Metric 18:	Consistency of Outcome Assessment	High	Outcome assessment appeared to be consistent among treatment and control groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	No reported differences in environmental conditions were reported.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	No information was provided to suggest differences in animal attrition or health outcomes unrelated to exposure were present to influence outcome assessment.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	LC50 estimation methods included moving average (Harris 1959) and binomial tests (Siegel 1965) with the probit method used for graphical determination of LC50 and 95% CIs.
	Metric 22:	Reporting of Data	Low	No 95% CIs are presented and the LC value in table 2 conflicts with footnote C in Table 2.
	Metric 23:	Explanation of Unexpected Outcomes	High	No unexpected outcomes were reported.

Additional Comments: This form is for 9-10 day old mysid acute tox data. The paper only presents the LC50 with no 95% CIs. The LC50 presented in table 2 represents the highes TBBPA concentration tested. Ninety-six hour LC50 values, based on mean measured exposure concentrations was reported by the study authors. The range of acute test concentrations used, however, was not reported.

Overall Quality Determination Medium

Study Citation:	Goodman, L. R., Cripe, G. M., Moody, P. H., Halsell, D. G. (1988). Acute toxicity of malathion, tetrabromobisphenol-a, and tirbutyltin chloride to mysids (mysidopsis bahia) of three ages. Bulletin of Environmental Contamination and Toxicology 41(5):746.
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; *Mysidopsis bahia*; Juvenile
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5757963

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	No CAS or structure/formula presented, compound referred to by name ”tetrabromobisphenol- A [2,2-bis(3,5- dibromo-4-hydroxy phenyl) propane]”
	Metric 2:	Test Substance Source	Low	Great lakes chemical company was listed as the source and reported LOT # 051450. The lab did perform in house testing of compound.
	Metric 3:	Test Substance Purity	Low	Purity from the manufacturer was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	The study used both a solvent control and non solvent control. The solvent control was 90% triethylene glycol 10 % acetone at a final concentration of 96 ul/L.
	Metric 5:	Negative Control Response	High	Control survival was reported in the results section as >94%.
	Metric 6:	Randomized Allocation	Low	Distribution was described as ”equal” but not random.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	Treatment concentrations were administered with a flow through system at a rate of 13L/hour. test media preparation was well described on the top of page 3/8.
	Metric 8:	Consistency of Exposure Administration	High	Exposure groups and the three age groups of juveniles were carried out the same.
	Metric 9:	Measurement of Test Substance Concentration	Medium	The concentrations in seawater were analyzed but only recovery at three concentrations (535, 445, and 84 ug/L) were reported.
	Metric 10:	Exposure Duration and Frequency	High	The 96 hour duration for acute toxicity were appropriate.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	The number and range of concentrations was not described.
	Metric 12:	Testing at or Below Solubility Limit	Low	Authors did not provide the range of concentrations tested so it is not possible to determine if the solubility was exceeded.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	Authors reported the test organisms as lab reared. Age was described as it served as an objective in the acute tests.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Each age group was reared in similar conditions as per the first paragraph of the materials and methods (Page 2/8). ”Juveniles were reared on the same food and at approximately the same salinity, temperature, and photoperiod as used during testing.”
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Authors reported that each age and treatment concentration was represented by 20 mysids.
Domain 5: Outcome Assessment				

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Study Citation:	Goodman, L. R., Cripe, G. M., Moody, P. H., Halsell, D. G. (1988). Acute toxicity of malathion, tetrabromobisphenol-a, and tirbutyltin chloride to mysids (mysidopsis bahia) of three ages. Bulletin of Environmental Contamination and Toxicology 41(5):746.
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; *Mysidopsis bahia*; Juvenile
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5757963

Domain	Metric		Rating	Comments
	Metric 16:	Adequacy of Test Conditions	High	Test conditions are reported on page 3/8 in table 1.
	Metric 17:	Outcome Assessment Methodology	Medium	The authors did not provide specific criteria for mortality (response, appearance etc.).
	Metric 18:	Consistency of Outcome Assessment	High	Outcome assessment appeared to be consistent among treatment and control groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	No reported differences in environmental conditions were reported.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	No information was provided to suggest differences in animal attrition or health outcomes unrelated to exposure were present to influence outcome assessment.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	LC50 estimation methods included moving average (Harris 1959) and binomial tests (Siegel 1965) with the probit method used for graphical determination of LC50 and 95% CIs.
	Metric 22:	Reporting of Data	Low	No 95% CIs are presented and the LC value in table 2 conflicts with footnote C in the Table 2.
	Metric 23:	Explanation of Unexpected Outcomes	High	No unexpected outcomes were reported.

Additional Comments: This form is for 4-5 day old mysid acute tox data. The paper only presents the LC50 with no 95% CIs. Ninety-six hour LC50 values, based on mean measured exposure concentrations was reported by the study authors. The range of acute test concentrations used, however, was not reported.

Overall Quality Determination Medium

Study Citation:	Halldin, K., Berg, C., Bergman, A., Brandt, I., Brunström, B. (2001). Distribution of bisphenol A and tetrabromobisphenol A in quail eggs, embryos and laying birds and studies on reproduction variables in adults following in ovo exposure. Archives of Toxicology 75(10):597-603.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; Food/Diet, N/A (e.g., injection); Injection, Dietary
Taxa, Species, Age:	Vertebrate; Avian; *Coturnix japonica*; Adult
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1416717

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%
Domain 2: Test Design				
	Metric 4:	Negative Controls	N/A	This ADME portion of the study did not have a traditional treatment control.
	Metric 5:	Negative Control Response	N/A	The biological response of the negative control group was reported and reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The test media preparation methods were adequately reported
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	there is only one exposure concentration with an appropriate solvent concentration
	Metric 12:	Testing at or Below Solubility Limit	High	exposure was either by injection or orally administered.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	There are minor reservations about the source of test organisms,
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms,
	Metric 15:	Number of Organisms and Replicates per Group	Medium	each hen is used as a replicate, typical for these studies
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:	Halldin, K., Berg, C., Bergman, A., Brandt, I., Brunström, B. (2001). Distribution of bisphenol A and tetrabromobisphenol A in quail eggs, embryos and laying birds and studies on reproduction variables in adults following in ovo exposure. Archives of Toxicology 75(10):597-603.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; Food/Diet, N/A (e.g., injection); Injection, Dietary
Taxa, Species, Age:	Vertebrate; Avian; *Coturnix japonica*; Adult
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1416717

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	N/A	Statistical analysis was not conducted for uptake or distribution (figure 2, page 5).
	Metric 22:	Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment group, but results were described in the text.
	Metric 23:	Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained

Additional Comments: This form represents fed and injected hens for distribution of TBBPA data (Figure 2, page 5).

Overall Quality Determination Medium

Study Citation:	Halldin, K., Berg, C., Bergman, A., Brandt, I., Brunström, B. (2001). Distribution of bisphenol A and tetrabromobisphenol A in quail eggs, embryos and laying birds and studies on reproduction variables in adults following in ovo exposure. Archives of Toxicology 75(10):597-603.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary
Taxa, Species, Age:	Vertebrate; Avian; *Coturnix japonica*; Adult
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1416717

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%
Domain 2: Test Design				
	Metric 4:	Negative Controls	N/A	This ADME portion of the study did not have a traditional treatment control.
	Metric 5:	Negative Control Response	N/A	The biological response of the negative control group was reported and reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The test media preparation methods were adequately reported
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	there is only one exposure concentration with an appropriate solvent concentration
	Metric 12:	Testing at or Below Solubility Limit	High	exposure was either by injection or orally administered.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	There are minor reservations about the source of test organisms,
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms,
	Metric 15:	Number of Organisms and Replicates per Group	Medium	each hen is used as a replicate, typical for these studies
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:	Halldin, K., Berg, C., Bergman, A., Brandt, I., Brunström, B. (2001). Distribution of bisphenol A and tetrabromobisphenol A in quail eggs, embryos and laying birds and studies on reproduction variables in adults following in ovo exposure. Archives of Toxicology 75(10):597-603.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Terrestrial; Food/Diet; Dietary
Taxa, Species, Age:	Vertebrate; Avian; *Coturnix japonica*; Adult
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1416717

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	N/A	Statistical analysis was not conducted for uptake or distribution (figure 2, page 5).
	Metric 22:	Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment group, but results were described in the text.
	Metric 23:	Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained

Additional Comments: This form represents hens exposed orally to TBBPA for 9 days.

Overall Quality Determination Medium

Study Citation:	Halldin, K., Berg, C., Bergman, A., Brandt, I., Brunström, B. (2001). Distribution of bisphenol A and tetrabromobisphenol A in quail eggs, embryos and laying birds and studies on reproduction variables in adults following in ovo exposure. Archives of Toxicology 75(10):597-603.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection
Taxa, Species, Age:	Vertebrate; Avian; *Coturnix japonica*; Embryo
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1416717

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%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The test media preparation methods were adequately reported
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	there is only one exposure concentration with an appropriate solvent concentration
	Metric 12:	Testing at or Below Solubility Limit	N/A	exposure was via injection
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	There are minor reservations about the source of test organisms,
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms,
	Metric 15:	Number of Organisms and Replicates per Group	Medium	each egg is used as a replicate, typical for these studies
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:	Halldin, K., Berg, C., Bergman, A., Brandt, I., Brunström, B. (2001). Distribution of bisphenol A and tetrabromobisphenol A in quail eggs, embryos and laying birds and studies on reproduction variables in adults following in ovo exposure. Archives of Toxicology 75(10):597-603.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection
Taxa, Species, Age:	Vertebrate; Avian; *Coturnix japonica*; Embryo
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1416717

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	N/A	Statistical analysis was not conducted for uptake or distribution
	Metric 22:	Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group, but results were described in the text
	Metric 23:	Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained

Additional Comments: None

Overall Quality Determination Medium

Study Citation:	Berg, C., Halldin, K., Brunström, B. (2001). Effects of bisphenol A and tetrabromobisphenol A on sex organ development in quail and chicken embryos. Environmental Toxicology and Chemistry 20(12):2836-2840.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection
Taxa, Species, Age:	Vertebrate; Avian; *Coturnix japonica*; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1416653

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%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent solvent control group
	Metric 5:	Negative Control Response	Medium	The biological response of the negative control group was reasonable but a little higher mortality than expected
	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 test media preparation methods were adequately reported.
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable
	Metric 12:	Testing at or Below Solubility Limit	N/A	The dose was administered via an injection to the egg.
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	No replicates were used
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest

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Study Citation:	Berg, C., Halldin, K., Brunström, B. (2001). Effects of bisphenol A and tetrabromobisphenol A on sex organ development in quail and chicken embryos. Environmental Toxicology and Chemistry 20(12):2836-2840.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection
Taxa, Species, Age:	Vertebrate; Avian; *Coturnix japonica*; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1416653

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

Additional Comments: None

Overall Quality Determination Medium

Study Citation:	Berg, C., Halldin, K., Brunström, B. (2001). Effects of bisphenol A and tetrabromobisphenol A on sex organ development in quail and chicken embryos. Environmental Toxicology and Chemistry 20(12):2836-2840.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection
Taxa, Species, Age:	Vertebrate; Avian; *Coturnix japonica*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1416653

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%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent solvennt control group
	Metric 5:	Negative Control Response	Medium	The biological response of the negative control group was reasonable but a little higher effect than expected
	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 test media preparation methods were adequately reported
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable
	Metric 12:	Testing at or Below Solubility Limit	N/A	The dose was administered via an injection to the egg.
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	No replicates were used
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups

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Study Citation:	Berg, C., Halldin, K., Brunström, B. (2001). Effects of bisphenol A and tetrabromobisphenol A on sex organ development in quail and chicken embryos. Environmental Toxicology and Chemistry 20(12):2836-2840.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection
Taxa, Species, Age:	Vertebrate; Avian; *Coturnix japonica*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1416653

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

Additional Comments: None

Overall Quality Determination Medium

Study Citation:	Halldin, K., Berg, C., Bergman, A., Brandt, I., Brunström, B. (2001). Distribution of bisphenol A and tetrabromobisphenol A in quail eggs, embryos and laying birds and studies on reproduction variables in adults following in ovo exposure. Archives of Toxicology 75(10):597-603.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection
Taxa, Species, Age:	Vertebrate; Avian; *Coturnix japonica*; Embryo
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1416717

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%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The test media preparation methods were adequately reported
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	there is only one exposure concentration with an appropriate solvent concentration
	Metric 12:	Testing at or Below Solubility Limit	N/A	exposure was via injection
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	There are minor reservations about the source of test organisms,
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms,
	Metric 15:	Number of Organisms and Replicates per Group	Medium	each egg is used as a replicate, typical for these studies
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:	Halldin, K., Berg, C., Bergman, A., Brandt, I., Brunström, B. (2001). Distribution of bisphenol A and tetrabromobisphenol A in quail eggs, embryos and laying birds and studies on reproduction variables in adults following in ovo exposure. Archives of Toxicology 75(10):597-603.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection
Taxa, Species, Age:	Vertebrate; Avian; *Coturnix japonica*; Embryo
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1416717

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	N/A	Statistical analysis was not conducted for uptake or distribution
	Metric 22:	Reporting of Data	Low	Data for exposure-related findings were not shown for each treatment and control group, but results were described in the text
	Metric 23:	Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained

Additional Comments: None

Overall Quality Determination Medium

Study Citation:	Halldin, K., Berg, C., Bergman, A., Brandt, I., Brunström, B. (2001). Distribution of bisphenol A and tetrabromobisphenol A in quail eggs, embryos and laying birds and studies on reproduction variables in adults following in ovo exposure. Archives of Toxicology 75(10):597-603.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection
Taxa, Species, Age:	Vertebrate; Avian; *Coturnix japonica*; Embryo
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1416717

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%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable
	Metric 6:	Randomized Allocation	Low	Allocation to treatment groups was not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The test media preparation methods were adequately reported
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	there is only one exposure concentration with an appropriate solvent concentration
	Metric 12:	Testing at or Below Solubility Limit	N/A	exposure was via injection
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	There are minor reservations about the source of test organisms,
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms,
	Metric 15:	Number of Organisms and Replicates per Group	Medium	each egg is used as a replicate, typical for these studies
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:	Halldin, K., Berg, C., Bergman, A., Brandt, I., Brunström, B. (2001). Distribution of bisphenol A and tetrabromobisphenol A in quail eggs, embryos and laying birds and studies on reproduction variables in adults following in ovo exposure. Archives of Toxicology 75(10):597-603.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection
Taxa, Species, Age:	Vertebrate; Avian; *Coturnix japonica*; Embryo
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1416717

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	Low	Statistics were described in the methods section, however, many statistical tests were not well described in the results section.
	Metric 22:	Reporting of Data	Low	Data for exposure-related findings were shown for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained

Additional Comments: Reproductive outputs from this work include: female egg output, and oviduct morphology. Data are reported for males in Table 2 and Table 3 for females.

Overall Quality Determination Medium

Study Citation:	Halldin, K., Berg, C., Bergman, A., Brandt, I., Brunström, B. (2001). Distribution of bisphenol A and tetrabromobisphenol A in quail eggs, embryos and laying birds and studies on reproduction variables in adults following in ovo exposure. Archives of Toxicology 75(10):597-603.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection
Taxa, Species, Age:	Vertebrate; Avian; *Coturnix japonica*; Embryo
Health Outcome:	Mechanistic-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1416717

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%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The test media preparation methods were adequately reported
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	there is only one exposure concentration with an appropriate solvent concentration
	Metric 12:	Testing at or Below Solubility Limit	N/A	exposure was via injection
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	There are minor reservations about the source of test organisms,
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms,
	Metric 15:	Number of Organisms and Replicates per Group	Medium	each egg is used as a replicate, typical for these studies
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:	Halldin, K., Berg, C., Bergman, A., Brandt, I., Brunström, B. (2001). Distribution of bisphenol A and tetrabromobisphenol A in quail eggs, embryos and laying birds and studies on reproduction variables in adults following in ovo exposure. Archives of Toxicology 75(10):597-603.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection
Taxa, Species, Age:	Vertebrate; Avian; *Coturnix japonica*; Embryo
Health Outcome:	Mechanistic-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1416717

Domain	Metric		Rating	Comments
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistics were described in the methods, however, statistical tests were not well described in the results.
	Metric 22:	Reporting of Data	Medium	Data for exposure-related findings were shown for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	unexpected outcomes were satisfactorily explained

Additional Comments: This form represents plasma testosterone values from TBBPA treatment (15ug/g egg) compared to control (Table 2, page 5).

Overall Quality Determination Medium

Study Citation:	Fini, J. B., Riu, A., Debrauwer, L., Hillenweck, A., Mével, Le, S., Chevolleau, S., Boulahtouf, A., Palmier, K., Balaguer, P., Cravedi, J. P., Demeneix, B. A., Zalko, D. (2012). Parallel biotransformation of tetrabromobisphenol A in Xenopus laevis and mammals: Xenopus as a model for endocrine perturbation studies. Toxicological Sciences 125(2):359-367.
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:	Vertebrate; Fish; *Danio rerio*; In vitro Thyroid Hormone Receptor Construct; Adult
Health Outcome:	Mechanistic-Receptor binding/ regulation of receptor activity
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1293799

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was only identified by name (tetrabromobisphenol A, TBBPA), but no other descriptors such as CASRN or structure were reported.
	Metric 2:	Test Substance Source	High	Reporter in vitro cell assay: TBBPA or TBBPA-monosulfate. TBBPA (>97% purity) was purchased from Sigma-Aldrich. TBBPA-monosulfate was synthesized by the laboratory.
	Metric 3:	Test Substance Purity	High	Reporter in vitro cell assay: TBBPA (>97% purity) was purchased from Sigma-Aldrich. TBBPA-monosulfate was synthesized by the laboratory.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Reporter in vitro cell assay: Details are missing in the methods, but authors indicated a control group in Figure 4.
	Metric 5:	Negative Control Response	High	Reporter in vitro cell assay: The biological response (D. rerio TRalpha binding activity in the absence/presence of tri-iodothyronine) of the control group was reported and appeared appropriate (Figure 4).
	Metric 6:	Randomized Allocation	Low	Randomized allocation not stated.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study lacked details on how TBBPA and TBBPA-monosulfate solutions were prepared to add to the cell culture. the exposure solutions.
	Metric 8:	Consistency of Exposure Administration	High	Evidence suggests that exposure administration was administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	TBBPA or TBBPA-monosulfate were not measured in the vitro assays.
	Metric 10:	Exposure Duration and Frequency	Uninformative	D. rerio TRalpha using a reporter in vitro cell assay: The exposure duration or incubation period was not reported, it is unclear if bindings response readings were done immediately or after an incubation period which is typical of these reporter cell assays.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	D. rerio TRalpha using a reporter in vitro cell assay: cells were exposed to either TBBPA or TBBPA-monosulfate at 1, 3, and 10 uMconcentrations. But it unclear why these concentrations were selected.

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Study Citation:	Fini, J. B., Riu, A., Debrauwer, L., Hillenweck, A., Mével, Le, S., Chevolleau, S., Boulahtouf, A., Palmier, K., Balaguer, P., Cravedi, J. P., Demeneix, B. A., Zalko, D. (2012). Parallel biotransformation of tetrabromobisphenol A in Xenopus laevis and mammals: Xenopus as a model for endocrine perturbation studies. Toxicological Sciences 125(2):359-367.
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:	Vertebrate; Fish; *Danio rerio*; In vitro Thyroid Hormone Receptor Construct; Adult
Health Outcome:	Mechanistic-Receptor binding/ regulation of receptor activity
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1293799

Domain	Metric		Rating	Comments
	Metric 12:	Testing at or Below Solubility Limit	Uninformative	D. rerio TRalpha using a reporter in vitro cell assay: cells were exposed to either TBBPA or TBBPA-monosulfate at 1, 3, and 10 uM concentrations. Therefore, in vitro exposure concentrations were above the solubility limit (TBBPA's solubility limit is 4.15 mg/L at 25 C pH 7.56). It is unclear what solvents authors used in the in vitro assay.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The in vitro construct was described. D. rerio TRalpha construct transfected into in HeLa cells to perform the binding assay.
	Metric 14:	Acclimatization and Pretreatment Conditions	Medium	Authors did not mention acclimatizing cells used in the in vitro assay, but these details are less common of in vitro assays.
	Metric 15:	Number of Organisms and Replicates per Group	Low	D. rerio TRalpha using a reporter in vitro cell assay: Authors reported that luciferase assays were performed with the Promega dual-reporterkit according to the manufacturer's instructions. Authors reported that transfection assays were performed twice in triplicate. However, they did not report specific number of cells tested.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	These were in vitro transfection assays. The authors did not report how they maintained the HeLA cells used in the transfection assays.
	Metric 17:	Outcome Assessment Methodology	High	D. rerio TRalpha using a reporter in vitro cell assay: The outcome methodology (preparation of construct transfected into HeLA ceels for binding in vitro assay) was reported and appropriate for the intended outcome (TRalpha activity in the presence or absence of TBBPA and TBBPA-monosulfate).
	Metric 18:	Consistency of Outcome Assessment	High	The outcome assessment was consistent across 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.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were described (ANOVA Kruskal-Wallis test followed by Dunn's test).
	Metric 22:	Reporting of Data	High	D. rerio TRalpha using a reporter in vitro cell assay: Data were presented for each control and treatment groups (Figure 4).

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Study Citation:	Fini, J. B., Riu, A., Debrauwer, L., Hillenweck, A., Mével, Le, S., Chevolleau, S., Boulahtouf, A., Palmier, K., Balaguer, P., Cravedi, J. P., Demeneix, B. A., Zalko, D. (2012). Parallel biotransformation of tetrabromobisphenol A in Xenopus laevis and mammals: Xenopus as a model for endocrine perturbation studies. Toxicological Sciences 125(2):359-367.
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:	Vertebrate; Fish; *Danio rerio*; In vitro Thyroid Hormone Receptor Construct; Adult
Health Outcome:	Mechanistic-Receptor binding/ regulation of receptor activity
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1293799

Domain	Metric	Rating	Comments
	Metric 23: Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This study examined thyroid disruption effects of TBBPA in Xenopus laevis. To do this the authors (1) assessed the kinetics of TBBPA (uptake and excretion) in X. laevis tadpoles, (2) investigated if antithyroid effects are due to the parent TBBPA or a metabolite using two readouts: tadpole morphology and an in vivo TH reporter assay, and (3) examined the ability of sulfate metabolites andTBBPA to exert direct actions on the TRalpha using a reporter in vitro cell assay. This form was used to evaluate the the ability of sulfate metabolites and TBBPA to exert direct actions on the D. rerio TRalpha using a reporter in vitro cell assay.

Overall Quality Determination — Uninformative

Study Citation:	Berg, C., Halldin, K., Brunström, B. (2001). Effects of bisphenol A and tetrabromobisphenol A on sex organ development in quail and chicken embryos. Environmental Toxicology and Chemistry 20(12):2836-2840.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection
Taxa, Species, Age:	Vertebrate; Avian; *Gallus domesticus*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1416653

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%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent solvennt control group
	Metric 5:	Negative Control Response	Medium	The biological response of the negative control group was reasonable but a little higher effect than expected
	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 test media preparation methods were adequately reported
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable
	Metric 12:	Testing at or Below Solubility Limit	Medium	The dose was administered via an injection to the egg.
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	No replicates were used
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest

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Study Citation:	Berg, C., Halldin, K., Brunström, B. (2001). Effects of bisphenol A and tetrabromobisphenol A on sex organ development in quail and chicken embryos. Environmental Toxicology and Chemistry 20(12):2836-2840.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection
Taxa, Species, Age:	Vertebrate; Avian; *Gallus domesticus*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1416653

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

Additional Comments: None

Overall Quality Determination Medium

Study Citation:	Berg, C., Halldin, K., Brunström, B. (2001). Effects of bisphenol A and tetrabromobisphenol A on sex organ development in quail and chicken embryos. Environmental Toxicology and Chemistry 20(12):2836-2840.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection
Taxa, Species, Age:	Vertebrate; Avian; *Gallus domesticus*; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1416653

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%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent solvennt control group
	Metric 5:	Negative Control Response	Medium	The biological response of the negative control group was reasonable but a little higher mortality than expected
	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 test media preparation methods were adequately reported
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable
	Metric 12:	Testing at or Below Solubility Limit	N/A	The dose was administered via an injection to the egg.
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	No replicates were used
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups

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Study Citation:	Berg, C., Halldin, K., Brunström, B. (2001). Effects of bisphenol A and tetrabromobisphenol A on sex organ development in quail and chicken embryos. Environmental Toxicology and Chemistry 20(12):2836-2840.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Injection
Taxa, Species, Age:	Vertebrate; Avian; *Gallus domesticus*; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1416653

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

Additional Comments: None

Overall Quality Determination Medium

Study Citation:	Behl, M., Hsieh, J. H., Shafer, T. J., Mundy, W. R., Rice, J. R., Boyd, W. A., Freedman, J. H., Hunter, E. S., Jarema, K. A., Padilla, S., Tice, R. R. (2015). Use of alternative assays to identify and prioritize organophosphorus flame retardants for potential developmental and neurotoxicity. Neurotoxicology and Teratology 52(Pt B):181-193.
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:	Vertebrate; Mammalian; *Mus musculus*; embryonic stem cell; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Cell signaling/function-Cytotoxicity-Developmental and juvenile toxicology
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3479540

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent solvent control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided few details on exposure media preparation
	Metric 8:	Consistency of Exposure Administration	Low	The study provided few details on exposure administration
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were justified for a dose response by study authors
	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	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
	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	2500 cells per well, did not see evidence of replicates being used
Domain 5: Outcome Assessment				

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Study Citation:	Behl, M., Hsieh, J. H., Shafer, T. J., Mundy, W. R., Rice, J. R., Boyd, W. A., Freedman, J. H., Hunter, E. S., Jarema, K. A., Padilla, S., Tice, R. R. (2015). Use of alternative assays to identify and prioritize organophosphorus flame retardants for potential developmental and neurotoxicity. Neurotoxicology and Teratology 52(Pt B):181-193.
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:	Vertebrate; Mammalian; *Mus musculus*; embryonic stem cell; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Cell signaling/function-Cytotoxicity-Developmental and juvenile toxicology
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3479540

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

Additional Comments: mouse cell differentiation and cytotoxicity

Overall Quality Determination Medium

Study Citation:	Zhang, H., Liu, W., Chen, B., He, J., Chen, F., Shan, X., Du, Q., Li, N., Jia, X., Tang, J. (2018). Differences in reproductive toxicity of TBBPA and TCBPA exposure in male Rana nigromaculata. Environmental Pollution 243(Pt A):394-403.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Rana nigromaculata*; Adult
Health Outcome:	Mechanistic-Cell signaling/function-Endocrine toxicity
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547039

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Nomenclature and CASRN provided.
	Metric 2:	Test Substance Source	Low	TBBPA was purchased from Aladdin Industrial Corporation (FengXian, Shanghai, China) but was not analytically verified.
	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 groups was acceptable.
	Metric 6:	Randomized Allocation	Medium	Frogs were randomly assigned to study groups (Section 2.2).
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The experimental system and test media preparation methods were reported but not in sufficient detail (type, closed or open, etc). Concentration of test substance was measured.
	Metric 8:	Consistency of Exposure Administration	Medium	Details of exposure administration were reported but not in sufficient detail. Exposure volume used for treatment groups was not reported.
	Metric 9:	Measurement of Test Substance Concentration	Medium	Exposure concentrations were measured and are similar to nominal concentrations (results given in supplemental document) but timing of measurement (before renewal or at start or end of the experiment) was not given.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were adequate to address the purpose of the study. 4 exposure concentrations were used- 0.001, 0.01, 0.1, and 1 mg/L TBBPA.
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate (i.e., no effects on biological responses were observed in the solvent control and no interactions were expected between the solvent and test substance).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	”Three year old male R. nigromaculata frogs were purchased fromthe outskirts of Huzhou (Zhejiang, China)”. Minor reservation regarding the source of the test organisms.

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Study Citation:	Zhang, H., Liu, W., Chen, B., He, J., Chen, F., Shan, X., Du, Q., Li, N., Jia, X., Tang, J. (2018). Differences in reproductive toxicity of TBBPA and TCBPA exposure in male Rana nigromaculata. Environmental Pollution 243(Pt A):394-403.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Rana nigromaculata*; Adult
Health Outcome:	Mechanistic-Cell signaling/function-Endocrine toxicity
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547039

Domain	Metric		Rating	Comments
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions for 14 days and all pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of replicates was not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Organism housing and environmental conditions were conducive to maintenance of health. Standard experimental conditions(temperature range of 18oC-20oC; pH range of 6.0-7.0; dissolvedoxygen range of 6-8 mg/L) were maintained for treatment groups and control organisms. Frogs were fed Eisenia fetida twice a day.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodologies for hormone concentrations and AR gene expression were given in detail and addressed the intended outcomes of interest.
	Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups.
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 that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately. The type of statistical test performed was not indicated, only software details were given.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings (testosterone, estradiol, LH and FSH concentrations and fold change in AR gene expression) were presented for each treatment and control group in Figures 4, 5 & 6.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: This evaluation form is relevant for mechanistic endpoints- testosterone, estradiol, LH and FSH concentrations and fold change in AR gene expression and results were quantitative.

Overall Quality Determination High

Study Citation:	Zhang, H., Liu, W., Chen, B., He, J., Chen, F., Shan, X., Du, Q., Li, N., Jia, X., Tang, J. (2018). Differences in reproductive toxicity of TBBPA and TCBPA exposure in male Rana nigromaculata. Environmental Pollution 243(Pt A):394-403.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Rana nigromaculata*; Adult
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547039

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Nomenclature and CASRN provided.
	Metric 2:	Test Substance Source	Low	TBBPA was purchased from Aladdin Industrial Corporation (FengXian, Shanghai, China) but was not analytically verified.
	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 groups was acceptable.
	Metric 6:	Randomized Allocation	Medium	Frogs were randomly assigned to study groups (Section 2.2).
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The experimental system and test media preparation methods were reported but not in sufficient detail (type, closed or open, etc). Concentration of test substance was measured.
	Metric 8:	Consistency of Exposure Administration	Medium	Details of exposure administration were reported but not in sufficient detail. Exposure volume used for treatment groups was not reported.
	Metric 9:	Measurement of Test Substance Concentration	Medium	Exposure concentrations were measured and are similar to nominal concentrations (results given in supplemental document) but timing of measurement (before renewal or at start or end of the experiment) was not given.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were adequate to address the purpose of the study. 4 exposure concentrations were used- 0.001, 0.01, 0.1, and 1 mg/L TBBPA.
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate (i.e., no effects on biological responses were observed in the solvent control and no interactions were expected between the solvent and test substance).
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	”Three year old male R. nigromaculata frogs were purchased fromthe outskirts of Huzhou (Zhejiang, China)”. Minor reservation regarding the source of the test organisms.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions for 14 days and all pretreatment conditions were the same for control and exposed organisms.

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Study Citation:	Zhang, H., Liu, W., Chen, B., He, J., Chen, F., Shan, X., Du, Q., Li, N., Jia, X., Tang, J. (2018). Differences in reproductive toxicity of TBBPA and TCBPA exposure in male Rana nigromaculata. Environmental Pollution 243(Pt A):394-403.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Water; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vertebrate; Amphibian; *Rana nigromaculata*; Adult
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5547039

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of replicates was not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Organism housing and environmental conditions were conducive to maintenance of health. Standard experimental conditions(temperature range of 18oC-20oC; pH range of 6.0-7.0; dissolvedoxygen range of 6-8 mg/L) were maintained for treatment groups and control organisms. Frogs were fed Eisenia fetida twice a day.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodologies for sperm count, mobility and deformity were given in detail and addressed the intended outcomes of interest.
	Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups.
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 that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately. The type of statistical test performed was not indicated, only software details were given.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings (sperm count, mobility and deformity) were presented for each treatment and control group in Fig 1.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: This evaluation form is relevant for reproductive endpoints- sperm count, mobility and deformity and results were quantitative.

Overall Quality Determination High

Study Citation:	Behl, M., Hsieh, J. H., Shafer, T. J., Mundy, W. R., Rice, J. R., Boyd, W. A., Freedman, J. H., Hunter, E. S., Jarema, K. A., Padilla, S., Tice, R. R. (2015). Use of alternative assays to identify and prioritize organophosphorus flame retardants for potential developmental and neurotoxicity. Neurotoxicology and Teratology 52(Pt B):181-193.
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:	Vertebrate; Mammalian; *Rattus norvegicus*; Long Evans, neocortical cells; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Cell signaling/function-Cytotoxicity-Developmental and juvenile toxicology
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3479540

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent solvent control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided few details on exposure media preparation
	Metric 8:	Consistency of Exposure Administration	Low	The study provided few details on exposure administration
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were justified for a dose response by study authors
	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	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed
	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	2500 cells per well, did not see evidence of replicates being used
Domain 5: Outcome Assessment				

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Study Citation:	Behl, M., Hsieh, J. H., Shafer, T. J., Mundy, W. R., Rice, J. R., Boyd, W. A., Freedman, J. H., Hunter, E. S., Jarema, K. A., Padilla, S., Tice, R. R. (2015). Use of alternative assays to identify and prioritize organophosphorus flame retardants for potential developmental and neurotoxicity. Neurotoxicology and Teratology 52(Pt B):181-193.
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:	Vertebrate; Mammalian; *Rattus norvegicus*; Long Evans, neocortical cells; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Cell signaling/function-Cytotoxicity-Developmental and juvenile toxicology
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3479540

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

Additional Comments: rat neural cell outgrowth, viability and cytotoxicity

Overall Quality Determination Medium

Study Citation:	Behl, M., Hsieh, J. H., Shafer, T. J., Mundy, W. R., Rice, J. R., Boyd, W. A., Freedman, J. H., Hunter, E. S., Jarema, K. A., Padilla, S., Tice, R. R. (2015). Use of alternative assays to identify and prioritize organophosphorus flame retardants for potential developmental and neurotoxicity. Neurotoxicology and Teratology 52(Pt B):181-193.
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:	Vertebrate; Mammalian; *Rattus norvegicus*; Long Evans, neocortical cells; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Cell signaling/function-Cytotoxicity-Developmental and juvenile toxicology
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3479540

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent solvent control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and used as a comparator
	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 few details on exposure media preparation
	Metric 8:	Consistency of Exposure Administration	Low	The study provided few details on exposure administration
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were justified for a dose response by study authors
	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	Another cited methodology needed to assess this metric was unavailable, therefore this metric score reflects the details provided in the study being reviewed Harrill, 2015
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups
	Metric 15:	Number of Organisms and Replicates per Group	Medium	experiment was repeated at least three times
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 reported but few dtails were provided

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Study Citation:	Behl, M., Hsieh, J. H., Shafer, T. J., Mundy, W. R., Rice, J. R., Boyd, W. A., Freedman, J. H., Hunter, E. S., Jarema, K. A., Padilla, S., Tice, R. R. (2015). Use of alternative assays to identify and prioritize organophosphorus flame retardants for potential developmental and neurotoxicity. Neurotoxicology and Teratology 52(Pt B):181-193.
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:	Vertebrate; Mammalian; *Rattus norvegicus*; Long Evans, neocortical cells; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Cell signaling/function-Cytotoxicity-Developmental and juvenile toxicology
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3479540

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

Additional Comments: rat neural cell network activity

Overall Quality Determination Medium

Study Citation:	Fini, J. B., Riu, A., Debrauwer, L., Hillenweck, A., Mével, Le, S., Chevolleau, S., Boulahtouf, A., Palmier, K., Balaguer, P., Cravedi, J. P., Demeneix, B. A., Zalko, D. (2012). Parallel biotransformation of tetrabromobisphenol A in Xenopus laevis and mammals: Xenopus as a model for endocrine perturbation studies. Toxicological Sciences 125(2):359-367.
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:	Vertebrate; Amphibian; *Xenopus laevis*; In vitro Thyroid Hormone Receptor Construct; Adult
Health Outcome:	Mechanistic-Receptor binding/ regulation of receptor activity
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1293799

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was only identified by name (tetrabromobisphenol A, TBBPA), but no other descriptors such as CASRN or structure were reported.
	Metric 2:	Test Substance Source	High	Reporter in vitro cell assay: TBBPA or TBBPA-monosulfate. TBBPA (>97% purity) was purchased from Sigma-Aldrich. TBBPA-monosulfate was synthesized by the laboratory.
	Metric 3:	Test Substance Purity	High	Reporter in vitro cell assay: TBBPA (>97% purity) was purchased from Sigma-Aldrich. TBBPA-monosulfate was synthesized by the laboratory.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Reporter in vitro cell assay: Details are missing in the methods, but authors indicated a control group in Figure 4.
	Metric 5:	Negative Control Response	High	Reporter in vitro cell assay: The biological response (X. laevis TRalpha binding activity in the absence/presence of tri-iodothyronine) of the control group was reported and appeared appropriate (Figure 4).
	Metric 6:	Randomized Allocation	Low	Randomized allocation not stated.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study lacked details on how TBBPA and TBBPA-monosulfate solutions were prepared to add to the cell culture. the exposure solutions.
	Metric 8:	Consistency of Exposure Administration	High	Evidence suggests that exposure administration was administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	TBBPA or TBBPA-monosulfate were not measured in the vitro assays.
	Metric 10:	Exposure Duration and Frequency	Uninformative	X. laevis TRalpha using a reporter in vitro cell assay: The exposure duration or incubation period was not reported, it is unclear if bindings response readings were done immediately or after an incubation period which is typical of these reporter cell assays.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	X. laevis TRalpha using a reporter in vitro cell assay: cells were exposed to either TBBPA or TBBPA-monosulfate at 1, 3, and 10 uMconcentrations. But it unclear why these concentrations were selected.

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Study Citation:	Fini, J. B., Riu, A., Debrauwer, L., Hillenweck, A., Mével, Le, S., Chevolleau, S., Boulahtouf, A., Palmier, K., Balaguer, P., Cravedi, J. P., Demeneix, B. A., Zalko, D. (2012). Parallel biotransformation of tetrabromobisphenol A in Xenopus laevis and mammals: Xenopus as a model for endocrine perturbation studies. Toxicological Sciences 125(2):359-367.
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:	Vertebrate; Amphibian; *Xenopus laevis*; In vitro Thyroid Hormone Receptor Construct; Adult
Health Outcome:	Mechanistic-Receptor binding/ regulation of receptor activity
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1293799

Domain	Metric		Rating	Comments
	Metric 12:	Testing at or Below Solubility Limit	Uninformative	X. laevis TRalpha using a reporter in vitro cell assay: cells were exposed to either TBBPA or TBBPA-monosulfate at 1, 3, and 10 uM concentrations. Therefore, in vitro exposure concentrations were above the solubility limit (TBBPA's solubility limit is 4.15 mg/L at 25 C pH 7.56). It is unclear what solvents authors used in the in vitro assay.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The in vitro construct was described. X. laevis TRalpha construct transfected into in HeLa cells to perform the binding assay.
	Metric 14:	Acclimatization and Pretreatment Conditions	Medium	Authors did not mention acclimatizing cells used in the in vitro assay, but these details are less common of in vitro assays.
	Metric 15:	Number of Organisms and Replicates per Group	Low	X. laevis TRalpha using a reporter in vitro cell assay: Authors reported that luciferase assays were performed with the Promega dual-reporterkit according to the manufacturer's instructions. Authors reported that transfection assays were performed twice in triplicate. However, they did not report specific number of cells tested.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	These were in vitro transfection assays. The authors did not report how they maintained the HeLA cells used in the transfection assays.
	Metric 17:	Outcome Assessment Methodology	High	X. laevis TRalpha using a reporter in vitro cell assay: The outcome methodology (preparation of construct transfected into HeLA ceels for binding in vitro assay) was reported and appropriate for the intended outcome (TRalpha activity in the presence or absence of TBBPA and TBBPA-monosulfate).
	Metric 18:	Consistency of Outcome Assessment	High	The outcome assessment was consistent across 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.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were described (ANOVA Kruskal-Wallis test followed by Dunn's test).
	Metric 22:	Reporting of Data	High	X. laevis TRalpha using a reporter in vitro cell assay: Data were presented for each control and treatment groups (Figure 4).

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Study Citation:	Fini, J. B., Riu, A., Debrauwer, L., Hillenweck, A., Mével, Le, S., Chevolleau, S., Boulahtouf, A., Palmier, K., Balaguer, P., Cravedi, J. P., Demeneix, B. A., Zalko, D. (2012). Parallel biotransformation of tetrabromobisphenol A in Xenopus laevis and mammals: Xenopus as a model for endocrine perturbation studies. Toxicological Sciences 125(2):359-367.
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:	Vertebrate; Amphibian; *Xenopus laevis*; In vitro Thyroid Hormone Receptor Construct; Adult
Health Outcome:	Mechanistic-Receptor binding/ regulation of receptor activity
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	1293799

Domain	Metric		Rating	Comments
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This study examined thyroid disruption effects of TBBPA in Xenopus laevis. To do this the authors (1) assessed the kinetics of TBBPA (uptake and excretion) in X. laevis tadpoles, (2) investigated if antithyroid effects are due to the parent TBBPA or a metabolite using two readouts: tadpole morphology and an in vivo TH reporter assay, and (3) examined the ability of sulfate metabolites andTBBPA to exert direct actions on the TRalpha using a reporter in vitro cell assay. This form was used to evaluate the the ability of sulfate metabolites and TBBPA to exert direct actions on the X. laevis TRalpha using a reporter in vitro cell assay.

Overall Quality Determination — Uninformative

Study Citation:	Li, J., Dai, L., Feng, Y., Cao, Z., Ding, Y., Xu, H., Xu, A., Du, H. (2024). Multigenerational effects and mutagenicity of three flame retardants on germ cells in Caenorhabditis elegans. Ecotoxicology and Environmental Safety 269:115815.
Duration:	Overall Duration: Not-reported; 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*; Larvae
Health Outcome:	Mechanistic-Cell signaling/function-Genotox (including DNA repair)-Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	11365410

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The TBBPA was identified by CASRN.
	Metric 2:	Test Substance Source	Low	The TBBPA was reported to be purchased from Merck, but it was not reported to be analytically verified.
	Metric 3:	Test Substance Purity	Low	The purity was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of a concurrent negative control.
	Metric 5:	Negative Control Response	High	The negative control response was reported in Figure 4 A and D and was appropriate for the outcome of interest.
	Metric 6:	Randomized Allocation	Low	It was not reported how the worms were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The preparation of the test substance was not reported in detail. Little information was provided regarding the test system.
	Metric 8:	Consistency of Exposure Administration	Low	Little information was provided on the test substance preparation and the test system. This creates uncertainty regarding whether the exposure was carried out consistently.
	Metric 9:	Measurement of Test Substance Concentration	Low	It was not reported if the test substance was measured at any point.
	Metric 10:	Exposure Duration and Frequency	High	The transgenerational assay exposure was for 36h to the parental generation. This assay then continued through the F6 generation. This appeared adequate for the outcome of interest.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	There was only one exposure level for this portion of the study. The goal was to look for transgenerational effects after exposure of the parental generation to TBBPA.
	Metric 12:	Testing at or Below Solubility Limit	Low	This test concentration was above the water solubility limit, but details regarding the preparation of the test substance and the method of exposure, make it unclear if this is acceptable or not. Likely the exposure occurred via the NGS, but this was not specified. If this was the case, insolubility in water, may not be an issue.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Study authors reported the worms were from the Caenorhabditis Genetic Center in Minneapolis, MN, USA. The worms were reported to be synchronized and in the L4 larval stage at the start of the exposure.

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Study Citation:	Li, J., Dai, L., Feng, Y., Cao, Z., Ding, Y., Xu, H., Xu, A., Du, H. (2024). Multigenerational effects and mutagenicity of three flame retardants on germ cells in Caenorhabditis elegans. Ecotoxicology and Environmental Safety 269:115815.
Duration:	Overall Duration: Not-reported; 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*; Larvae
Health Outcome:	Mechanistic-Cell signaling/function-Genotox (including DNA repair)-Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	11365410

Domain	Metric		Rating	Comments
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the C. elegans were acclimated at any point prior to the start of the study.
	Metric 15:	Number of Organisms and Replicates per Group	Low	Worms were exposed in 24-well plates. However, it was unclear how many worms there were per plate well. It was reported that every treatment group was tested in triplicate.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Worms were cultivated in nematode growth media plates with E. coli strain OP50 as a food source. They were maintained at 20C in the dark. Biomass loading was unclear.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome(s) of interest–changes in germ cell apoptosis and number of oocytes in the F1-F6 generations after F0 exposure to TBBPA.
	Metric 18:	Consistency of Outcome Assessment	Low	It is unclear at what points the oocytes and the germ cell corpse per gonad arm were assessed or how they were assessed.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistics were reported in section 2.10 and were appropriate for the study.
	Metric 22:	Reporting of Data	High	Control and exposure results were reported in Figure 4 A and D.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.

Additional Comments: This evaluation was for the change in germ cell development as well oogenesis in F1-F6 generations after the F0 generation was exposed to TBBPA.

Overall Quality Determination Medium

Study Citation:	Li, J., Dai, L., Feng, Y., Cao, Z., Ding, Y., Xu, H., Xu, A., Du, H. (2024). Multigenerational effects and mutagenicity of three flame retardants on germ cells in Caenorhabditis elegans. Ecotoxicology and Environmental Safety 269:115815.
Duration:	Overall Duration: Not-reported; 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*; Larvae
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	11365410

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The TBBPA was identified by CASRN.
	Metric 2:	Test Substance Source	Low	The TBBPA was reported to be purchased from Merck, but it was not reported to be analytically verified.
	Metric 3:	Test Substance Purity	Low	The purity was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of a concurrent negative control.
	Metric 5:	Negative Control Response	High	The negative control response was reported in Figure 3A and was appropriate for the outcome of interest.
	Metric 6:	Randomized Allocation	Low	It was not reported how the worms were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The preparation of the test substance was not reported in detail. Little information was provided regarding the test system.
	Metric 8:	Consistency of Exposure Administration	Low	Little information was provided on the test substance preparation and the test system. This creates uncertainty regarding whether the exposure was carried out consistently.
	Metric 9:	Measurement of Test Substance Concentration	Low	It was not reported if the test substance was measured at any point.
	Metric 10:	Exposure Duration and Frequency	High	The brood assay had a 60h exposure to L1 stage worms to TBBPA. This was followed by transferring young-adult worms onto new NGS plates every 24h until egg laying stopped. All hatched larvae and unhatched eggs were counted under a dissecting scope. Progeny produced over 5 days were documented for brood size.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	There were four exposure levels. Spacing appeared adequate to observe the outcome interest.
	Metric 12:	Testing at or Below Solubility Limit	Medium	The highest test concentration was above the water solubility limit. The preparation of the test substance was not reported in detail, so it is unclear if a solvent was used.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Study authors reported the worms were from the Caenorhabditis Genetic Center in Minneapolis, MN, USA. The worms were reported to be synchronized and in the L4 larval stage at the start of the exposure.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the C. elegans were acclimated at any point prior to the start of the study.
	Metric 15:	Number of Organisms and Replicates per Group	Low	Worms were exposed in 24-well plates. However, it was unclear how many worms there were per plate well. It was reported that every treatment group was tested in triplicate.

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Study Citation:	Li, J., Dai, L., Feng, Y., Cao, Z., Ding, Y., Xu, H., Xu, A., Du, H. (2024). Multigenerational effects and mutagenicity of three flame retardants on germ cells in Caenorhabditis elegans. Ecotoxicology and Environmental Safety 269:115815.
Duration:	Overall Duration: Not-reported; 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*; Larvae
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	11365410

Domain	Metric		Rating	Comments
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Worms were cultivated in nematode growth media plates with E. coli strain OP50 as a food source. They were maintained at 20C in the dark. Biomass loading was unclear.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome(s) of interest–changes in brood size due to TBBPA exposure.
	Metric 18:	Consistency of Outcome Assessment	High	Brood size was assessed after a 60h exposure after which worms were transferred to new NGS plates every 24h until egg laying ceased. All hatched larvae and unhatched eggs were counted under a dissecting scope, and progeny produced over a five day period was documented as brood size.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistics were reported in section 2.10 and were appropriate for the study.
	Metric 22:	Reporting of Data	High	Control and exposure results were reported in Figure 3A.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.

Additional Comments: This evaluation was for the change in brood size after exposure of L1-stage C. elegans larvae to four TBBPA test concentrations for 60h. Results are reported in Fig. 3A. The outcome of interest was reproduction. There were other reproductive outcomes in this study, but those were conducted in a separate test, so are included in a different evaluation form.

Overall Quality Determination Medium

Study Citation:	Li, J., Dai, L., Feng, Y., Cao, Z., Ding, Y., Xu, H., Xu, A., Du, H. (2024). Multigenerational effects and mutagenicity of three flame retardants on germ cells in Caenorhabditis elegans. Ecotoxicology and Environmental Safety 269:115815.
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*; Larvae
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	11365410

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The TBBPA was identified by CASRN.
	Metric 2:	Test Substance Source	Low	The TBBPA was reported to be purchased from Merck, but it was not reported to be analytically verified.
	Metric 3:	Test Substance Purity	Low	The purity was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of a concurrent negative control.
	Metric 5:	Negative Control Response	High	The negative control response was reported in Figure 1A and was appropriate for the outcome of interest.
	Metric 6:	Randomized Allocation	Low	It was not reported how the worms were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The preparation of the test substance was not reported in detail. Little information was provided regarding the test system.
	Metric 8:	Consistency of Exposure Administration	Low	Little information was provided on the test substance preparation and the test system. This creates uncertainty regarding whether the exposure was carried out consistently.
	Metric 9:	Measurement of Test Substance Concentration	Low	It was not reported if the test substance was measured at any point.
	Metric 10:	Exposure Duration and Frequency	High	The locomotion assay exposure duration was reported to be 24h. Basal activities were then measured for 24h. This appeared adequate to observe the outcome of interest.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	There were four exposure levels. Spacing appeared adequate to observe the outcome interest.
	Metric 12:	Testing at or Below Solubility Limit	Medium	The highest test concentration was above the water solubility limit. The preparation of the test substance was not reported in detail, so it is unclear if a solvent was used.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Study authors reported the worms were from the Caenorhabditis Genetic Center in Minneapolis, MN, USA. The worms were reported to be synchronized and in the L4 larval stage at the start of the exposure.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the C. elegans were acclimated at any point prior to the start of the study.
	Metric 15:	Number of Organisms and Replicates per Group	Low	Worms were exposed in 24-well plates. However, it was unclear how many worms there were per plate well. It was reported that every treatment group was tested in triplicate.
Domain 5: Outcome Assessment				

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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*; Larvae
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	11365410

Domain	Metric		Rating	Comments
	Metric 16:	Adequacy of Test Conditions	Medium	Worms were cultivated in nematode growth media plates with E. coli strain OP50 as a food source. They were maintained at 20C in the dark. Biomass loading was unclear.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome(s) of interest–changes in locomotive activities after exposure. This was assessed using a WMicrotracker.
	Metric 18:	Consistency of Outcome Assessment	High	Locomotion was assessed using a WMicrotracker. Following exposure, worms were transferred to M9 buffer into a 96-well flat microtiter plated sealed with parafilm. After 1h rest, basal activities were measured for 24h.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistics were reported in section 2.10 and were appropriate for the study.
	Metric 22:	Reporting of Data	High	Control and exposure results were reported in Figure 1A and were adequate.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.

Additional Comments: This evaluation was for the change in locomotive activity in L4 larval C. elegans after exposure to TBBPA at four different concentrations. WMicrotracker was used to evaluate the activity. Behavioral was selected as the outcome of interest.

Overall Quality Determination Medium

Study Citation:	Li, J., Dai, L., Feng, Y., Cao, Z., Ding, Y., Xu, H., Xu, A., Du, H. (2024). Multigenerational effects and mutagenicity of three flame retardants on germ cells in Caenorhabditis elegans. Ecotoxicology and Environmental Safety 269:115815.
Duration:	Overall Duration: Not-reported; 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*; Larvae
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	11365410

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The TBBPA was identified by CASRN.
	Metric 2:	Test Substance Source	Low	The TBBPA was reported to be purchased from Merck, but it was not reported to be analytically verified.
	Metric 3:	Test Substance Purity	Low	The purity was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of a concurrent negative control.
	Metric 5:	Negative Control Response	High	The negative control response was reported in Figure 4 G and H and was appropriate for the outcome of interest.
	Metric 6:	Randomized Allocation	Low	It was not reported how the worms were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The preparation of the test substance was not reported in detail. Little information was provided regarding the test system.
	Metric 8:	Consistency of Exposure Administration	Low	Little information was provided on the test substance preparation and the test system. This creates uncertainty regarding whether the exposure was carried out consistently.
	Metric 9:	Measurement of Test Substance Concentration	Low	It was not reported if the test substance was measured at any point.
	Metric 10:	Exposure Duration and Frequency	High	The transgenerational assay exposure was for 36h to the parental generation. This assay then continued through the F6 generation. This appeared adequate for the outcome of interest.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	There was only one exposure level for this portion of the study. The goal was to look for transgenerational effects after exposure of the parental generation to TBBPA.
	Metric 12:	Testing at or Below Solubility Limit	Low	This test concentration was above the water solubility limit, but details regarding the preparation of the test substance and the method of exposure, make it unclear if this is acceptable or not. Likely the exposure occurred via the NGS, but this was not specified. If this was the case, insolubility in water, may not be an issue.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Study authors reported the worms were from the Caenorhabditis Genetic Center in Minneapolis, MN, USA. The worms were reported to be synchronized and in the L4 larval stage at the start of the exposure.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the C. elegans were acclimated at any point prior to the start of the study.

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Study Citation:	Li, J., Dai, L., Feng, Y., Cao, Z., Ding, Y., Xu, H., Xu, A., Du, H. (2024). Multigenerational effects and mutagenicity of three flame retardants on germ cells in Caenorhabditis elegans. Ecotoxicology and Environmental Safety 269:115815.
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Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Caenorhabditis elegans*; Larvae
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	11365410

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Low	Worms were exposed in 24-well plates. However, it was unclear how many worms there were per plate well. It was reported that every treatment group was tested in triplicate.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Worms were cultivated in nematode growth media plates with E. coli strain OP50 as a food source. They were maintained at 20C in the dark. Biomass loading was unclear.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome(s) of interest–changes in brood size in several generations due to TBBPA exposure after exposure to the F0 generation.
	Metric 18:	Consistency of Outcome Assessment	Low	Brood size was assessed for F1 and F2 progenies after the F0 generation was exposed to 10mg/L of TBBPA. It was unclear when and how the broods were assessed after the 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	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	Statistics were reported in section 2.10 and were appropriate for the study.
	Metric 22:	Reporting of Data	High	Control and exposure results were reported in Figure 4 G and H.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.

Additional Comments: This evaluation was for the change in brood size after exposure of L3-stage C. elegans larvae to 10mg/L of TBBPA for 36h in the F0 generation. Brood sizes from the F1 and F2 generations were assessed. Reproduction was the outcome of interest.

Overall Quality Determination Medium

Study Citation:	Lieke, T., Steinberg, C. E., Ju, J., Saul, N. (2015). Natural Marine and Synthetic Xenobiotics Get on Nematode's Nerves: Neuro-Stimulating and Neurotoxic Findings in Caenorhabditis elegans. Marine Drugs 13(5):2785-812. [Marine drugs].
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*; BZ555; Larvae
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Cytotoxicity-Neurotoxicology
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555762

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 >97%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Low	The study provided few details on consistency of exposure administration
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	Medium	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	There were two TBBPA treatment groups.
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The 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	Environmental conditions were not sufficiently reported to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	the assessment methodology was sensitive and appropriate for the outcomes of interest.

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Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Caenorhabditis elegans*; BZ555; Larvae
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Cytotoxicity-Neurotoxicology
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555762

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

Additional Comments: None

Overall Quality Determination Medium

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Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Caenorhabditis elegans*; N2; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Neurotoxicology
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555762

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	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 >97%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Low	The study provided few details on consistency of exposure administration
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	Medium	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	There were deficiencies regarding the number of exposure groups
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The 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	Environmental conditions were not sufficiently reported to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	the assessment methodology was sensitive and appropriate for the outcomes of interest.
	Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups

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Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Caenorhabditis elegans*; N2; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Neurotoxicology
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555762

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

Additional Comments: multiple behavioral measurements, author referred to several tables that may be part of supplemental information but I wasn't able to find a link or a means of accessing them (Table S1, S2...), mixtures also reported

Overall Quality Determination Medium

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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*; N2; Larvae
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555762

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 >97%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Low	The study provided few details on consistency of exposure administration
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	Medium	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	There were deficiencies regarding the number of exposure groups. Only a low concentration (0.1 uM) and high (50 uM) were used, with no intervening concentrations that would have allowed a better characterization of the dose-response relationship.
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The 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	Environmental conditions were not sufficiently reported to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	the assessment methodology was sensitive and appropriate for the outcomes of interest.
	Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups

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Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555762

Domain	Metric		Rating	Comments
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
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Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Caenorhabditis elegans*; DA1240; Larvae
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Cytotoxicity-Neurotoxicology
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
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Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
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Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
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	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
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HERO ID:	5555762

Domain	Metric		Rating	Comments
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described
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Additional Comments: None

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Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Caenorhabditis elegans*; EG1285; Larvae
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Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Caenorhabditis elegans*; EG1285; Larvae
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Additional Comments: None

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Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Caenorhabditis elegans*; LX929; Larvae
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Cytotoxicity-Neurotoxicology
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Domain 6: Confounding / Variable Control				
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Study Citation:	Lieke, T., Steinberg, C. E., Ju, J., Saul, N. (2015). Natural Marine and Synthetic Xenobiotics Get on Nematode's Nerves: Neuro-Stimulating and Neurotoxic Findings in Caenorhabditis elegans. Marine Drugs 13(5):2785-812. [Marine drugs].
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*; N2; Larvae
Health Outcome:	Gastrointestinal
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555762

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 >97%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Low	The study provided few details on consistency of exposure administration
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	Medium	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	There were deficiencies regarding the number of exposure groups. Only a low concentration (0.1 uM) and high (50 uM) were used, with no intervening concentrations that would have allowed a better characterization of the dose-response relationship.
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The 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	Environmental conditions were not sufficiently reported to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	the assessment methodology was sensitive and appropriate for the outcomes of interest.
	Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups

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Study Citation:	Lieke, T., Steinberg, C. E., Ju, J., Saul, N. (2015). Natural Marine and Synthetic Xenobiotics Get on Nematode's Nerves: Neuro-Stimulating and Neurotoxic Findings in Caenorhabditis elegans. Marine Drugs 13(5):2785-812. [Marine drugs].
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*; N2; Larvae
Health Outcome:	Gastrointestinal
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555762

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

Additional Comments: This evaluation is for the assessment of defecation.

Overall Quality Determination Medium

Study Citation:	Lieke, T., Steinberg, C. E., Ju, J., Saul, N. (2015). Natural Marine and Synthetic Xenobiotics Get on Nematode's Nerves: Neuro-Stimulating and Neurotoxic Findings in Caenorhabditis elegans. Marine Drugs 13(5):2785-812. [Marine drugs].
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*; N2; Larvae
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Cytotoxicity-Neurotoxicology
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555762

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 >97%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Low	The study provided few details on consistency of exposure administration
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	Medium	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	There were two TBBPA treatment groups.
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The 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	Environmental conditions were not sufficiently reported to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	the assessment methodology was sensitive and appropriate for the outcomes of interest.
	Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups

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Study Citation:	Lieke, T., Steinberg, C. E., Ju, J., Saul, N. (2015). Natural Marine and Synthetic Xenobiotics Get on Nematode's Nerves: Neuro-Stimulating and Neurotoxic Findings in Caenorhabditis elegans. Marine Drugs 13(5):2785-812. [Marine drugs].
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*; N2; Larvae
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Cytotoxicity-Neurotoxicology
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555762

Domain	Metric		Rating	Comments
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were 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:	Lieke, T., Steinberg, C. E., Ju, J., Saul, N. (2015). Natural Marine and Synthetic Xenobiotics Get on Nematode's Nerves: Neuro-Stimulating and Neurotoxic Findings in Caenorhabditis elegans. Marine Drugs 13(5):2785-812. [Marine drugs].
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*; GR1366; Larvae
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Cytotoxicity-Neurotoxicology
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555762

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 >97%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Low	The study provided few details on consistency of exposure administration
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	Medium	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	There were two TBBPA treatment groups.
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The 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	Environmental conditions were not sufficiently reported to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	the assessment methodology was sensitive and appropriate for the outcomes of interest.
	Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups

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Study Citation:	Lieke, T., Steinberg, C. E., Ju, J., Saul, N. (2015). Natural Marine and Synthetic Xenobiotics Get on Nematode's Nerves: Neuro-Stimulating and Neurotoxic Findings in Caenorhabditis elegans. Marine Drugs 13(5):2785-812. [Marine drugs].
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*; GR1366; Larvae
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Cytotoxicity-Neurotoxicology
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555762

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

Additional Comments: None

Overall Quality Determination Medium

Study Citation:	Liu, F., Zaman, W. Q., Peng, H., Li, C., Cao, X., Huang, K., Cui, C., Zhang, W., Lin, K., Luo, Q. (2019). Ecotoxicity of Caenorhabditis elegans following a step and repeated chronic exposure to tetrabromobisphenol A. Ecotoxicology and Environmental Safety 169:273-281.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater), Terrestrial; Water, 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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5497487

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was 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	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for this study type
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of replicates was not reported
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Reporting omissions were unlikely to have a substantial impact on results
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest

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Study Citation:	Liu, F., Zaman, W. Q., Peng, H., Li, C., Cao, X., Huang, K., Cui, C., Zhang, W., Lin, K., Luo, Q. (2019). Ecotoxicity of Caenorhabditis elegans following a step and repeated chronic exposure to tetrabromobisphenol A. Ecotoxicology and Environmental Safety 169:273-281.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater), Terrestrial; Water, 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:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5497487

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
	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: None

Overall Quality Determination High

Study Citation:	Liu, F., Zaman, W. Q., Peng, H., Li, C., Cao, X., Huang, K., Cui, C., Zhang, W., Lin, K., Luo, Q. (2019). Ecotoxicity of Caenorhabditis elegans following a step and repeated chronic exposure to tetrabromobisphenol A. Ecotoxicology and Environmental Safety 169:273-281.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater), Terrestrial; Water, 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:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5497487

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was 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	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for this study type
	Metric 12:	Testing at or Below Solubility Limit	High	Exposure concentrations were at or below the water solubility limit
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of replicates was not reported
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Reporting omissions were unlikely to have a substantial impact on results
	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				

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Study Citation:	Liu, F., Zaman, W. Q., Peng, H., Li, C., Cao, X., Huang, K., Cui, C., Zhang, W., Lin, K., Luo, Q. (2019). Ecotoxicity of Caenorhabditis elegans following a step and repeated chronic exposure to tetrabromobisphenol A. Ecotoxicology and Environmental Safety 169:273-281.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Aquatic (freshwater), Terrestrial; Water, 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:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5497487

Domain	Metric		Rating	Comments
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups
	Metric 20:	Outcomes Unrelated to Exposure	High	there were no differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	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 High

Study Citation:	Saul, N., Baberschke, N., Chakrabarti, S., Stürzenbaum, S. R., Lieke, T., Menzel, R., Jonáš, A., Steinberg, C. E. (2014). Two organobromines trigger lifespan, growth, reproductive and transcriptional changes in Caenorhabditis elegans. Environmental Science and Pollution Research 21(17):10419-10431.
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*; strain N2 (var. Bristol); Larvae
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5549514

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Nomenclature of test substance was provided. No other information (structure, CASRN, etc.) was given,
	Metric 2:	Test Substance Source	Low	The source was reported but the test substance identity was NOT analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Controls were not mentioned in the methods but Fig 5 shows control results. Unclear whether it was solvent or blank control.
	Metric 5:	Negative Control Response	Medium	The biological responses of the negative control group was reported in Fig 5. Unclear whether it was solvent or blank control response.
	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 concentration of test substance was not measured during the study.
	Metric 8:	Consistency of Exposure Administration	Low	Details of exposure administration were not given in sufficient detail. The exposure duration for F0 generation were not reported. L4 larvae of F1 generation were moved to new treatment plates each day until reproduction was completed. In a second experiment, gravid animals of F0 generation were moved to treatment plates for 30 min and number of offspring counted after 75 hours Unclear whether treatment groups and control were treated the same way in both experiments.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	Medium	The duration of exposure of F1 larvae was 4 days for obtaining the total number of offspring and for F0 larvae, the duration of exposure was 30 minutes for initial reproduction counts recorded at 75 hours after egg stage. These durations are appropriate for the outcome of interest
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	5 exposure groups were tested for the reproductive assay of F1 larvae (Fig 5b) and 2 exposure groups were tested for the reproductive assay of F0 larvae (Fig 5d), and were adequate to address the purpose of the study.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure medium was solid NGM.

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Study Citation:	Saul, N., Baberschke, N., Chakrabarti, S., Stürzenbaum, S. R., Lieke, T., Menzel, R., Jonáš, A., Steinberg, C. E. (2014). Two organobromines trigger lifespan, growth, reproductive and transcriptional changes in Caenorhabditis elegans. Environmental Science and Pollution Research 21(17):10419-10431.
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*; strain N2 (var. Bristol); Larvae
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5549514

Domain	Metric		Rating	Comments
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized and/or whether pre-treatment conditions were the same for control and exposed groups.
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of replicates was not reported. The figure caption (for fig 5) mention that 27-30 nematodes were used for the reproductive assay of F1 larvae and 20 nematodes per concentration was used for the reproductive assay of F0 larvae.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reporting of housing and environmental conditions were not sufficiently reported to evaluate if adequate and whether differences occurred between control and exposed populations.
	Metric 17:	Outcome Assessment Methodology	Low	Outcome assessment methodology was not clearly reported. It was reported that the number of offspring were counted but no specific details were given.
	Metric 18:	Consistency of Outcome Assessment	Medium	The number of offspring were counted daily for the reproductive assay of F1 larvae and total numbers were counted after 75 hours from the second day of adulthood for the reproductive assay of F0 larvae. Unclear whether controls and exposure groups were assessed similarly. Incomplete reporting of minor details of outcome assessment protocol execution.
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	Statistical analysis was performed but not described adequately. It was reported that Student's t-test was conducted for results of reproductive assays, which is not an appropriate test when more than 2 exposure groups were involved.
	Metric 22:	Reporting of Data	High	The number of offspring/ day and total counts for the exposure groups and controls were given for the reproductive assay of F1 larvae (Fog 5 a) and total number of offspring (after 75 hours from the second day of adulthood) for the reproductive assay of F0 larvae. (Fig 5 d) were given as a percentage of the control.
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

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Study Citation:	Saul, N., Baberschke, N., Chakrabarti, S., Stürzenbaum, S. R., Lieke, T., Menzel, R., Jonáš, A., Steinberg, C. E. (2014). Two organobromines trigger lifespan, growth, reproductive and transcriptional changes in Caenorhabditis elegans. Environmental Science and Pollution Research 21(17):10419-10431.
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*; strain N2 (var. Bristol); Larvae
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5549514

Domain	Metric	Rating	Comments
Additional Comments:	This evaluation form pertains to reproductive outcome obtained for F1 and F0 generation following exposure of C. elegans to TBBPA concentrations ranging from 0.1-50uM.		

Overall Quality Determination Low

Study Citation:	Saul, N., Baberschke, N., Chakrabarti, S., Stürzenbaum, S. R., Lieke, T., Menzel, R., Jonáš, A., Steinberg, C. E. (2014). Two organobromines trigger lifespan, growth, reproductive and transcriptional changes in Caenorhabditis elegans. Environmental Science and Pollution Research 21(17):10419-10431.
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*; strain N2 (var. Bristol); Larvae
Health Outcome:	Mechanistic-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5549514

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Nomenclature of test substance was provided. No other information (structure, CASRN, etc.) was given,
	Metric 2:	Test Substance Source	Low	The source was reported but the test substance identity was NOT analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Controls were not mentioned in the methods but in the results section, it was reported that a solvent control group was used.
	Metric 5:	Negative Control Response	High	The biological responses of the negative control group was adequate (provided in supplemental information available online; https://link.springer.com/article/10.1007/s11356-014-2932-6#Sec20)
	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 concentration of test substance was not measured during the study.
	Metric 8:	Consistency of Exposure Administration	Medium	Exposure administration details were reported and appears to be consistent based on the details in the methods and results section.
	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 for 3 days (given in results section/page 10424) and was justified by the authors.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	2 exposure groups (0.1 and 50 µM TBBP) were tested for the gene expression assays and was adequate to address the purpose of the study.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure medium was solid NGM.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized and/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 organism per test concentration was not reported. Experiment was conducted in triplicates.

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Study Citation:	Saul, N., Baberschke, N., Chakrabarti, S., Stürzenbaum, S. R., Lieke, T., Menzel, R., Jonáš, A., Steinberg, C. E. (2014). Two organobromines trigger lifespan, growth, reproductive and transcriptional changes in Caenorhabditis elegans. Environmental Science and Pollution Research 21(17):10419-10431.
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*; strain N2 (var. Bristol); Larvae
Health Outcome:	Mechanistic-Cell signaling/function
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5549514

Domain	Metric		Rating	Comments
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reporting of housing and environmental conditions were not sufficiently reported to evaluate if adequate and whether differences occurred between control and exposed populations.
	Metric 17:	Outcome Assessment Methodology	High	Outcome assessment methodology (RNA preparation, DNA microarray and qRt-PCR) reported the intended outcome(s) of interest.
	Metric 18:	Consistency of Outcome Assessment	Medium	Incomplete reporting of minor details of outcome assessment protocol execution. Outcome assessment was conducted on the first day of adulthood but unclear how that was verified and whether all study groups and controls reached adulthood at the same time.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure (e.g., infection) that could influence the outcome assessment.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group (provided in supplemental information available online; https://link.springer.com/article/10.1007/s11356-014-2932-6#Sec20).
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This evaluation form pertains to mechanistic endpoints. The experimental design for mechanistic experiment was different from other assays that looked at apical endpoints within the study. The parent generation was not exposed to the test substance for mechanistic study.

Overall Quality Determination Medium

Study Citation:	Saul, N., Baberschke, N., Chakrabarti, S., Stürzenbaum, S. R., Lieke, T., Menzel, R., Jonáš, A., Steinberg, C. E. (2014). Two organobromines trigger lifespan, growth, reproductive and transcriptional changes in Caenorhabditis elegans. Environmental Science and Pollution Research 21(17):10419-10431.
Duration:	Overall Duration: > 21 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Caenorhabditis elegans*; strain N2 (var. Bristol); Larvae
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5549514

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Nomenclature of test substance was provided. No other information (structure, CASRN, etc.) was given.
	Metric 2:	Test Substance Source	Low	The source was reported but the test substance identity was NOT analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	”Treated and untreated nematodes” were used in the experiment. Unclear whether this is a solvent or blank control.
	Metric 5:	Negative Control Response	Uninformative	The biological responses of the negative control group was reported (Fig 3). Mortality in controls was > 30%.
	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 concentration of test substance was not measured during the study.
	Metric 8:	Consistency of Exposure Administration	Low	Details of exposure administration were not given in sufficient detail. The exposure duration for F0 generation were not reported. Nematodes were transferred daily to new plates but unclear whether controls and treatment groups were treated similarly.
	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 for six days, after which ”treated and untreated nematodes were moved either to 35 °C for 6.5 h”.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	Only two exposure groups were tested (0.1 and 50uM) and was not justified by the authors.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure medium was solid NGM.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized and/or whether pre-treatment conditions were the same for control and exposed groups.

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Study Citation:	Saul, N., Baberschke, N., Chakrabarti, S., Stürzenbaum, S. R., Lieke, T., Menzel, R., Jonáš, A., Steinberg, C. E. (2014). Two organobromines trigger lifespan, growth, reproductive and transcriptional changes in Caenorhabditis elegans. Environmental Science and Pollution Research 21(17):10419-10431.
Duration:	Overall Duration: > 21 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Caenorhabditis elegans*; strain N2 (var. Bristol); Larvae
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5549514

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of replicates was not reported.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reporting of housing and environmental conditions were not sufficiently reported to evaluate if adequate and whether differences occurred between control and exposed populations.
	Metric 17:	Outcome Assessment Methodology	Low	Outcome assessment methodology was not clearly reported. It was reported that surviving and dead organisms were counted starting on the 6th day of adulthood after moving to 35oC for 6.5 hours. Unclear whether authors confirmed death of nematodes when they failed do respond to contact stimuli as reported for the lifespan assay.
	Metric 18:	Consistency of Outcome Assessment	Medium	Outcome assessment appears to be consistent. It was reported that surviving and dead organisms were counted starting on the 6th day of adulthood after moving to 35oC for 6.5 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	Low	Mortality was assessed on the 6th day of adulthood after moving the nematodes to 35oC for 6.5 hours. Temperature may have had an an effect on mortality.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22:	Reporting of Data	Medium	Percent survivors for 2 treatment groups and and control was shown in Fig 3
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This evaluation form pertains to mortality outcome measured as part of thermal stress resistance assay following exposure to TBBPA concentrations (0.1uM and 50uM) for 6 days,

Overall Quality Determination Uninformative

Study Citation:	Saul, N., Baberschke, N., Chakrabarti, S., Stürzenbaum, S. R., Lieke, T., Menzel, R., Jonáš, A., Steinberg, C. E. (2014). Two organobromines trigger lifespan, growth, reproductive and transcriptional changes in Caenorhabditis elegans. Environmental Science and Pollution Research 21(17):10419-10431.
Duration:	Overall Duration: > 21 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Caenorhabditis elegans*; strain N2 (var. Bristol); Larvae
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5549514

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Nomenclature of test substance was provided. No other information (structure, CASRN, etc.) was given,
	Metric 2:	Test Substance Source	Low	The source was reported but the test substance identity was NOT analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Use of 'treated' and 'untreated' nematodes mentioned in the methods but unclear whether it was solvent control or blank control.
	Metric 5:	Negative Control Response	Medium	The biological responses of the negative control group was not explicitly reported. Results were reported as percentage of the control.
	Metric 6:	Randomized Allocation	Medium	Test organisms were randomly selected and transferred to treatment plates.
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 concentration of test substance was not measured during the study.
	Metric 8:	Consistency of Exposure Administration	Low	Details of exposure administration were not given in sufficient detail. The exposure duration for F0 generation were not reported. F1 larvae were moved to new treatment plates and then on 6th day of adulthood, they were moved to 45oC for 2.5 hours. Unclear whether treatment groups and control were treated the same way and how they were subjected to high temperature.
	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 6 days and was appropriate for the outcome of interest. This test was conducted as part of a thermal stress resistance and nematodes were moved to 45oC for 2.5 hours before measuring the body length.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	5 exposure groups were tested and were adequate to address the purpose of the study.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure medium was solid NGM.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized and/or whether pretreatment conditions were the same for control and exposed groups.

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Study Citation:	Saul, N., Baberschke, N., Chakrabarti, S., Stürzenbaum, S. R., Lieke, T., Menzel, R., Jonáš, A., Steinberg, C. E. (2014). Two organobromines trigger lifespan, growth, reproductive and transcriptional changes in Caenorhabditis elegans. Environmental Science and Pollution Research 21(17):10419-10431.
Duration:	Overall Duration: > 21 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Caenorhabditis elegans*; strain N2 (var. Bristol); Larvae
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5549514

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of replicates was not reported. The figure caption (for fig 4) mention that 3-5 trails were conducted.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reporting of housing and environmental conditions were not sufficiently reported to evaluate if adequate and whether differences occurred between control and exposed populations.
	Metric 17:	Outcome Assessment Methodology	Medium	Outcome assessment methodology was reported. Body length was measured by means of a digital Microscope at 100-fold magnification.
	Metric 18:	Consistency of Outcome Assessment	Medium	Outcome assessment appears to be consistent. Body length measurements were taken on the sixth day of adulthood, after moving the nematodes to 45oC for 2.5 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	Low	The nematodes were subjected to high temperature (45oC for 2.5 hours) before body length was measured. High temperature may have had an effect on body length.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately. It was reported that Student's t-test was conducted for body length measurements, which is not an appropriate test when more than 2 exposure groups were involved.
	Metric 22:	Reporting of Data	Medium	Body length data was given as percentage of the control body length measurements. (Fig 4).
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: This evaluation form pertains to C. elegans growth outcome (body length) following exposure to TBBPA concentration ranging from 0.1-50uM.

Overall Quality Determination Low

Study Citation:	Saul, N., Baberschke, N., Chakrabarti, S., Stürzenbaum, S. R., Lieke, T., Menzel, R., Jonáš, A., Steinberg, C. E. (2014). Two organobromines trigger lifespan, growth, reproductive and transcriptional changes in Caenorhabditis elegans. Environmental Science and Pollution Research 21(17):10419-10431.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Caenorhabditis elegans*; strain N2 (var. Bristol); Larvae
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5549514

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Nomenclature of test substance was provided. No other information (structure, CASRN, etc.) was given,
	Metric 2:	Test Substance Source	Low	The source was reported but the test substance identity was NOT analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity and/or grade of test substance were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	A negative control group was not mentioned in the methods but results were expressed as a percentage of the control (Figures 1 and 2) . Unclear whether it is a solvent control or not.
	Metric 5:	Negative Control Response	Medium	The biological responses of the negative control group was reported (Fig 1 and 2) but survival variability in the control was not shown.
	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 concentration of test substance was not measured during the study.
	Metric 8:	Consistency of Exposure Administration	Low	Details of exposure administration were not given in sufficient detail. It was reported that adult nematodes were moved to new treatment plates regularly, not clear whether this was done consistently for control and treatment 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 and exposure frequency were not clearly reported. From the results figures, it looks like the experiments (exposure to 5 treatments groups of TBBPA (Fig 1) and exposure to 2 treatments groups in mixture study (Fig 2)) and was conducted for 27 days. It was reported that adult nematodes were moved to new treatment plates regularly but the frequency was not reported.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	5 exposure groups were used for lifespan assay (Fig 1) and 2 exposure groups were used for the mixture study (Fig 2), and were adequate to address the purpose of the study.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure medium was solid NGM.
Domain 4: Test Organism				

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Study Citation:	Saul, N., Baberschke, N., Chakrabarti, S., Stürzenbaum, S. R., Lieke, T., Menzel, R., Jonáš, A., Steinberg, C. E. (2014). Two organobromines trigger lifespan, growth, reproductive and transcriptional changes in Caenorhabditis elegans. Environmental Science and Pollution Research 21(17):10419-10431.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Caenorhabditis elegans*; strain N2 (var. Bristol); Larvae
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5549514

Domain	Metric		Rating	Comments
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether test organisms were acclimatized and/or whether pre-treatment conditions were the same for control and exposed groups.
	Metric 15:	Number of Organisms and Replicates per Group	Low	The number of replicates was not reported for life span assays using single chemical and mixtures.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Reporting of housing and environmental conditions were not sufficiently reported to evaluate if adequate and whether differences occurred between control and exposed populations.
	Metric 17:	Outcome Assessment Methodology	Low	Outcome assessment methodology was not clearly reported. Surviving and dead organisms were counted starting the first day of adulthood but identification methods for confirming adulthood were not provided. It was reported that nematodes suffering from internal hatch and those that escaped from the NGM agar were censored.
	Metric 18:	Consistency of Outcome Assessment	Low	Details regarding the execution of the study protocol for outcome assessment (e.g., timing of assessment across groups) were limited.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Low	” Nematodes suffering from internal hatch and those thatescaped from the NGM agar were censored”. This could mean that one or more study groups experienced disproportionate test organism attrition.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was performed but not described adequately.
	Metric 22:	Reporting of Data	Medium	Mean lifespan data was shown as a percentage of control data and Kaplan-Meier survival curve was shown only for the lowest and highest concentration (Fig 1). Survival variability in the control was not provided for lifespan assays using single chemical and mixtures (Fig 1 and 2).
	Metric 23:	Explanation of Unexpected Outcomes	Low	Insufficient information was provided to determine if excessive variability occurred in the controls

Additional Comments: This evaluation form is relevant to mortality outcome (lifespan and survivorship) in lifespan assays using single chemical (5 concentrations of TBBPA) and mixture study (2 concentrations of TBBPA).

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Study Citation:	Saul, N., Baberschke, N., Chakrabarti, S., Stürzenbaum, S. R., Lieke, T., Menzel, R., Jonáš, A., Steinberg, C. E. (2014). Two organobromines trigger lifespan, growth, reproductive and transcriptional changes in Caenorhabditis elegans. Environmental Science and Pollution Research 21(17):10419-10431.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Caenorhabditis elegans*; strain N2 (var. Bristol); Larvae
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5549514

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

Study Citation:	Li, J., Dai, L., Feng, Y., Cao, Z., Ding, Y., Xu, H., Xu, A., Du, H. (2024). Multigenerational effects and mutagenicity of three flame retardants on germ cells in Caenorhabditis elegans. Ecotoxicology and Environmental Safety 269:115815.
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported
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*; Larvae
Health Outcome:	Mechanistic-Cell signaling/function-Genotox (including DNA repair)-Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	11365410

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The TBBPA was identified by CASRN.
	Metric 2:	Test Substance Source	Low	The TBBPA was reported to be purchased from Merck, but it was not reported to be analytically verified.
	Metric 3:	Test Substance Purity	Low	The purity was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of a concurrent negative control.
	Metric 5:	Negative Control Response	High	The negative control response was reported in Figure 5 and Tables 1 and 2.
	Metric 6:	Randomized Allocation	Low	It was not reported how the worms were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The preparation of the test substance was not reported in detail. Little information was provided regarding the test system.
	Metric 8:	Consistency of Exposure Administration	Low	Little information was provided on the test substance preparation and the test system. This creates uncertainty regarding whether the exposure was carried out consistently.
	Metric 9:	Measurement of Test Substance Concentration	Low	It was not reported if the test substance was measured at any point.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration for this portion of the study was reported to be through the F4 generation. It appears the exposure was conducted through the F3 generation.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	There was one exposure level in this portion of the study to determine mutations in DNA.
	Metric 12:	Testing at or Below Solubility Limit	Low	The test concentration was above the water solubility limit, but the exposure methods were not reported in detail, so it is unclear if a solvent was used, and the exposure method was unclear.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Study authors reported the worms were from the Caenorhabditis Genetic Center in Minneapolis, MN, USA. The worms were reported to be synchronized and in the L4 larval stage at the start of the exposure.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the C. elegans were acclimated at any point prior to the start of the study.
	Metric 15:	Number of Organisms and Replicates per Group	Low	Worms were exposed in 24-well plates. However, it was unclear how many worms there were per plate well. It was reported that every treatment group was tested in triplicate.

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Study Citation:	Li, J., Dai, L., Feng, Y., Cao, Z., Ding, Y., Xu, H., Xu, A., Du, H. (2024). Multigenerational effects and mutagenicity of three flame retardants on germ cells in Caenorhabditis elegans. Ecotoxicology and Environmental Safety 269:115815.
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported
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*; Larvae
Health Outcome:	Mechanistic-Cell signaling/function-Genotox (including DNA repair)-Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	11365410

Domain	Metric		Rating	Comments
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Worms were cultivated in nematode growth media plates with E. coli strain OP50 as a food source. They were maintained at 20C in the dark. Biomass loading was unclear.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome(s) of interest–effects on germ cell mutations and mutation frequencies after exposure.
	Metric 18:	Consistency of Outcome Assessment	High	An EasyPure Genomic DNA Kit was used to extract genomic DNA and assess the outcomes of interest.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistics were reported in section 2.10 and were appropriate for the study.
	Metric 22:	Reporting of Data	High	Control and exposure results were reported in Figure 5 and in Tables 1 and 2.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.

Additional Comments: This evaluation was for the effect of TBBPA germ cell mutations including mutations frequencies, indels, and SNVs.

Overall Quality Determination Medium

Study Citation:	Li, J., Dai, L., Feng, Y., Cao, Z., Ding, Y., Xu, H., Xu, A., Du, H. (2024). Multigenerational effects and mutagenicity of three flame retardants on germ cells in Caenorhabditis elegans. Ecotoxicology and Environmental Safety 269:115815.
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported
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*; Larvae
Health Outcome:	Mechanistic-Cell signaling/function-Genotox (including DNA repair)-Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	11365410

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	The TBBPA was identified by CASRN.
	Metric 2:	Test Substance Source	Low	The TBBPA was reported to be purchased from Merck, but it was not reported to be analytically verified.
	Metric 3:	Test Substance Purity	Low	The purity was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of a concurrent negative control.
	Metric 5:	Negative Control Response	High	The negative control response was reported in Figure 2 and was appropriate for the outcome of interest.
	Metric 6:	Randomized Allocation	Low	It was not reported how the worms were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The preparation of the test substance was not reported in detail. Little information was provided regarding the test system.
	Metric 8:	Consistency of Exposure Administration	Low	Little information was provided on the test substance preparation and the test system. This creates uncertainty regarding whether the exposure was carried out consistently.
	Metric 9:	Measurement of Test Substance Concentration	Low	It was not reported if the test substance was measured at any point.
	Metric 10:	Exposure Duration and Frequency	Uninformative	The exposure duration for this portion of the study was not reported.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	There were four exposure levels. Spacing appeared adequate to observe the outcome interest.
	Metric 12:	Testing at or Below Solubility Limit	Medium	The highest test concentration was above the water solubility limit. The preparation of the test substance was not reported in detail, so it is unclear if a solvent was used.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Study authors reported the worms were from the Caenorhabditis Genetic Center in Minneapolis, MN, USA. The worms were reported to be synchronized and in the L4 larval stage at the start of the exposure.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the C. elegans were acclimated at any point prior to the start of the study.
	Metric 15:	Number of Organisms and Replicates per Group	Low	Worms were exposed in 24-well plates. However, it was unclear how many worms there were per plate well. It was reported that every treatment group was tested in triplicate.
Domain 5: Outcome Assessment				

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Study Citation:	Li, J., Dai, L., Feng, Y., Cao, Z., Ding, Y., Xu, H., Xu, A., Du, H. (2024). Multigenerational effects and mutagenicity of three flame retardants on germ cells in Caenorhabditis elegans. Ecotoxicology and Environmental Safety 269:115815.
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported
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*; Larvae
Health Outcome:	Mechanistic-Cell signaling/function-Genotox (including DNA repair)-Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	11365410

Domain	Metric		Rating	Comments
	Metric 16:	Adequacy of Test Conditions	Medium	Worms were cultivated in nematode growth media plates with E. coli strain OP50 as a food source. They were maintained at 20C in the dark. Biomass loading was unclear.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome(s) of interest–germ cell apoptosis and oogenesis after TBBPA exposure.
	Metric 18:	Consistency of Outcome Assessment	Low	Apoptotic cells were visualized under a Leica DM4B microscope stimulated by blue excitation light. The quantification method of the oocytes was unclear.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistics were reported in section 2.10 and were appropriate for the study.
	Metric 22:	Reporting of Data	High	Control and exposure results were reported in Figure 2.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.

Additional Comments: This evaluation was for the effect of TBBPA on germ cell apoptosis and oogenesis after exposure. The exposure duration was not reported for this portion of the study, so the this evaluation received an unacceptable rating.

Overall Quality Determination — Uninformative

Study Citation:	Lieke, T., Steinberg, C. E., Ju, J., Saul, N. (2015). Natural Marine and Synthetic Xenobiotics Get on Nematode's Nerves: Neuro-Stimulating and Neurotoxic Findings in Caenorhabditis elegans. Marine Drugs 13(5):2785-812. [Marine drugs].
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported
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*; LX929; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Neurotoxicology
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555762

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 >97%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Low	The study provided few details on consistency of exposure administration
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	Medium	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	There were deficiencies regarding the number of exposure groups
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The 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	Environmental conditions were not sufficiently reported to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	the assessment methodology was sensitive and appropriate for the outcomes of interest.

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Study Citation:	Lieke, T., Steinberg, C. E., Ju, J., Saul, N. (2015). Natural Marine and Synthetic Xenobiotics Get on Nematode's Nerves: Neuro-Stimulating and Neurotoxic Findings in Caenorhabditis elegans. Marine Drugs 13(5):2785-812. [Marine drugs].
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported
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*; LX929; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Neurotoxicology
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555762

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

Additional Comments: multiple behavioral measurements, author referred to several tables that may be part of supplemental information but I wasn't able to find a link or a means of accessing them (Table S1, S2...), mixtures also reported

Overall Quality Determination Medium

Study Citation:	Lieke, T., Steinberg, C. E., Ju, J., Saul, N. (2015). Natural Marine and Synthetic Xenobiotics Get on Nematode's Nerves: Neuro-Stimulating and Neurotoxic Findings in Caenorhabditis elegans. Marine Drugs 13(5):2785-812. [Marine drugs].
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported
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*; BZ555; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Neurotoxicology
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555762

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 >97%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Low	The study provided few details on consistency of exposure administration
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	Medium	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	There were deficiencies regarding the number of exposure groups
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The 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	Environmental conditions were not sufficiently reported to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	the assessment methodology was sensitive and appropriate for the outcomes of interest.
	Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups

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Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported
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*; BZ555; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Neurotoxicology
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555762

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

Additional Comments: multiple behavioral measurements, author referred to several tables that may be part of supplemental information but I wasn't able to find a link or a means of accessing them (Table S1, S2...), mixtures also reported

Overall Quality Determination Medium

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Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported
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*; GR1366; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Neurotoxicology
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555762

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 >97%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Low	The study provided few details on consistency of exposure administration
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	Medium	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	There were deficiencies regarding the number of exposure groups
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The 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	Environmental conditions were not sufficiently reported to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	the assessment methodology was sensitive and appropriate for the outcomes of interest.
	Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups

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Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported
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Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Caenorhabditis elegans*; GR1366; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Neurotoxicology
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555762

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

Additional Comments: multiple behavioral measurements, author referred to several tables that may be part of supplemental information but I wasn't able to find a link or a means of accessing them (Table S1, S2...), mixtures also reported

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Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported
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*; EG1285; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Neurotoxicology
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555762

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
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	Metric 3:	Test Substance Purity	High	Chemical purity reported as >97%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Low	The study provided few details on consistency of exposure administration
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	Medium	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	There were deficiencies regarding the number of exposure groups
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Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups
	Metric 15:	Number of Organisms and Replicates per Group	Medium	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	Environmental conditions were not sufficiently reported to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	the assessment methodology was sensitive and appropriate for the outcomes of interest.
	Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups

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Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported
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*; EG1285; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Neurotoxicology
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HERO ID:	5555762

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

Additional Comments: multiple behavioral measurements, author referred to several tables that may be part of supplemental information but I wasn't able to find a link or a means of accessing them (Table S1, S2...), mixtures also reported

Overall Quality Determination Medium

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Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported
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*; DA1240; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Neurotoxicology
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555762

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
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	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Low	The study provided few details on consistency of exposure administration
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	Medium	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	There were deficiencies regarding the number of exposure groups
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The 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	Environmental conditions were not sufficiently reported to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	the assessment methodology was sensitive and appropriate for the outcomes of interest.
	Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups

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Study Citation:	Lieke, T., Steinberg, C. E., Ju, J., Saul, N. (2015). Natural Marine and Synthetic Xenobiotics Get on Nematode's Nerves: Neuro-Stimulating and Neurotoxic Findings in Caenorhabditis elegans. Marine Drugs 13(5):2785-812. [Marine drugs].
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported
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*; DA1240; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Neurotoxicology
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555762

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

Additional Comments: multiple behavioral measurements, author referred to several tables that may be part of supplemental information but I wasn't able to find a link or a means of accessing them (Table S1, S2...), mixtures also reported

Overall Quality Determination Medium

Study Citation:	Lieke, T., Steinberg, C. E., Ju, J., Saul, N. (2015). Natural Marine and Synthetic Xenobiotics Get on Nematode's Nerves: Neuro-Stimulating and Neurotoxic Findings in Caenorhabditis elegans. Marine Drugs 13(5):2785-812. [Marine drugs].
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported
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*; N2; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Neurotoxicology
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555762

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 >97%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Low	The study provided few details on consistency of exposure administration
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	Medium	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	There were deficiencies regarding the number of exposure groups
	Metric 12:	Testing at or Below Solubility Limit	High	The solvent concentration was appropriate
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The 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	Environmental conditions were not sufficiently reported to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	the assessment methodology was sensitive and appropriate for the outcomes of interest.
	Metric 18:	Consistency of Outcome Assessment	High	Details of the outcome assessment protocol were reported, and outcomes were assessed consistently across study groups

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Study Citation:	Lieke, T., Steinberg, C. E., Ju, J., Saul, N. (2015). Natural Marine and Synthetic Xenobiotics Get on Nematode's Nerves: Neuro-Stimulating and Neurotoxic Findings in Caenorhabditis elegans. Marine Drugs 13(5):2785-812. [Marine drugs].
Duration:	Overall Duration: Not-reported; Exposure Duration: Not-reported
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*; N2; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Neurotoxicology
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5555762

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

Additional Comments: multiple behavioral measurements, author referred to several tables that may be part of supplemental information but I wasn't able to find a link or a means of accessing them (Table S1, S2...), multiple developmental stages examined, mixtures also reported

Overall Quality Determination Medium

Study Citation:	Gu, J., Chen, X., Wang, Y., Wang, L., Szlavecz, K., Ma, Y., Ji, R. (2020). Bioaccumulation, physiological distribution, and biotransformation of tetrabromobisphenol a (TBBPA) in the geophagous earthworm Metaphire guillelmi - hint for detoxification strategy. Journal of Hazardous Materials 388:122027.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Dermal (topical application)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Eisenia fetida*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6775143

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The TBBPA was identified by name only.
	Metric 2:	Test Substance Source	Low	The source of the non-radiolabeled TBBPA was reported to be Sigma Corp. It was not reported to be analytically verified. The 14C ring-labeled TBBPA was synthesized from uniformly 14C-ring -labeled phenol.
	Metric 3:	Test Substance Purity	High	The radiochemical purity was reported to be 99%, and the chemical purity was reported to be 97%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of a concurrent negative control in which no TBBPA was present as well as a solvent control for this portion of the study.
	Metric 5:	Negative Control Response	Low	The negative control response was not reported for this portion of the study.
	Metric 6:	Randomized Allocation	Low	It was not reported how the E. fetida were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	Details regarding the test substance preparation were not reported in great detail. The test substance was dissolved in dichloromethane solution and evenly dispersed on filter paper. The filter paper covered the inner wall of the test vials. The dichloromethane was allowed to evaporate and then the filter paper was wet again with ultrapure water. One test organism was added to each vial. Size of test vials was not reported, nor was photoperiod.
	Metric 8:	Consistency of Exposure Administration	Low	The experimental system was not reported in great detail, so consistency cannot be determined.
	Metric 9:	Measurement of Test Substance Concentration	Low	It was not reported if the test substance concentrations were measured at any point during this portion of the study.
	Metric 10:	Exposure Duration and Frequency	Low	The study duration was reported to be 24h. This is a shorter duration than is typical for a filter paper earthworm test, but the outcome of interest was able to be assessed.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	There were four exposure levels for the TBBPA exposure, which is a little lower than the standard five exposure levels in a filter paper test.
	Metric 12:	Testing at or Below Solubility Limit	N/A	This study was conducted on filter paper. The test substance was dissolved in dichloromethane and then dispersed evenly on filter paper. The solvent was allowed to dissolve, and then filter paper was then wetted again with ultrapure water.
Domain 4: Test Organism				

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Study Citation:	Gu, J., Chen, X., Wang, Y., Wang, L., Szlavecz, K., Ma, Y., Ji, R. (2020). Bioaccumulation, physiological distribution, and biotransformation of tetrabromobisphenol a (TBBPA) in the geophagous earthworm Metaphire guillelmi - hint for detoxification strategy. Journal of Hazardous Materials 388:122027.
Duration:	Overall Duration: 0 - 4 days (0-96h); Exposure Duration: 0 - 4 days (0-96h)
Exposure Route, Media, Path:	Terrestrial; N/A (e.g., injection); Dermal (topical application)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Eisenia fetida*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6775143

Domain	Metric		Rating	Comments
	Metric 13:	Test Organism Characteristics	Low	The source of the E. fetida was not reported, nor was the age.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the E. fetida were acclimated to test conditions at any point prior to the test.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	There was one earthworm per test vial, and there were 10 replicates per tests concentration.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	The culture/holding conditions were not reported. It was reported the organisms were held at 20C for the duration of the study, but nothing else was reported.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome(s) of interest-activities of antioxidant enzymes including SOD, CAT, GST, GSH, MDA, and AChE. These were reported to be analyzed using diagnostic reagent kits.
	Metric 18:	Consistency of Outcome Assessment	High	The outcome assessment was reported and appeared to be conducted consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups in animal attrition or health outcomes unrelated to exposure.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Uninformative	Statistical analysis for this portion of the study was not reported.
	Metric 22:	Reporting of Data	Low	LOAEL values were reported in supplemental Table 2. Results for each test concentration were not reported.
	Metric 23:	Explanation of Unexpected Outcomes	Low	Variability was not reported for this portion of the study.

Additional Comments: This evaluation is for the effect of TBBPA on E. fetida ROS after 24h of exposure. This study was described briefly in the supplemental material only. It received an unacceptable rating due to the lack of information regarding statistical analysis.

Overall Quality Determination Uninformative

Study Citation:	Chen, X., Gu, X., Zhao, X., Ma, X., Pan, Y., Wang, X., Ji, R. (2018). Species-dependent toxicity, accumulation, and subcellular partitioning of cadmium in combination with tetrabromobisphenol A in earthworms. Chemosphere 210:1042-1050.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Eisenia fetida*; Adult
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5115416

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 >97%
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Study authors reported using an appropriate concurrent negative control group but unclear whether it was a solvent control. Solvent was used to spike the soil with the test substance, so ideally a solvent control should be used for the experiment.
	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	Methods for preparation of stock solution and TBBPA spiked soil were not given in detail. The solvent concentration used was not reported. Concentrations of test substance in the spiked soil were not measured before or during the study.
	Metric 8:	Consistency of Exposure Administration	Medium	Details of exposure administration were reported. Since exposure concentration in the spiked soil was not measured, unclear whether the TBBPA concentration was homogenously distributed in the soil.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure (14 days) and exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response. Three exposure groups 10, 100,and 500 mg/kg) were used.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms. "The earthworms were acclimated in non-spiked soil at 20C for at least 2 weeks."
	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

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Study Citation:	Chen, X., Gu, X., Zhao, X., Ma, X., Pan, Y., Wang, X., Ji, R. (2018). Species-dependent toxicity, accumulation, and subcellular partitioning of cadmium in combination with tetrabromobisphenol A in earthworms. Chemosphere 210:1042-1050.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Eisenia fetida*; Adult
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5115416

Domain	Metric		Rating	Comments
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health. "The beakers were covered with medical gauze and incubated for 14 days at 20 C, 75% humidity and a 16-h light (400-800 Lux) and 8-h dark cycle."
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology (bioamass determination) 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 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	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 evaluation form is for the biomass measurements taken after 14 days exposure to TBBP spiked soil

Overall Quality Determination Medium

Study Citation:	Chen, X., Gu, X., Zhao, X., Ma, X., Pan, Y., Wang, X., Ji, R. (2018). Species-dependent toxicity, accumulation, and subcellular partitioning of cadmium in combination with tetrabromobisphenol A in earthworms. Chemosphere 210:1042-1050.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Eisenia fetida*; Adult
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5115416

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 >97%
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Study authors reported using an appropriate concurrent negative control group but unclear whether it was a solvent control. Solvent was used to spike the soil with the test substance, so ideally a solvent control should be used for the experiment.
	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	Methods for preparation of stock solution and TBBPA spiked soil were not given in detail. The solvent concentration used was not reported. Concentrations of test substance in the spiked soil were not measured before or during the study.
	Metric 8:	Consistency of Exposure Administration	Medium	Details of exposure administration were reported. Since exposure concentration in the spiked soil was not measured, unclear whether the TBBPA concentration was homogenously distributed in the soil.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure (14 days) and exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response. Three exposure groups were tested (10, 100, and 500 mg/kg of TBBPA).
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms. "The earthworms were acclimated in non-spiked soil at 20oC for at least 2 weeks".
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				

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Study Citation:	Chen, X., Gu, X., Zhao, X., Ma, X., Pan, Y., Wang, X., Ji, R. (2018). Species-dependent toxicity, accumulation, and subcellular partitioning of cadmium in combination with tetrabromobisphenol A in earthworms. Chemosphere 210:1042-1050.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Eisenia fetida*; Adult
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5115416

Domain	Metric		Rating	Comments
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health. "The beakers were covered with medical gauze and incubated for 14 days at 20oC, 75% humidity and a 16-h light (400 - 800 Lux) and 8-h dark cycle."
	Metric 17:	Outcome Assessment Methodology	Medium	The outcome assessment methodologies for mechanistic outcomes (ROS, SOD, GPx, GST, GSH and lipid peroxidation) was briefly described and appropriate references were cited for methods.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described in Section 2.6.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group (Fig 3).
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes, or unexpected outcomes were satisfactorily explained.

Additional Comments: Methods for preparing spiked soil were not given in detail. Unclear whether the negative control was a solvent control or not. Exposure concentrations were not verified.

Overall Quality Determination Medium

Study Citation:	Shi, Y. J., Xu, X. B., Zheng, X. Q., Lu, Y. L. (2015). Responses of growth inhibition and antioxidant gene expression in earthworms (Eisenia fetida) exposed to tetrabromobisphenol A, hexabromocyclododecane and decabromodiphenyl ether. Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 174-175:32-38.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Eisenia fetida*; Adult
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Oxidative stress (including redox biology)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2965902

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name, CAS# and structure
	Metric 2:	Test Substance Source	Low	The test substance source was identified, but identity was not analytically verified – "Tetrabromobisphenol A (CAS No. 79-94-7, 98% purity), was produced by TCI (Shanghai) Chemicals"
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using appropriate concurrent negative control groups
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	There are minor reservations about the source of test organisms
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				

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Study Citation:	Shi, Y. J., Xu, X. B., Zheng, X. Q., Lu, Y. L. (2015). Responses of growth inhibition and antioxidant gene expression in earthworms (Eisenia fetida) exposed to tetrabromobisphenol A, hexabromocyclododecane and decabromodiphenyl ether. Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 174-175:32-38.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Eisenia fetida*; Adult
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Oxidative stress (including redox biology)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2965902

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

Additional Comments: gene expression

Overall Quality Determination High

Study Citation:	Shi, Y. J., Xu, X. B., Zheng, X. Q., Lu, Y. L. (2015). Responses of growth inhibition and antioxidant gene expression in earthworms (Eisenia fetida) exposed to tetrabromobisphenol A, hexabromocyclododecane and decabromodiphenyl ether. Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 174-175:32-38.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Eisenia fetida*; Adult
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2965902

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name, CAS# and structure
	Metric 2:	Test Substance Source	High	The test substance source was identified, but identity was not analytically verified – "Tetrabromobisphenol A (CAS No. 79-94-7, 98% purity), was produced by TCI (Shanghai) Chemicals"
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 98%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using appropriate concurrent negative control groups
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Medium	There are minor reservations about the source of test organisms
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	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:	Shi, Y. J., Xu, X. B., Zheng, X. Q., Lu, Y. L. (2015). Responses of growth inhibition and antioxidant gene expression in earthworms (Eisenia fetida) exposed to tetrabromobisphenol A, hexabromocyclododecane and decabromodiphenyl ether. Comparative Biochemistry and Physiology - Part C: Toxicology and Pharmacology 174-175:32-38.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Eisenia fetida*; Adult
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	2965902

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 clearly described
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: None

Overall Quality Determination High

Study Citation:	Ltd,, W.I. (2003). Effect of tetrabromobisphenol A on the survival and reproduction of the earthworm, Eisenia fetida.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Eisenia fetida*; Adult
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	7415561

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance was obtained from Wildlife International Ltd. but the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 98.91%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group (Table 8).
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Medium	The study reported that "earthworms were impartially added to a set of labeled containers. Each container was randomly assigned to treatment replicate by a random number generator." Worms were then added to soil in each test chamber.
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.Soil characterization, preparation and spiking methods were given in detail.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure (28 days) 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 seemed suitable. Nominal soil concentrations of 0.63, 1.3, 2.5, 5.0, 10, 20, and 40 mg TBBPA per kg dry soil were used for the second definitive test.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions and all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and suitable. 10 worms per concentration were used and each concentration had 4 replicates. Control group had eight replicates.

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Study Citation:	Ltd,, W.I. (2003). Effect of tetrabromobisphenol A on the survival and reproduction of the earthworm, Eisenia fetida.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Eisenia fetida*; Adult
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	7415561

Domain	Metric		Rating	Comments
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health. Housing, environmental conditions and feeding details were provided in the report.
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology for determining tissue concentration of TBBPA was reported. Tissue samples were composited per treatment and no variability measurements were reported.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical analysis was not applied to the BAF evaluation
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability (e.g., SE, SD, confidence intervals).

Additional Comments: This form is for the ADME (bioaccumulation of TBBPA) measured in the second definitive test.

Overall Quality Determination High

Study Citation:	Ltd,, W.I. (2003). Effect of tetrabromobisphenol A on the survival and reproduction of the earthworm, Eisenia fetida.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Eisenia fetida*; Adult
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	7415561

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 98.91%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Medium	The study reported that "earthworms were impartially added to a set of labeled containers. Each container was randomly assigned to treatment replicate by a random number generator." Worms were then added to soil in each test chamber.
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. Soil characterization, preparation and spiking methods were given in detail.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported (56 days) 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. Nominal soil concentrations of 0.63, 1.3, 2.5, 5.0, 10, 20, and 40 mg TBBPA per kg dry soil were used for the second definitive test.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions and all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and suitable. 10 worms per concentration were used and each concentration had 4 replicates. Control group had eight replicates.
Domain 5: Outcome Assessment				

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Study Citation:	Ltd,, W.I. (2003). Effect of tetrabromobisphenol A on the survival and reproduction of the earthworm, Eisenia fetida.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Eisenia fetida*; Adult
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	7415561

Domain	Metric		Rating	Comments
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health. Housing, environmental conditions and feeding details were provided in the report.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest. The number of juvenile worms was "determined at test termination by rinsing the test soil through a series of sieves (e.g., 710,500, and 425 μm mesh), collecting, and enumerating the animals retained within the sieves."
	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 in section 2.6.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: None

Overall Quality Determination High

Study Citation:	Ltd,, W.I. (2003). Effect of tetrabromobisphenol A on the survival and reproduction of the earthworm, Eisenia fetida.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Eisenia fetida*; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	7415561

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance was obtained from Wildlife International Ltd. but the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 98.91%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and adequate.
	Metric 6:	Randomized Allocation	Medium	The study reported that "earthworms were impartially added to a set of labeled containers. Each container was randomly assigned to treatment replicate by a random number generator." Worms were then added to soil in each test chamber.
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. Soil characterization, preparation and spiking methods were given in detail.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure (28 days) 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. Nominal soil concentrations of 313, 625, 1,250, 2,500, and 5,000 mg TBBPA per kg dry soil were used based on the initial range finding tests results.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions and all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and suitable. 10 worms per concentration were used and each concentration had 4 replicates. Control group had eight replicates.
Domain 5: Outcome Assessment				

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Study Citation:	Ltd,, W.I. (2003). Effect of tetrabromobisphenol A on the survival and reproduction of the earthworm, Eisenia fetida.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Eisenia fetida*; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	7415561

Domain	Metric		Rating	Comments
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health. Housing, environmental conditions and feeding details were provided in the report.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest. Criteria for identifying dead worms were provided.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described in Section 2.6.
	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: Well-conducted and well-written study. Nominal concentrations were verified. This form is for the mortality outcome for the first definitive test.

Overall Quality Determination High

Study Citation:	Ltd,, W.I. (2003). Effect of tetrabromobisphenol A on the survival and reproduction of the earthworm, Eisenia fetida.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Eisenia fetida*; Adult
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	7415561

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#.
	Metric 2:	Test Substance Source	Low	The test substance was obtained from Wildlife International Ltd. but the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 98.91%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Medium	The study reported that "earthworms were impartially added to a set of labeled containers. Each container was randomly assigned to treatment replicate by a random number generator." Worms were then added to soil in each test chamber.
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. Soil characterization, preparation and spiking methods were given in detail.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type. Progeny count was done after 56 days of exposure.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	The number of exposure groups and spacing of exposure levels seemed suitable but worms did not reproduce at tested levels.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions and all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and suitable.10 worms per concentration were used and each concentration had 4 replicates. Control group had eight replicates.
Domain 5: Outcome Assessment				

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Study Citation:	Ltd,, W.I. (2003). Effect of tetrabromobisphenol A on the survival and reproduction of the earthworm, Eisenia fetida.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Eisenia fetida*; Adult
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	7415561

Domain	Metric		Rating	Comments
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health. Housing, environmental conditions and feeding details were provided in the report.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest. The number of juvenile worms were "determined at testtermination by rinsing the test soil through a series of sieves (e.g.,710,500, and425 μmmesh), collecting, and enumerating the animalsretained within the sieves."
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical methods were described in general in Section 2.6 . However, for reproductive outcome results from initial definitive test, statistical test was not applied. "Due to the lack of reproductive activity in the treatments, a second range-finding and definitive test were performed".
	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	No reproductive dose response was unexpected.

Additional Comments: Well-conducted and well-written study. Nominal concentrations were verified. This form is for the reproductive outcome for the first definitive test."Due to the lack of reproductive activity in the treatments, a second range-finding and definitive test were performed".

Overall Quality Determination High

Study Citation:	Ltd,, W.I. (2003). Effect of tetrabromobisphenol A on the survival and reproduction of the earthworm, Eisenia fetida.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Eisenia fetida*; Adult
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	7415561

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance was obtained from Wildlife International Ltd. but the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 98.91%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.
	Metric 6:	Randomized Allocation	Medium	The study reported that "earthworms were impartially added to a set of labeled containers. Each container was randomly assigned to treatment replicate by a random number generator." Worms were then added to soil in each test chamber.
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. Soil characterization, preparation and spiking methods were given in detail.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported (28 days) and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	The number of exposure groups and spacing of exposure levels seemed suitable but worms did not respond (weight change) in a dose dependent manner.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions and all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and suitable. 10 worms per concentration were used and each concentration had 4 replicates. Control group had eight replicates.
Domain 5: Outcome Assessment				

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Study Citation:	Ltd,, W.I. (2003). Effect of tetrabromobisphenol A on the survival and reproduction of the earthworm, Eisenia fetida.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Eisenia fetida*; Adult
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	7415561

Domain	Metric		Rating	Comments
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health. Housing, environmental conditions and feeding details were provided in the report.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology (for measuring change in body weight) 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	Low	It was not clear if statistical analysis was applied to the growth evaluation as NOEC or LOEC values were not reported.
	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	No clear dose response for weight change was unexpected.

Additional Comments: Well-conducted and well-written study. Nominal concentrations were verified. This form is for the growth outcome for the first definitive test. No clear dose response for worm weight after 28 days of exposure was observed. A second range finding and definitive test was performed.

Overall Quality Determination High

Study Citation:	Ltd,, W.I. (2003). Effect of tetrabromobisphenol A on the survival and reproduction of the earthworm, Eisenia fetida.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Eisenia fetida*; Adult
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	7415561

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#.
	Metric 2:	Test Substance Source	Low	The test substance was obtained from Wildlife International Ltd. but the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 98.91%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Medium	The study reported that "earthworms were impartially added to a set of labeled containers. Each container was randomly assigned to treatment replicate by a random number generator." Worms were then added to soil in each test chamber.
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. Soil characterization, preparation and spiking methods were given in detail.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure (28 days) 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 seemed suitable.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions and all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and suitable.10 worms per concentration were used and each concentration had 4 replicates. Control group had eight replicates.
Domain 5: Outcome Assessment				

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Study Citation:	Ltd,, W.I. (2003). Effect of tetrabromobisphenol A on the survival and reproduction of the earthworm, Eisenia fetida.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Eisenia fetida*; Adult
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	7415561

Domain	Metric		Rating	Comments
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health. Housing, environmental conditions and feeding details were provided in the report.
	Metric 17:	Outcome Assessment Methodology	Medium	The outcome assessment methodology was reported. Burrowing behavior was observed after 10 minutes and 6 hours post addition of worms to each test chamber. Absence of burrowing was noted after 28 days of exposure.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	N/A	Statistical methods were described in general in Section 2.6. Negative findings across all groups for behavioral outcome and was not quantified.
	Metric 22:	Reporting of Data	High	Negative findings were described qualitatively in the text.
	Metric 23:	Explanation of Unexpected Outcomes	Medium	No dose dependent behavior response was observed.

Additional Comments: Well-conducted and well-written study. Nominal concentrations were verified. This form is for the behavioral outcome for the first definitive test. "No abnormal burrowing or avoidance behaviors were recorded during the test."

Overall Quality Determination High

Study Citation:	Ltd,, W.I. (2003). Effect of tetrabromobisphenol A on the survival and reproduction of the earthworm, Eisenia fetida.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Eisenia fetida*; Adult
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	7415561

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance was obtained from Wildlife International Ltd. but the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 98.91%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Medium	The study reported that "earthworms were impartially added to a set of labeled containers. Each container was randomly assigned to treatment replicate by a random number generator." Worms were then added to soil in each test chamber.
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. Soil characterization, preparation and spiking methods were given in detail.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure (28 days) was reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	The number of exposure groups and spacing of exposure levels seemed suitable but worms did not respond in a dose dependent manner. Nominal soil concentrations of 0.63, 1.3, 2.5, 5.0, 10, 20, and 40 mg TBBPA per kg dry soil were used for the second definitive test.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions and all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and suitable. 10 worms per concentration were used and each concentration had 4 replicates. Control group had eight replicates.

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Study Citation:	Ltd,, W.I. (2003). Effect of tetrabromobisphenol A on the survival and reproduction of the earthworm, Eisenia fetida.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Eisenia fetida*; Adult
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	7415561

Domain	Metric		Rating	Comments
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health. Housing, environmental conditions and feeding details were provided in the report.
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology was reported but there are limitations. Worms were not individually weighed for each replicate per treatment. Only surviving organisms were weighed but methods states that weight change was normalized to 10 organism per replicate. Also, worms were rinsed, not mentioned whether these were blotted dry before weighing, which could have contributed to the variability.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	It was not clear if statistical analysis was applied to the growth evaluation as NOEC or LOEC values were not reported.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	Low	The study did not report any measures of variability (e.g., SE, SD, confidence intervals). The increase in growth rates in higher concentrations was unexpected and not explained in the results.

Additional Comments: None

Overall Quality Determination High

Study Citation:	Ltd,, W.I. (2003). Effect of tetrabromobisphenol A on the survival and reproduction of the earthworm, Eisenia fetida.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Eisenia fetida*; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	7415561

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance was obtained from Wildlife International Ltd. but the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 98.91%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Medium	The study reported that "earthworms were impartially added to a set of labeled containers. Each container was randomly assigned to treatment replicate by a random number generator." Worms were then added to soil in each test chamber.
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. Soil characterization, preparation and spiking methods were given in detail.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure (28 days) was reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	7 exposure groups were used. Nominal soil concentrations of 0.63, 1.3, 2.5, 5.0, 10, 20, and 40 mg TBBPA per kg dry soil were used for the second definitive test.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions and all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and suitable. 10 worms per concentration were used and each concentration had 4 replicates. Control group had eight replicates.
Domain 5: Outcome Assessment				

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Study Citation:	Ltd,, W.I. (2003). Effect of tetrabromobisphenol A on the survival and reproduction of the earthworm, Eisenia fetida.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Eisenia fetida*; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	7415561

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. Criteria for identifying dead worms were provided.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	It was not clear if statistical analysis was applied to the survival evaluation as NOEC or LOEC values were not reported for mortality results from second definitive test.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	Medium	No mortality in the second definitive test was unexpected.

Additional Comments: Well-conducted and well-written study. Nominal concentrations were verified. This form is for the mortality outcome for the second definitive test. NOEC or EC 50 values were not reported for mortality data.

Overall Quality Determination High

Study Citation:	Warbritton, R., Kendall, T. Z., Nixon, W. B. (2005). Effect of tetrabromobisphenol A on the reproduction of the earthworm, Eisenia fetida.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Eisenia fetida*; Adult
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6311100

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.2%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes (Table 7, page 25/77).
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were impartially allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods, HPLC.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type, 56 days for reproduction.
	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. There were 7 treatment concentrations.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions and all pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects (OECD 850.6200).
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:	Warbritton, R., Kendall, T. Z., Nixon, W. B. (2005). Effect of tetrabromobisphenol A on the reproduction of the earthworm, Eisenia fetida.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Eisenia fetida*; Adult
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6311100

Domain	Metric		Rating	Comments
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest (page 12/77).
	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 were no information presented 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: Reproduction of adult worms was described in the results section on page 14/77 and also presented in table 7 (page 25/77).

Overall Quality Determination High

Study Citation:	Warbritton, R., Kendall, T. Z., Nixon, W. B. (2005). Effect of tetrabromobisphenol A on the reproduction of the earthworm, Eisenia fetida.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Eisenia fetida*; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6311100

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.2%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group (page 17/77).
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes Page 21/77).
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were impartially allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods (HPLC).
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type (28 days and 56 days).
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	The number of exposure groups and spacing of exposure levels were reasonable but did not produce a dose response (7 concentrations and a control).
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions and all pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	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 12/77).

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Study Citation:	Warbritton, R., Kendall, T. Z., Nixon, W. B. (2005). Effect of tetrabromobisphenol A on the reproduction of the earthworm, Eisenia fetida.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Eisenia fetida*; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6311100

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups (Page 12/77).
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 information presented 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: Mortality assessments conducted on day 28 for adult worms are reported page 14/77.

Overall Quality Determination High

Study Citation:	Warbritton, R., Kendall, T. Z., Nixon, W. B. (2005). Effect of tetrabromobisphenol A on the reproduction of the earthworm, Eisenia fetida.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Eisenia fetida*; Adult
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6311100

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.2%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group (page 17/77).
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes Page 21/77).
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were impartially allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods (HPLC).
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type (behavior assessed at 28 days).
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	The number of exposure groups and spacing of exposure levels were reasonable but did not produce a dose response (7 concentrations and a control).
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions and all pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	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 12/77).

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Study Citation:	Warbritton, R., Kendall, T. Z., Nixon, W. B. (2005). Effect of tetrabromobisphenol A on the reproduction of the earthworm, Eisenia fetida.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Eisenia fetida*; Adult
Health Outcome:	Behavioral
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6311100

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups (Page 12/77).
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 information presented 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 (Table 5).
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: Behavioral assessments conducted on day 28 for adult worms are reported on page 14/77.

Overall Quality Determination High

Study Citation:	Warbritton, R., Kendall, T. Z., Nixon, W. B. (2005). Effect of tetrabromobisphenol A on the reproduction of the earthworm, Eisenia fetida.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Eisenia fetida*; Adult
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6311100

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Chemical was identified by name and CAS#
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.2%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes (page 14).
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were impartially allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods (page 11-12/77).
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type (28 and 56 day endpoints).
	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 (7 treatments concentrations).
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The test organisms were acclimatized to test conditions and all pretreatment conditions were the same for control and exposed organisms.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects (OPPTS 850.6200).
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 (Page 12/77).

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Study Citation:	Warbritton, R., Kendall, T. Z., Nixon, W. B. (2005). Effect of tetrabromobisphenol A on the reproduction of the earthworm, Eisenia fetida.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Eisenia fetida*; Adult
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6311100

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 were no information presented 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 reported.

Additional Comments: Growth of adult worms was reported at 28-days of exposure on page 14/77 and in Table 6 on page 23/77.

Overall Quality Determination High

Study Citation:	Sverdrup, L. E., Hartnik, T., Mariussen, E., Jensen, J. (2006). Toxicity of three halogenated flame retardants to nitrifying bacteria, red clover (Trifolium pratense), and a soil invertebrate (Enchytraeus crypticus). Chemosphere 64(1):96-103.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Enchytraeus crypticus*; Adult
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	938766

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Identified as Tetrabromobisphenol A (TBBPA; Cas no 79-94-7).
	Metric 2:	Test Substance Source	High	Purchased FLUKA (Germany) and analytically verified.
	Metric 3:	Test Substance Purity	Low	Purity and grade not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Solvent and negative controls reported.
	Metric 5:	Negative Control Response	High	Response of controls were acceptable.
	Metric 6:	Randomized Allocation	Low	Allocation method not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	Experimental system and test media well described. concentrations were measured in sediment, however, the study reported effects based on nominal concentrations even though table 1 (page 4/8) reports deviations between nominal and actual concentrations. Differences would potentially reduce the reported toxicity of the compound in the results.
	Metric 8:	Consistency of Exposure Administration	High	Exposure administration was consistent across groups.
	Metric 9:	Measurement of Test Substance Concentration	High	TBBPA dissolved in acetone before application to soil. Results were reported with nominal concentrations. Analytical concentrations were separately reported and compared to nominal concentrations.
	Metric 10:	Exposure Duration and Frequency	High	Exposure duration and frequency were reported and acceptable.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Number of exposure groups and spacing were appropriate. A range finding test was conducted.
	Metric 12:	Testing at or Below Solubility Limit	N/A	concentrations and exposures in sediment.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Species, life stage, and source were adequately described.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	acclimation was not reported. Pretreatment conditions were equivalent across groups.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Number of organisms and replicates were appropriate for test.
Domain 5: Outcome Assessment				

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Study Citation:	Sverdrup, L. E., Hartnik, T., Mariussen, E., Jensen, J. (2006). Toxicity of three halogenated flame retardants to nitrifying bacteria, red clover (Trifolium pratense), and a soil invertebrate (Enchytraeus crypticus). Chemosphere 64(1):96-103.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Enchytraeus crypticus*; Adult
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	938766

Domain	Metric		Rating	Comments
	Metric 16:	Adequacy of Test Conditions	High	Housing and environmental conditions were appropriate for test.
	Metric 17:	Outcome Assessment Methodology	High	Intended outcomes were reported.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences between groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	Outcomes unrelated to exposures were not reported.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical analyses were well described.
	Metric 22:	Reporting of Data	High	Data for all exposures were reported.
	Metric 23:	Explanation of Unexpected Outcomes	High	No unexpected outcomes were reported.

Additional Comments: ” TBBPA showed a high toxicity to enchytraeid reproduction, with small, but significant effects already atthe 10 mg kg1 exposure level (ANOVA, Dunnett’s $p < 0.05$). Figure 3, page 7/8.”

Overall Quality Determination High

Study Citation:	Chen, X., Gu, X., Zhao, X., Ma, X., Pan, Y., Wang, X., Ji, R. (2018). Species-dependent toxicity, accumulation, and subcellular partitioning of cadmium in combination with tetrabromobisphenol A in earthworms. Chemosphere 210:1042-1050.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Metaphire guillelmi*; Adult
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5115416

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 >97%
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Study authors reported using an appropriate concurrent negative control group but unclear whether it was a solvent control. Solvent was used to spike the soil with the test substance, so ideally a solvent control should be used for the experiment.
	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	Methods for preparation of stock solution and TBBPA spiked soil were not given in detail. The solvent concentration used was not reported. Concentrations of test substance in the spiked soil were not measured before or during the study.
	Metric 8:	Consistency of Exposure Administration	Medium	Details of exposure administration were reported. Since exposure concentration in the spiked soil was not measured, unclear whether the TBBPA concentration was homogenously distributed in the soil.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure (14 days) and exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms. "The earthworms were acclimated in non-spiked soil at 20oC for at least 2 weeks".
	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 earthworms per beaker and 3 replicate beakers per concentration were used.

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Study Citation:	Chen, X., Gu, X., Zhao, X., Ma, X., Pan, Y., Wang, X., Ji, R. (2018). Species-dependent toxicity, accumulation, and subcellular partitioning of cadmium in combination with tetrabromobisphenol A in earthworms. Chemosphere 210:1042-1050.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Metaphire guillelmi*; Adult
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5115416

Domain	Metric		Rating	Comments
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health. "The beakers were covered with medical gauze and incubated for 14 days at 20oC, 75% humidity and a 16-h light (400 - 800 Lux) and 8-h dark cycle."
	Metric 17:	Outcome Assessment Methodology	Medium	The outcome assessment methodologies for mechanistic outcomes (ROS, SOD, GPx, GST, GSH and lipid peroxidation) was briefly described and appropriate references were cited for methods.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
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: Methods for preparing spiked soil were not given in detail. Unclear whether the negative control was a solvent control or not. Exposure concentrations were not verified.

Overall Quality Determination Medium

Study Citation:	Chen, X., Gu, X., Zhao, X., Ma, X., Pan, Y., Wang, X., Ji, R. (2018). Species-dependent toxicity, accumulation, and subcellular partitioning of cadmium in combination with tetrabromobisphenol A in earthworms. Chemosphere 210:1042-1050.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Metaphire guillelmi*; Adult
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5115416

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 >97%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Study authors reported using an appropriate concurrent negative control group but unclear whether it was a solvent control. Solvent was used to spike the soil with the test substance, so ideally a solvent control should be used for the experiment.
	Metric 5:	Negative Control Response	Medium	The biological response of the negative control group was reported but there are limitations. Biomass in control decreased after 14 days of exposure. Control response for the other earthworm species tested showed increase in biomass.
	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	Methods for preparation of stock solution and TBBPA spiked soil were not given in detail. The solvent concentration used was not reported. Concentrations of test substance in the spiked soil were not measured before or during the study.
	Metric 8:	Consistency of Exposure Administration	Medium	Details of exposure administration were reported. Since exposure concentration in the spiked soil was not measured, unclear whether the TBBPA concentration was homogenously distributed in the soil.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure (14 days) and exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	The number of exposure groups were reported (10, 50, and100 mg/kg)but since there was 100% mortality in the highest treatment group, biomass could be measured only in 2 groups.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms. "The earthworms wereacclimated in non-spiked soil at 20C for at least 2 weeks."
	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

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Study Citation:	Chen, X., Gu, X., Zhao, X., Ma, X., Pan, Y., Wang, X., Ji, R. (2018). Species-dependent toxicity, accumulation, and subcellular partitioning of cadmium in combination with tetrabromobisphenol A in earthworms. Chemosphere 210:1042-1050.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Metaphire guillelmi*; Adult
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5115416

Domain	Metric		Rating	Comments
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health. "The beakers were covered with medical gauze and incubated for 14 days at20 C, 75% humidity and a 16-h light (400-800 Lux) and 8-h dark cycle."
	Metric 17:	Outcome Assessment Methodology	Low	The outcome assessment methodology (bioamass determination) 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 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	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: Biomass of earthworms in control groups decreased after 14 days.

Overall Quality Determination Medium

Study Citation:	Chen, X., Gu, X., Zhao, X., Ma, X., Pan, Y., Wang, X., Ji, R. (2018). Species-dependent toxicity, accumulation, and subcellular partitioning of cadmium in combination with tetrabromobisphenol A in earthworms. Chemosphere 210:1042-1050.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Metaphire guillelmi*; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5115416

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only.
	Metric 2:	Test Substance Source	Low	Source was reported as Sigma Aldrich but the test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	High	Chemical purity reported as >97%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	Low	Study authors reported using an appropriate concurrent negative control group but unclear whether it was a solvent control. Solvent was used to spike the soil with the test substance, so ideally a solvent control should be used for the experiment.
	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	Methods for preparation of spiked soil were not given in detail. The solvent concentration used was not reported. Concentrations of test substance in the spiked soil were not measure before or during the study.
	Metric 8:	Consistency of Exposure Administration	Medium	Details of exposure administration were reported. Since exposure concentration in the spiked soil were mot measured, unclear whether the TBBPA concentration was achieved and was uniform.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured.
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure (14 days) and exposure frequency were reported and appropriate for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were adequate to address the purpose of the study.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	All pretreatment conditions were the same for control and exposed organisms. "The earthworms were acclimated in non-spiked soil at 20 oC for at least 2 weeks".
	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 earthworms per beaker and 3 replicate beakers per concentration were used.

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Study Citation:	Chen, X., Gu, X., Zhao, X., Ma, X., Pan, Y., Wang, X., Ji, R. (2018). Species-dependent toxicity, accumulation, and subcellular partitioning of cadmium in combination with tetrabromobisphenol A in earthworms. Chemosphere 210:1042-1050.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Metaphire guillelmi*; Adult
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5115416

Domain	Metric		Rating	Comments
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health.
	Metric 17:	Outcome Assessment Methodology	Medium	The outcome assessment methodology was reported but criteria used to assess dead worms were not reported.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions or other non-treatment-related factors across study groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Statistical methods were adequately described but was not incorporated for the discussion of results on mortality.
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group (Figure 1).
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes.

Additional Comments: Methods for preparing spiked soil were not given in detail. Unclear whether the negative control was a solvent control or not. Exposure concentrations were not verified.

Overall Quality Determination Medium

Study Citation:	Gu, J., Chen, X., Wang, Y., Wang, L., Szlavecz, K., Ma, Y., Ji, R. (2020). Bioaccumulation, physiological distribution, and biotransformation of tetrabromobisphenol a (TBBPA) in the geophagous earthworm Metaphire guillelmi - hint for detoxification strategy. Journal of Hazardous Materials 388:122027.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Dermal (topical application), Dietary
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Metaphire guillelmi*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6775143

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The TBBPA was identified by name only.
	Metric 2:	Test Substance Source	Low	The source of the non-radiolabeled TBBPA was reported to be Sigma Corp. It was not reported to be analytically verified. The 14C ring-labeled TBBPA was synthesized from uniformly 14C-ring -labeled phenol.
	Metric 3:	Test Substance Purity	High	The radiochemical purity was reported to be 99%, and the chemical purity was reported to be 97%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of a concurrent negative control in which no TBBPA was present.
	Metric 5:	Negative Control Response	High	The biological response of the negative control was reported in Figure S1 in the supplemental material.
	Metric 6:	Randomized Allocation	Low	It was not reported how the M. guillelmi were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	0.83mL of 14C methanolic solution with a radioactivity concentration of 0.35MBq/mL and chemical concentration of 5ug/mL was added dropwise to 330g of air dried soil through mixing. The final concentration of TBBPA and radioactivity were reported to be 9.96ug/g and 0.65kBq/g respectively. This was allowed to sit for 24h and then 30g of soil were placed in 100mL flasks with one earthworm per flask. The flasks were then stoppered.
	Metric 8:	Consistency of Exposure Administration	High	Details of the exposure administration were reported, and the exposures appeared to be administered consistently. 100mL flasks were used with 30g of treated soil. The flasks were kept under the same environmental conditions.
	Metric 9:	Measurement of Test Substance Concentration	Medium	The radioactivity and the chemical concentration were determined at the start of the test. These were not monitored in the soil for the rest of the study duration, though presumably the earthworms would be taking up the chemical.
	Metric 10:	Exposure Duration and Frequency	High	The earthworms were exposed to the test substance for 30 days. There was a depuration portion to the study that lasted up to 18 days. This was adequate to observe the outcomes of interest.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	There was one exposure level, as the goal of the study was not to observe a dose response, but to observe accumulation, metabolism, and depuration of TBBPA in earthworms.

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Study Citation:	Gu, J., Chen, X., Wang, Y., Wang, L., Szlavecz, K., Ma, Y., Ji, R. (2020). Bioaccumulation, physiological distribution, and biotransformation of tetrabromobisphenol a (TBBPA) in the geophagous earthworm Metaphire guillelmi - hint for detoxification strategy. Journal of Hazardous Materials 388:122027.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Dermal (topical application), Dietary
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Metaphire guillelmi*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6775143

Domain	Metric		Rating	Comments
	Metric 12:	Testing at or Below Solubility Limit	N/A	This study was conducted in soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The M. guillelmi were field collected from a farm in Jurong, Jiangsu Province, China. This creates concern regarding the health of the organisms.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The earthworms were kept in uncontaminated soil at the test temperature for 15 days prior to the start of the test.
	Metric 15:	Number of Organisms and Replicates per Group	Low	There was one earthworm per flask, but the number of replicates for the study was unclear for the TBBPA. There were three flasks without the addition of TBBPA that served as controls.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	The soil characteristics were described adequately. The earthworms were kept in 100mL flasks that were stoppered except for once daily for 10 minutes to refresh the headspace. There was one worm per flask. They were held at 20C in the dark, and the soil was maintained at 60% water holding capacity.
	Metric 17:	Outcome Assessment Methodology	Low	This study focused mainly on ADME outcomes, but biomass of the earthworms was also monitored for the duration of the exposure. Since earthworm growth was not the main focus of the study, no information was provided on the outcome assessment methodology for this portion of the study.
	Metric 18:	Consistency of Outcome Assessment	Low	Details of the outcome assessment were limited for this portion of the study.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	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 for the biomass portion of the study was conducted but not described.
	Metric 22:	Reporting of Data	High	Control and exposure results are reported in the supplemental material in Fig S1.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.

Additional Comments: This evaluation is for the biomass outcome in M. guillelmi after exposure to TBBPA. This was not the main focus of the study, so assessment methods were not reported in great detail. Biomass outcomes are reported in the supplemental material in Figure S1.

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Study Citation:	Gu, J., Chen, X., Wang, Y., Wang, L., Szlavecz, K., Ma, Y., Ji, R. (2020). Bioaccumulation, physiological distribution, and biotransformation of tetrabromobisphenol a (TBBPA) in the geophagous earthworm Metaphire guillelmi - hint for detoxification strategy. Journal of Hazardous Materials 388:122027.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Dermal (topical application), Dietary
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Metaphire guillelmi*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6775143

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

Study Citation:	Gu, J., Chen, X., Wang, Y., Wang, L., Szlavecz, K., Ma, Y., Ji, R. (2020). Bioaccumulation, physiological distribution, and biotransformation of tetrabromobisphenol a (TBBPA) in the geophagous earthworm Metaphire guillelmi - hint for detoxification strategy. Journal of Hazardous Materials 388:122027.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Dermal (topical application), Dietary
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Metaphire guillelmi*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6775143

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The TBBPA was identified by name only.
	Metric 2:	Test Substance Source	Low	The source of the non-radiolabeled TBBPA was reported to be Sigma Corp. It was not reported to be analytically verified. The 14C ring-labeled TBBPA was synthesized from uniformly 14C-ring -labeled phenol.
	Metric 3:	Test Substance Purity	High	The radiochemical purity was reported to be 99%, and the chemical purity was reported to be 97%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of a concurrent negative control in which no TBBPA was present.
	Metric 5:	Negative Control Response	High	It was reported that all earthworm individuals survived during the experiments.
	Metric 6:	Randomized Allocation	Low	It was not reported how the M. guillelmi were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	0.83mL of 14C methanolic solution with a radioactivity concentration of 0.35MBq/mL and chemical concentration of 5ug/mL was added dropwise to 330g of air dried soil through mixing. The final concentration of TBBPA and radioactivity were reported to be 9.96ug/g and 0.65kBq/g respectively. This was allowed to sit for 24h and then 30g of soil were placed in 100mL flasks with one earthworm per flask. The flasks were then stoppered.
	Metric 8:	Consistency of Exposure Administration	High	Details of the exposure administration were reported, and the exposures appeared to be administered consistently. 100mL flasks were used with 30g of treated soil. The flasks were kept under the same environmental conditions.
	Metric 9:	Measurement of Test Substance Concentration	Medium	The radioactivity and the chemical concentration were determined at the start of the test. These were not monitored in the soil for the rest of the study duration, though presumably the earthworms would be taking up the chemical.
	Metric 10:	Exposure Duration and Frequency	High	The earthworms were exposed to the test substance for 30 days. There was a depuration portion to the study that lasted up to 18 days. This was adequate to observe the outcomes of interest.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	There was one exposure level, as the goal of the study was not to observe a dose response, but to observe accumulation, metabolism, and depuration of TBBPA in earthworms.
	Metric 12:	Testing at or Below Solubility Limit	N/A	This study was conducted in soil.
Domain 4: Test Organism				

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Study Citation:	Gu, J., Chen, X., Wang, Y., Wang, L., Szlavecz, K., Ma, Y., Ji, R. (2020). Bioaccumulation, physiological distribution, and biotransformation of tetrabromobisphenol a (TBBPA) in the geophagous earthworm Metaphire guillelmi - hint for detoxification strategy. Journal of Hazardous Materials 388:122027.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Dermal (topical application), Dietary
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Metaphire guillelmi*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6775143

Domain	Metric		Rating	Comments
	Metric 13:	Test Organism Characteristics	Low	The M. guillelmi were field collected from a farm in Jurong, Jiangsu Province, China. This creates concern regarding the health of the organisms.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The earthworms were kept in uncontaminated soil at the test temperature for 15 days prior to the start of the test.
	Metric 15:	Number of Organisms and Replicates per Group	Low	There was one earthworm per flask, but the number of replicates for the study was unclear for the TBBPA. There were three flasks without the addition of TBBPA that served as controls.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	The soil characteristics were described adequately. The earthworms were kept in 100mL flasks that were stoppered except for once daily for 10 minutes to refresh the headspace. There was one worm per flask. They were held at 20C in the dark, and the soil was maintained at 60% water holding capacity.
	Metric 17:	Outcome Assessment Methodology	Low	This study focused mainly on ADME outcomes, but mortality was also monitored. It is unclear what methods were used for determining if an organism was dead.
	Metric 18:	Consistency of Outcome Assessment	Low	Details of the outcome assessment were limited for this portion of the study.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences among the study groups in environmental conditions or other factors that could influence the outcome assessment.
	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	N/A	Study authors reported all M. guillelmi earthworms survived during the experiment, so statistical analysis was not necessary.
	Metric 22:	Reporting of Data	Low	Survival was reported in the the text of the Results section. All earthworm individuals survived during the experiments.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.

Additional Comments: This evaluation is for the mortality outcome in M. guillelmi after exposure to TBBPA. This was not the main focus of the study, so assessment methods were not reported in great detail. Mortality was reported in the text of the Results section.

Overall Quality Determination Medium

Study Citation:	Gu, J., Chen, X., Wang, Y., Wang, L., Szlavecz, K., Ma, Y., Ji, R. (2020). Bioaccumulation, physiological distribution, and biotransformation of tetrabromobisphenol a (TBBPA) in the geophagous earthworm Metaphire guillelmi - hint for detoxification strategy. Journal of Hazardous Materials 388:122027.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Dermal (topical application), Dietary
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Metaphire guillelmi*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6775143

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The TBBPA was identified by name only.
	Metric 2:	Test Substance Source	Low	The source of the non-radiolabeled TBBPA was reported to be Sigma Corp. It was not reported to be analytically verified. The 14C ring-labeled TBBPA was synthesized from uniformly 14C-ring -labeled phenol.
	Metric 3:	Test Substance Purity	High	The radiochemical purity was reported to be 99%, and the chemical purity was reported to be 97%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of a concurrent negative control in which no TBBPA was present.
	Metric 5:	Negative Control Response	Low	The biological response of the negative control was not reported.
	Metric 6:	Randomized Allocation	Low	It was not reported how the M. guillelmi were allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	0.83mL of 14C methanolic solution with a radioactivity concentration of 0.35MBq/mL and chemical concentration of 5ug/mL was added dropwise to 330g of air dried soil through mixing. The final concentration of TBBPA and radioactivity were reported to be 9.96ug/g and 0.65kBq/g respectively. This was allowed to sit for 24h and then 30g of soil were placed in 100mL flasks with one earthworm per flask. The flasks were then stoppered.
	Metric 8:	Consistency of Exposure Administration	High	Details of the exposure administration were reported, and the exposures appeared to be administered consistently. 100mL flasks were used with 30g of treated soil. The flasks were kept under the same environmental conditions.
	Metric 9:	Measurement of Test Substance Concentration	Medium	The radioactivity and the chemical concentration were determined at the start of the test. These were not monitored in the soil for the rest of the study duration, though presumably the earthworms would be taking up the chemical.
	Metric 10:	Exposure Duration and Frequency	High	The earthworms were exposed to the test substance for 30 days. There was a depuration portion to the study that lasted up to 18 days. This was adequate to observe the outcomes of interest.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	There was one exposure level, as the goal of the study was not to observe a dose response, but to observe accumulation, metabolism, and depuration of TBBPA in earthworms.
	Metric 12:	Testing at or Below Solubility Limit	N/A	This study was conducted in soil.
Domain 4: Test Organism				

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Study Citation:	Gu, J., Chen, X., Wang, Y., Wang, L., Szlavecz, K., Ma, Y., Ji, R. (2020). Bioaccumulation, physiological distribution, and biotransformation of tetrabromobisphenol a (TBBPA) in the geophagous earthworm Metaphire guillelmi - hint for detoxification strategy. Journal of Hazardous Materials 388:122027.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Dermal (topical application), Dietary
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Metaphire guillelmi*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6775143

Domain	Metric		Rating	Comments
	Metric 13:	Test Organism Characteristics	Low	The M. guillelmi were field collected from a farm in Jurong, Jiangsu Province, China. This creates concern regarding the health of the organisms.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The earthworms were kept in uncontaminated soil at the test temperature for 15 days prior to the start of the test.
	Metric 15:	Number of Organisms and Replicates per Group	Low	There was one earthworm per flask, but the number of replicates for the study was unclear for the TBBPA. There were three flasks without the addition of TBBPA that served as controls.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	The soil characteristics were described adequately. The earthworms were kept in 100mL flasks that were stoppered except for once daily for 10 minutes to refresh the headspace. There was one worm per flask. They were held at 20C in the dark, and the soil was maintained at 60% water holding capacity.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome(s) of interest–accumulation and depuration of TBBPA in earthworm M. guillelmi during exposure to 14C-TBBPA in soil.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently. Earthworms were exposed to the test substance for 30 days with sampling on days 5,10, 15, 20, 25, and 30. The organisms were then transferred to clean soil for depuration for up to 18 days with sampling on days 3, 6, 9, 12, and 18. Organism dissections and chemical analyses were performed consistently across all organisms.
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	The statistical analysis was adequate for the study and can be found in section 2.6 ” Data analysis.”
	Metric 22:	Reporting of Data	Low	Negative control results were not reported, though they typically are not reported for this type of study. It is unknown if there was any contamination. ADME outcomes are reported in Figs. 1-5, as well as in Figure S2.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.

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Study Citation:	Gu, J., Chen, X., Wang, Y., Wang, L., Szlavecz, K., Ma, Y., Ji, R. (2020). Bioaccumulation, physiological distribution, and biotransformation of tetrabromobisphenol a (TBBPA) in the geophagous earthworm Metaphire guillelmi - hint for detoxification strategy. Journal of Hazardous Materials 388:122027.
Duration:	Overall Duration: > 21 days; Exposure Duration: > 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Dermal (topical application), Dietary
Taxa, Species, Age:	Invertebrate; Worms (e.g., Annelids, Nematodes); *Metaphire guillelmi*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6775143

Domain	Metric	Rating	Comments
Additional Comments:	This evaluation is for the ADME outcomes in M. guillelmi after exposure to TBBPA. Accumulation and radioactivity were assessed in earthworms and in earthworm tissues during a 30 day exposure and an 18 day depuration period. Transformation of the TBBPA was also assessed, and metabolites were determined.		

Overall Quality Determination Medium

Study Citation:	Hou, X., Wei, L., Tang, Y., Kong, W., Liu, J., Schnoor, J. L., Jiang, G. (2021). Two typical glycosylated metabolites of tetrabromobisphenol A formed in plants: Excretion and deglycosylation in plant root zones. Environmental Science & Technology Letters 8(4):313-319.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Terrestrial; Water; Root uptake
Taxa, Species, Age:	Vegetation; Non-vascular Plants; *Cucurbita sp.*; Cucurbita maxima x Cucurbita moschata; Juvenile
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10115348

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The TBBPA was identified by name only.
	Metric 2:	Test Substance Source	Low	The TBBPA was reported to be from Dr. Ehrenstorfer GmbH in Augsburg, Germany. It was not reported to be analytically verified.
	Metric 3:	Test Substance Purity	High	The purity was reported to be 99%.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors report the use of a concurrent negative control group in which three plants were in hydroponic solution without TBBPA.
	Metric 5:	Negative Control Response	High	Study authors reported in the text that no TBBPA was detected in pumpkin tissues or hydroponic solutions of the blank control group. This indicated that not volatilization, chemical transformation, or cross-contamination occurred.
	Metric 6:	Randomized Allocation	Low	It was not reported if the plants were randomly allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The preparation of the test substance was only vaguely reported. TBBPA working solution was added to ultrapure water to reach a concentration of 1umol/L. It is unclear how the working solution was prepared. A hydroponic exposure system was used with a volume of 40mL of hydroponic solution. The size of the test chambers were unclear, though there were three seedlings per reactor.
	Metric 8:	Consistency of Exposure Administration	Medium	The exposure appeared to be conducted consistently across study groups. The environmental conditions were reported and consistent, though exposure chamber size was not reported.
	Metric 9:	Measurement of Test Substance Concentration	High	LC-MS/MS was used to determine TBBPA in test solutions and in the plants at various sample times. This was reported in Figure 1.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration was reported to be 240h. This was adequate to observe the outcome of interest.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	There was only one exposure level in this study, as the goal was not to observe a dose response, but to determine uptake of TBBPA into roots, stems, and leaves and to observe effects of glycosylated TBBPA.
	Metric 12:	Testing at or Below Solubility Limit	High	The test concentration was below the water solubility limit.
Domain 4: Test Organism				

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Study Citation:	Hou, X., Wei, L., Tang, Y., Kong, W., Liu, J., Schnoor, J. L., Jiang, G. (2021). Two typical glycosylated metabolites of tetrabromobisphenol A formed in plants: Excretion and deglycosylation in plant root zones. Environmental Science & Technology Letters 8(4):313-319.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Terrestrial; Water; Root uptake
Taxa, Species, Age:	Vegetation; Non-vascular Plants; *Cucurbita sp.*; Cucurbita maxima x Cucurbita moschata; Juvenile
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10115348

Domain	Metric		Rating	Comments
	Metric 13:	Test Organism Characteristics	High	The pumpkin seeds were reported to be from Taigu Yinong Seed Co, LTD. in Shanxi Province, China. Seedlings were used in this study, and the germination process prior to the study was reported.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Study authors reported that test conditions were the same as the growth conditions.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	There were three seedling per test chamber with three replicates for each sampling time.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	The pumpkin seeds were cultivated in an illumination growth chamber with a photoperiod of 16L:8D. Temperatures were reported to be 25C during the day and 22C during the night. Once the reached about 5cm, they were transferred to brown glass reactors with ultrapure water for 2d of adaptive training prior to the start of the exposure. Study authors reported that test conditions were the same as the growth conditions.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome(s) of interest-accumulation of TBBPA in the roots, stems, and leaves of pumpkin seeds during a 240h exposure.
	Metric 18:	Consistency of Outcome Assessment	High	Samples were taken for assessment at 3, 6, 12, 24, 48, 96, 168, and 240h to determine TBBPA in the roots, stems, and leaves of the pumpkin plants as well in the test solution. An LC-MS/MS system was used for chemical analysis.
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	The statistical analysis was reported and adequate for the outcome of interest.
	Metric 22:	Reporting of Data	High	The control results were reported in the text in the "Results and Discussion" section. Accumulation of TBBPA in the root and stem was reported in Fig. 1. Accumulation in the leaves was reported in the text, as there was no accumulation in the leaves.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.

Additional Comments: This evaluation was for the accumulation of TBBPA in pumpkin plants after exposure for 240h. Sampling intervals of 3, 6, 12, 24, 48, 96, 168, and 240h.

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Study Citation:	Hou, X., Wei, L., Tang, Y., Kong, W., Liu, J., Schnoor, J. L., Jiang, G. (2021). Two typical glycosylated metabolites of tetrabromobisphenol A formed in plants: Excretion and deglycosylation in plant root zones. Environmental Science & Technology Letters 8(4):313-319.
Duration:	Overall Duration: 4 - 10 days; Exposure Duration: 4 - 10 days
Exposure Route, Media, Path:	Terrestrial; Water; Root uptake
Taxa, Species, Age:	Vegetation; Non-vascular Plants; *Cucurbita sp.*; Cucurbita maxima x Cucurbita moschata; Juvenile
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10115348

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

Study Citation:	Ltd,, W.I. (2002). Tetrabromobisphenol-A: A toxicity test to determine the effects of the test substance on seedling emergence of six species of plants.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; *Allium cepa*; Texas Grano; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6391028

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 an independent laboratory, (Appendix 3).
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.17%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods (Appendix 4)
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups
	Metric 15:	Number of Organisms and Replicates per Group	Low	Replicates were not used
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health

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Study Citation:	Ltd,, W.I. (2002). Tetrabromobisphenol-A: A toxicity test to determine the effects of the test substance on seedling emergence of six species of plants.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; *Allium cepa*; Texas Grano; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6391028

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 were no information presented 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: Emergence, weight, heightAllium cepa (onion) data on survival, emergence, and growth data are presented in table 4, page 23/122. NOEC/Ecx data estimates are listed on page 27 in Table 8.

Overall Quality Determination High

Study Citation:	Ltd,, W.I. (2002). Tetrabromobisphenol-A: A toxicity test to determine the effects of the test substance on seedling emergence of six species of plants.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; *Allium cepa*; Texas Grano; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6391028

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 an independent laboratory, (Appendix 3).
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.17%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods (Appendix 4)
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups
	Metric 15:	Number of Organisms and Replicates per Group	Low	Replicates were not used
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups

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Study Citation:	Ltd,, W.I. (2002). Tetrabromobisphenol-A: A toxicity test to determine the effects of the test substance on seedling emergence of six species of plants.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; *Allium cepa*; Texas Grano; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6391028

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 were no information presented 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: Allium cepa (onion) data on survival, emergance, and growth data are presented in table 4, page 23/122. NOEC/Ecx data estimates are listed on page 27 in Table 8.

Overall Quality Determination High

Study Citation:	Jiang, Y., Lu, H., Wang, Y., Hong, H., Wang, Q., Liu, J., Yan, C. (2020). Uptake, biotransformation and physiological response of TBBPA in mangrove plants after hydroponics exposure. Marine Pollution Bulletin 151:110832.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Root uptake
Taxa, Species, Age:	Vegetation; Vascular Plants; *Avicennia marina*; Juvenile
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)-Photosynthesis
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10113700

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The TBBPA was identified by name only.
	Metric 2:	Test Substance Source	Low	The source of the TBBPA was not reported.
	Metric 3:	Test Substance Purity	Low	The purity of the TBBPA was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of appropriate concurrent negative controls.
	Metric 5:	Negative Control Response	High	The negative control responses were reported in Figures 3-7 for the mechanistic outcomes, including chlorophyll a and b, MDA, SOD, POD, and CAT.
	Metric 6:	Randomized Allocation	Low	It was not reported how the plants were allocated into study groups. Once the plants were placed into study pots, they were randomly placed in the experimental green house.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	Information regarding the preparation of the test substance was limited. The test substance was reported to be in Hoagland solution with DMSO to aid solubility, but little else was reported. 2.5L of solution was used , and the hydroponic solution was renewed every three days to maintain the test concentration.
	Metric 8:	Consistency of Exposure Administration	High	The exposure administration was administered consistently across study groups. Test groups were maintained in 2.5L of test substance with a 12h light cycle. Environmental conditions were maintained for the duration of the study.
	Metric 9:	Measurement of Test Substance Concentration	Low	It was not reported if the test concentrations were measured at any point during the study.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration was reported to be 14 days. This was adequate to observe the outcomes of interest.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	There were only two exposure groups, but this was adequate to observe the outcomes of interest. Study authors were primarily interested in comparing a high dose and a low dose of TBBPA.
	Metric 12:	Testing at or Below Solubility Limit	Medium	The high dose may be above the water solubility depending on the pH. The low dose appears to be below the water solubility limit. In both doses, DMSO was used to aid the dissolution of the test substance.
Domain 4: Test Organism				

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Study Citation:	Jiang, Y., Lu, H., Wang, Y., Hong, H., Wang, Q., Liu, J., Yan, C. (2020). Uptake, biotransformation and physiological response of TBBPA in mangrove plants after hydroponics exposure. Marine Pollution Bulletin 151:110832.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Root uptake
Taxa, Species, Age:	Vegetation; Vascular Plants; *Avicennia marina*; Juvenile
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)-Photosynthesis
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10113700

Domain	Metric		Rating	Comments
	Metric 13:	Test Organism Characteristics	Medium	The mangroves were reported to be from the Zhangjiang estuary in a typical National Nature Mangrove Reserve in Zhangzhou city, Fujian province, China. They appear to be field harvested, which creates concern regarding plant health, though plants were disinfected prior to the test and only viable propagules were chosen for cultivation for four weeks before the experiment start.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The plants were acclimated to test conditions for four weeks prior to the start of the test.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	There were three seedlings per black pot (10.0cm diameter, 14.0cm height) with three replicates performed for each treatment.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	The plants were placed in the experimental green house with a relative humidity of 60-80%, a temperature of 25C +- 5 degrees, and a 12h light photoperiod with 800-1400 umol photons/m^2/s. Hoagland solution provided nutrients.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome(s) of interest–mechanistic outcomes including chlorophyll a and b, MDA, SOD, POD, and CAT. The outcome assessment methodology was reported clearly.
	Metric 18:	Consistency of Outcome Assessment	High	The outcome assessment was carried out consistently across study groups. Methods were reported clearly.
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	The statistical analysis was reported in section 2.6, and was adequate for the outcomes of interest.
	Metric 22:	Reporting of Data	High	Outcomes for both the controls and the exposure responses were reported in Figures 3-7.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.

Additional Comments: This evaluation was for the effect of TBBPA on mechanistic outcomes in A. marina, including chlorophyll a and b, MDA, SOD, POD, and CAT, after exposure for 14 days. Samples were also taken at day 7.

Overall Quality Determination Medium

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Study Citation:	Jiang, Y., Lu, H., Wang, Y., Hong, H., Wang, Q., Liu, J., Yan, C. (2020). Uptake, biotransformation and physiological response of TBBPA in mangrove plants after hydroponics exposure. Marine Pollution Bulletin 151:110832.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Root uptake
Taxa, Species, Age:	Vegetation; Vascular Plants; *Avicennia marina*; Juvenile
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)-Photosynthesis
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10113700

Domain	Metric	Rating	Comments

Study Citation:	Jiang, Y., Lu, H., Wang, Y., Hong, H., Wang, Q., Liu, J., Yan, C. (2020). Uptake, biotransformation and physiological response of TBBPA in mangrove plants after hydroponics exposure. Marine Pollution Bulletin 151:110832.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Root uptake
Taxa, Species, Age:	Vegetation; Vascular Plants; *Avicennia marina*; Juvenile
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10113700

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The TBBPA was identified by name only.
	Metric 2:	Test Substance Source	Low	The source of the TBBPA was not reported.
	Metric 3:	Test Substance Purity	Low	The purity of the TBBPA was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of appropriate concurrent negative controls.
	Metric 5:	Negative Control Response	High	The negative control response was reported in Table 1, and was adequate for the outcomes of interest.
	Metric 6:	Randomized Allocation	Low	It was not reported how the plants were allocated into study groups. Once the plants were placed into study pots, they were randomly placed in the experimental green house.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	Information regarding the preparation of the test substance was limited. The test substance was reported to be in Hoagland solution with DMSO to aid solubility, but little else was reported. 2.5L of solution was used , and the hydroponic solution was renewed every three days to maintain the test concentration.
	Metric 8:	Consistency of Exposure Administration	High	The exposure administration was administered consistently across study groups. Test groups were maintained in 2.5L of test substance with a 12h light cycle. Environmental conditions were maintained for the duration of the study.
	Metric 9:	Measurement of Test Substance Concentration	Low	It was not reported if the test concentrations were measured at any point during the study.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration was reported to be 14 days. This was adequate to observe the outcomes of interest.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	There were only two exposure groups, but this was adequate to observe the outcomes of interest. Study authors were primarily interested in comparing a high dose and a low dose of TBBPA.
	Metric 12:	Testing at or Below Solubility Limit	Medium	The high dose may be above the water solubility depending on the pH. The low dose appears to be below the water solubility limit. In both doses, DMSO was used to aid the dissolution of the test substance.
Domain 4: Test Organism				

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Study Citation:	Jiang, Y., Lu, H., Wang, Y., Hong, H., Wang, Q., Liu, J., Yan, C. (2020). Uptake, biotransformation and physiological response of TBBPA in mangrove plants after hydroponics exposure. Marine Pollution Bulletin 151:110832.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Root uptake
Taxa, Species, Age:	Vegetation; Vascular Plants; *Avicennia marina*; Juvenile
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10113700

Domain	Metric		Rating	Comments
	Metric 13:	Test Organism Characteristics	Medium	The mangroves were reported to be from the Zhangjiang estuary in a typical National Nature Mangrove Reserve in Zhangzhou city, Fujian province, China. They appear to be field harvested, which creates concern regarding plant health, though plants were disinfected prior to the test and only viable propagules were chosen for cultivation for four weeks before the experiment start.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The plants were acclimated to test conditions for four weeks prior to the start of the test.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	There were three seedlings per black pot (10.0cm diameter, 14.0cm height) with three replicates performed for each treatment.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	The plants were placed in the experimental green house with a relative humidity of 60-80%, a temperature of 25C +- 5 degrees, and a 12h light photoperiod with 800-1400 umol photons/m^2/s. Hoagland solution provided nutrients.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome(s) of interest–mangrove biomass for the root, stem, leaf, and total weight.
	Metric 18:	Consistency of Outcome Assessment	High	The outcome assessment was carried out consistently across study groups. Mangroves were washed and rinsed and then divided into roots, stems, and leaves. The fresh weight was then recorded.
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	The statistical analysis was reported in section 2.6, and was adequate for the outcomes of interest.
	Metric 22:	Reporting of Data	High	The biomass outcomes for all exposure concentrations and the control response were reported in Table 1.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.

Additional Comments: This evaluation was for the changes in biomass in A. marina after exposure to TBBPA.

Overall Quality Determination Medium

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Study Citation:	Jiang, Y., Lu, H., Wang, Y., Hong, H., Wang, Q., Liu, J., Yan, C. (2020). Uptake, biotransformation and physiological response of TBBPA in mangrove plants after hydroponics exposure. Marine Pollution Bulletin 151:110832.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Root uptake
Taxa, Species, Age:	Vegetation; Vascular Plants; *Avicennia marina*; Juvenile
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10113700

Domain	Metric	Rating	Comments

Study Citation:	Jiang, Y., Lu, H., Wang, Y., Hong, H., Wang, Q., Liu, J., Yan, C. (2020). Uptake, biotransformation and physiological response of TBBPA in mangrove plants after hydroponics exposure. Marine Pollution Bulletin 151:110832.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Root uptake
Taxa, Species, Age:	Vegetation; Vascular Plants; *Avicennia marina*; Juvenile
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10113700

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The TBBPA was identified by name only.
	Metric 2:	Test Substance Source	Low	The source of the TBBPA was not reported.
	Metric 3:	Test Substance Purity	Low	The purity of the TBBPA was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of appropriate concurrent negative controls.
	Metric 5:	Negative Control Response	High	It was reported in section 3.1 that TBBPA was not detected in the roots, stems, and leaves of the control plants.
	Metric 6:	Randomized Allocation	Low	It was not reported how the plants were allocated into study groups. Once the plants were placed into study pots, they were randomly placed in the experimental green house.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	Information regarding the preparation of the test substance was limited. The test substance was reported to be in Hoagland solution with DMSO to aid solubility, but little else was reported. 2.5L of solution was used , and the hydroponic solution was renewed every three days to maintain the test concentration.
	Metric 8:	Consistency of Exposure Administration	High	The exposure administration was administered consistently across study groups. Test groups were maintained in 2.5L of test substance with a 12h light cycle. Environmental conditions were maintained for the duration of the study.
	Metric 9:	Measurement of Test Substance Concentration	Low	It was not reported if the test concentrations were measured at any point during the study.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration was reported to be 14 days. This was adequate to observe the outcomes of interest.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	There were only two exposure groups, but this was adequate to observe the outcomes of interest. Study authors were primarily interested in comparing a high dose and a low dose of TBBPA.
	Metric 12:	Testing at or Below Solubility Limit	Medium	The high dose may be above the water solubility depending on the pH. The low dose appears to be below the water solubility limit. In both doses, DMSO was used to aid the dissolution of the test substance.
Domain 4: Test Organism				

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Study Citation:	Jiang, Y., Lu, H., Wang, Y., Hong, H., Wang, Q., Liu, J., Yan, C. (2020). Uptake, biotransformation and physiological response of TBBPA in mangrove plants after hydroponics exposure. Marine Pollution Bulletin 151:110832.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Root uptake
Taxa, Species, Age:	Vegetation; Vascular Plants; *Avicennia marina*; Juvenile
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10113700

Domain	Metric		Rating	Comments
	Metric 13:	Test Organism Characteristics	Medium	The mangroves were reported to be from the Zhangjiang estuary in a typical National Nature Mangrove Reserve in Zhangzhou city, Fujian province, China. They appear to be field harvested, which creates concern regarding plant health, though plants were disinfected prior to the test and only viable propagules were chosen for cultivation for four weeks before the experiment start.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The plants were acclimated to test conditions for four weeks prior to the start of the test.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	There were three seedlings per black pot (10.0cm diameter, 14.0cm height) with three replicates performed for each treatment.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	The plants were placed in the experimental green house with a relative humidity of 60-80%, a temperature of 25C +- 5 degrees, and a 12h light photoperiod with 800-1400 umol photons/m^2/s. Hoagland solution provided nutrients.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome(s) of interest–accumulation of TBBPA in the root, stem, and leaf of the plants.
	Metric 18:	Consistency of Outcome Assessment	High	The outcome assessment was carried out consistently across study groups. Concentrations of TBBPA were determined in the root, stem, and leaf of plants at days 7 and 14. The extraction and purification was performed according to Liu et al (2016).
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	The statistical analysis was reported in section 2.6, and was adequate for the outcomes of interest.
	Metric 22:	Reporting of Data	High	The ADME outcomes were reported in Fig 1 for concentrations of TBBPA in the root, stem, and leaf of the plant at days seven and 14. Control results were reported in the text. BCFs were reported in Table S5. TBBPA and transformation products were reported in Table S6.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.

Additional Comments: This evaluation was for the accumulation of TBBPA in A. marina after a 14 day hydroponic exposure. Samples were taken at days 7 and 14.

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Study Citation:	Jiang, Y., Lu, H., Wang, Y., Hong, H., Wang, Q., Liu, J., Yan, C. (2020). Uptake, biotransformation and physiological response of TBBPA in mangrove plants after hydroponics exposure. Marine Pollution Bulletin 151:110832.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Root uptake
Taxa, Species, Age:	Vegetation; Vascular Plants; *Avicennia marina*; Juvenile
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10113700

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

Study Citation:	Ltd,, W.I. (2002). Tetrabromobisphenol-A: A toxicity test to determine the effects of the test substance on seedling emergence of six species of plants.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; *Cucumis sativa*; Straight Eight; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6391028

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 an independent laboratory, (Appendix 3).
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.17%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods (Appendix 4)
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups
	Metric 15:	Number of Organisms and Replicates per Group	Low	Replicates were not used
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health

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Study Citation:	Ltd,, W.I. (2002). Tetrabromobisphenol-A: A toxicity test to determine the effects of the test substance on seedling emergence of six species of plants.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; *Cucumis sativa*; Straight Eight; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6391028

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 were no information presented 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: Emergence, weight, heightCucumis sativa (cucumber) data on survival, emergence, and growth data are presented in table 3, page 22/122. NOEC/Ecx data estimates are listed on page 27 in Table 8.

Overall Quality Determination High

Study Citation:	Ltd,, W.I. (2002). Tetrabromobisphenol-A: A toxicity test to determine the effects of the test substance on seedling emergence of six species of plants.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; *Cucumis sativa*; Straight Eight; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6391028

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 an independent laboratory, (Appendix 3).
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.17%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods (Appendix 4)
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups
	Metric 15:	Number of Organisms and Replicates per Group	Low	Replicates were not used
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups

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Study Citation:	Ltd,, W.I. (2002). Tetrabromobisphenol-A: A toxicity test to determine the effects of the test substance on seedling emergence of six species of plants.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; *Cucumis sativa*; Straight Eight; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6391028

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 were no information presented 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: Cucumis sativa (cucumber) data on survival, emergence, and growth data are presented in table 3, page 22/122. NOEC/Ecx data estimates are listed on page 27 in Table 8.

Overall Quality Determination High

Study Citation:	Ge, H., Zhang, F. (2017). Effects of Tetrabromobisphenol A Stress on Growth and Physiological Characteristics of Soybean Seedling. Bulletin of Environmental Contamination and Toxicology 98(1):141-146.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake
Taxa, Species, Age:	Vegetation; Vascular Plants; *Glycine max*; Zhoudou22; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Oxidative stress (including redox biology)-Photosynthesis
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4687686

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity 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 reported and reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The experimental system was adequately reported, methods for preparation of test media were not
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Medium	Soil concentrations were measured after exposure
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest

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Study Citation:	Ge, H., Zhang, F. (2017). Effects of Tetrabromobisphenol A Stress on Growth and Physiological Characteristics of Soybean Seedling. Bulletin of Environmental Contamination and Toxicology 98(1):141-146.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake
Taxa, Species, Age:	Vegetation; Vascular Plants; *Glycine max*; Zhoudou22; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Oxidative stress (including redox biology)-Photosynthesis
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4687686

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	Low	Statistical analysis was performed but not described adequately
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: None

Overall Quality Determination High

Study Citation:	Ge, H., Zhang, F. (2017). Effects of Tetrabromobisphenol A Stress on Growth and Physiological Characteristics of Soybean Seedling. Bulletin of Environmental Contamination and Toxicology 98(1):141-146.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake
Taxa, Species, Age:	Vegetation; Vascular Plants; *Glycine max*; Zhoudou22; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4687686

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	Chemical was identified by name only
	Metric 2:	Test Substance Source	Low	The test substance identity was not analytically verified by the performing laboratory.
	Metric 3:	Test Substance Purity	Low	Purity 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 reported and reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Medium	The experimental system was adequately reported, methods for preparation of test media were not
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	Medium	Soil concentrations were measured after exposure
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups

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Study Citation:	Ge, H., Zhang, F. (2017). Effects of Tetrabromobisphenol A Stress on Growth and Physiological Characteristics of Soybean Seedling. Bulletin of Environmental Contamination and Toxicology 98(1):141-146.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake
Taxa, Species, Age:	Vegetation; Vascular Plants; *Glycine max*; Zhoudou22; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	4687686

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

Additional Comments: None

Overall Quality Determination High

Study Citation:	Ltd,, W.I. (2002). Tetrabromobisphenol-A: A toxicity test to determine the effects of the test substance on seedling emergence of six species of plants.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; *Glycine max*; Green Envy; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6391028

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 an independent laboratory, (Appendix 3).
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.17%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods (Appendix 4)
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups
	Metric 15:	Number of Organisms and Replicates per Group	Low	Replicates were not used
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health

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Study Citation:	Ltd,, W.I. (2002). Tetrabromobisphenol-A: A toxicity test to determine the effects of the test substance on seedling emergence of six species of plants.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; *Glycine max*; Green Envy; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6391028

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 were no information presented 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: Glycine max (soybeans) data on survival, emergence, and growth data are presented in table 6, page 25/122. NOEC/Ecx data estimates are listed on page 27 in Table 8.

Overall Quality Determination High

Study Citation:	Ltd,, W.I. (2002). Tetrabromobisphenol-A: A toxicity test to determine the effects of the test substance on seedling emergence of six species of plants.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; *Glycine max*; Green Envy; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6391028

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 an independent laboratory, (Appendix 3).
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.17%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods (Appendix 4)
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups
	Metric 15:	Number of Organisms and Replicates per Group	Low	Replicates were not used
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups

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Study Citation:	Ltd,, W.I. (2002). Tetrabromobisphenol-A: A toxicity test to determine the effects of the test substance on seedling emergence of six species of plants.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; *Glycine max*; Green Envy; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6391028

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 were no information presented 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: Emergence, weight, heightGlycine max (soybeans) data on survival, emergence, and growth data are presented in table 6, page 25/122. NOEC/Ecx data estimates are listed on page 27 in Table 8.

Overall Quality Determination High

Study Citation:	Jiang, Y., Lu, H., Wang, Y., Hong, H., Wang, Q., Liu, J., Yan, C. (2020). Uptake, biotransformation and physiological response of TBBPA in mangrove plants after hydroponics exposure. Marine Pollution Bulletin 151:110832.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Root uptake
Taxa, Species, Age:	Vegetation; Vascular Plants; *Kandelia obovata*; Juvenile
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10113700

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The TBBPA was identified by name only.
	Metric 2:	Test Substance Source	Low	The source of the TBBPA was not reported.
	Metric 3:	Test Substance Purity	Low	The purity of the TBBPA was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of appropriate concurrent negative controls.
	Metric 5:	Negative Control Response	High	It was reported in section 3.1 that TBBPA was not detected in the roots, stems, and leaves of the control plants.
	Metric 6:	Randomized Allocation	Low	It was not reported how the plants were allocated into study groups. Once the plants were placed into study pots, they were randomly placed in the experimental green house.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	Information regarding the preparation of the test substance was limited. The test substance was reported to be in Hoagland solution with DMSO to aid solubility, but little else was reported. 2.5L of solution was used , and the hydroponic solution was renewed every three days to maintain the test concentration.
	Metric 8:	Consistency of Exposure Administration	High	The exposure administration was administered consistently across study groups. Test groups were maintained in 2.5L of test substance with a 12h light cycle. Environmental conditions were maintained for the duration of the study.
	Metric 9:	Measurement of Test Substance Concentration	Low	It was not reported if the test concentrations were measured at any point during the study.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration was reported to be 14 days. This was adequate to observe the outcomes of interest.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	There were only two exposure groups, but this was adequate to observe the outcomes of interest. Study authors were primarily interested in comparing a high dose and a low dose of TBBPA.
	Metric 12:	Testing at or Below Solubility Limit	Medium	The high dose may be above the water solubility depending on the pH. The low dose appears to be below the water solubility limit. In both doses, DMSO was used to aid the dissolution of the test substance.
Domain 4: Test Organism				

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Study Citation:	Jiang, Y., Lu, H., Wang, Y., Hong, H., Wang, Q., Liu, J., Yan, C. (2020). Uptake, biotransformation and physiological response of TBBPA in mangrove plants after hydroponics exposure. Marine Pollution Bulletin 151:110832.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Root uptake
Taxa, Species, Age:	Vegetation; Vascular Plants; *Kandelia obovata*; Juvenile
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10113700

Domain	Metric		Rating	Comments
	Metric 13:	Test Organism Characteristics	Medium	The mangroves were reported to be from the Zhangjiang estuary in a typical National Nature Mangrove Reserve in Zhangzhou city, Fujian province, China. They appear to be field harvested, which creates concern regarding plant health, though plants were disinfected prior to the test and only viable propagules were chosen for cultivation for four weeks before the experiment start.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The plants were acclimated to test conditions for four weeks prior to the start of the test.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	There were three seedlings per black pot (10.0cm diameter, 14.0cm height) with three replicates performed for each treatment.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	The plants were placed in the experimental green house with a relative humidity of 60-80%, a temperature of 25C +- 5 degrees, and a 12h light photoperiod with 800-1400 umol photons/m^2/s. Hoagland solution provided nutrients.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome(s) of interest–accumulation of TBBPA in the root, stem, and leaf of the plants.
	Metric 18:	Consistency of Outcome Assessment	High	The outcome assessment was carried out consistently across study groups. Concentrations of TBBPA were determined in the root, stem, and leaf of plants at days 7 and 14. The extraction and purification was performed according to Liu et al (2016).
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	The statistical analysis was reported in section 2.6, and was adequate for the outcomes of interest.
	Metric 22:	Reporting of Data	High	The ADME outcomes were reported in Fig 1 for concentrations of TBBPA in the root, stem, and leaf of the plant at days seven and 14. Control results were reported in the text. BCFs were reported in Table S5. TBBPA and transformation products were reported in Table S6.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.

Additional Comments: This evaluation was for the accumulation of TBBPA in K. obovata after a 14 day hydroponic exposure. Samples were taken at days 7 and 14.

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Study Citation:	Jiang, Y., Lu, H., Wang, Y., Hong, H., Wang, Q., Liu, J., Yan, C. (2020). Uptake, biotransformation and physiological response of TBBPA in mangrove plants after hydroponics exposure. Marine Pollution Bulletin 151:110832.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Root uptake
Taxa, Species, Age:	Vegetation; Vascular Plants; *Kandelia obovata*; Juvenile
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10113700

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

Study Citation:	Jiang, Y., Lu, H., Wang, Y., Hong, H., Wang, Q., Liu, J., Yan, C. (2020). Uptake, biotransformation and physiological response of TBBPA in mangrove plants after hydroponics exposure. Marine Pollution Bulletin 151:110832.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Root uptake
Taxa, Species, Age:	Vegetation; Vascular Plants; *Kandelia obovata*; Juvenile
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10113700

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The TBBPA was identified by name only.
	Metric 2:	Test Substance Source	Low	The source of the TBBPA was not reported.
	Metric 3:	Test Substance Purity	Low	The purity of the TBBPA was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of appropriate concurrent negative controls.
	Metric 5:	Negative Control Response	High	The negative control response was reported in Table 1, and was adequate for the outcomes of interest.
	Metric 6:	Randomized Allocation	Low	It was not reported how the plants were allocated into study groups. Once the plants were placed into study pots, they were randomly placed in the experimental green house.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	Information regarding the preparation of the test substance was limited. The test substance was reported to be in Hoagland solution with DMSO to aid solubility, but little else was reported. 2.5L of solution was used , and the hydroponic solution was renewed every three days to maintain the test concentration.
	Metric 8:	Consistency of Exposure Administration	High	The exposure administration was administered consistently across study groups. Test groups were maintained in 2.5L of test substance with a 12h light cycle. Environmental conditions were maintained for the duration of the study.
	Metric 9:	Measurement of Test Substance Concentration	Low	It was not reported if the test concentrations were measured at any point during the study.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration was reported to be 14 days. This was adequate to observe the outcomes of interest.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	There were only two exposure groups, but this was adequate to observe the outcomes of interest. Study authors were primarily interested in comparing a high dose and a low dose of TBBPA.
	Metric 12:	Testing at or Below Solubility Limit	Medium	The high dose may be above the water solubility depending on the pH. The low dose appears to be below the water solubility limit. In both doses, DMSO was used to aid the dissolution of the test substance.
Domain 4: Test Organism				

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Study Citation:	Jiang, Y., Lu, H., Wang, Y., Hong, H., Wang, Q., Liu, J., Yan, C. (2020). Uptake, biotransformation and physiological response of TBBPA in mangrove plants after hydroponics exposure. Marine Pollution Bulletin 151:110832.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Root uptake
Taxa, Species, Age:	Vegetation; Vascular Plants; *Kandelia obovata*; Juvenile
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10113700

Domain	Metric		Rating	Comments
	Metric 13:	Test Organism Characteristics	Medium	The mangroves were reported to be from the Zhangjiang estuary in a typical National Nature Mangrove Reserve in Zhangzhou city, Fujian province, China. They appear to be field harvested, which creates concern regarding plant health, though plants were disinfected prior to the test and only viable propagules were chosen for cultivation for four weeks before the experiment start.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The plants were acclimated to test conditions for four weeks prior to the start of the test.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	There were three seedlings per black pot (10.0cm diameter, 14.0cm height) with three replicates performed for each treatment.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	The plants were placed in the experimental green house with a relative humidity of 60-80%, a temperature of 25C +- 5 degrees, and a 12h light photoperiod with 800-1400 umol photons/m^2/s. Hoagland solution provided nutrients.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome(s) of interest–mangrove biomass for the root, stem, leaf, and total weight.
	Metric 18:	Consistency of Outcome Assessment	High	The outcome assessment was carried out consistently across study groups. Mangroves were washed and rinsed and then divided into roots, stems, and leaves. The fresh weight was then recorded.
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	The statistical analysis was reported in section 2.6, and was adequate for the outcomes of interest.
	Metric 22:	Reporting of Data	High	The biomass outcomes for all exposure concentrations and the control response were reported in Table 1.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.

Additional Comments: This evaluation was for the changes in biomass in K. obovata after exposure to TBBPA.

Overall Quality Determination Medium

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Study Citation:	Jiang, Y., Lu, H., Wang, Y., Hong, H., Wang, Q., Liu, J., Yan, C. (2020). Uptake, biotransformation and physiological response of TBBPA in mangrove plants after hydroponics exposure. Marine Pollution Bulletin 151:110832.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Root uptake
Taxa, Species, Age:	Vegetation; Vascular Plants; *Kandelia obovata*; Juvenile
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10113700

Domain	Metric	Rating	Comments

Study Citation:	Jiang, Y., Lu, H., Wang, Y., Hong, H., Wang, Q., Liu, J., Yan, C. (2020). Uptake, biotransformation and physiological response of TBBPA in mangrove plants after hydroponics exposure. Marine Pollution Bulletin 151:110832.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Root uptake
Taxa, Species, Age:	Vegetation; Vascular Plants; *Kandelia obovata*; Juvenile
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)-Photosynthesis
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10113700

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The TBBPA was identified by name only.
	Metric 2:	Test Substance Source	Low	The source of the TBBPA was not reported.
	Metric 3:	Test Substance Purity	Low	The purity of the TBBPA was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of appropriate concurrent negative controls.
	Metric 5:	Negative Control Response	High	The negative control responses were reported in Figures 3-7 for the mechanistic outcomes, including chlorophyll a and b, MDA, SOD, POD, and CAT.
	Metric 6:	Randomized Allocation	Low	It was not reported how the plants were allocated into study groups. Once the plants were placed into study pots, they were randomly placed in the experimental green house.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	Information regarding the preparation of the test substance was limited. The test substance was reported to be in Hoagland solution with DMSO to aid solubility, but little else was reported. 2.5L of solution was used , and the hydroponic solution was renewed every three days to maintain the test concentration.
	Metric 8:	Consistency of Exposure Administration	High	The exposure administration was administered consistently across study groups. Test groups were maintained in 2.5L of test substance with a 12h light cycle. Environmental conditions were maintained for the duration of the study.
	Metric 9:	Measurement of Test Substance Concentration	Low	It was not reported if the test concentrations were measured at any point during the study.
	Metric 10:	Exposure Duration and Frequency	High	The exposure duration was reported to be 14 days. This was adequate to observe the outcomes of interest.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Medium	There were only two exposure groups, but this was adequate to observe the outcomes of interest. Study authors were primarily interested in comparing a high dose and a low dose of TBBPA.
	Metric 12:	Testing at or Below Solubility Limit	Medium	The high dose may be above the water solubility depending on the pH. The low dose appears to be below the water solubility limit. In both doses, DMSO was used to aid the dissolution of the test substance.
Domain 4: Test Organism				

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Study Citation:	Jiang, Y., Lu, H., Wang, Y., Hong, H., Wang, Q., Liu, J., Yan, C. (2020). Uptake, biotransformation and physiological response of TBBPA in mangrove plants after hydroponics exposure. Marine Pollution Bulletin 151:110832.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Root uptake
Taxa, Species, Age:	Vegetation; Vascular Plants; *Kandelia obovata*; Juvenile
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)-Photosynthesis
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10113700

Domain	Metric		Rating	Comments
	Metric 13:	Test Organism Characteristics	Medium	The mangroves were reported to be from the Zhangjiang estuary in a typical National Nature Mangrove Reserve in Zhangzhou city, Fujian province, China. They appear to be field harvested, which creates concern regarding plant health, though plants were disinfected prior to the test and only viable propagules were chosen for cultivation for four weeks before the experiment start.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	The plants were acclimated to test conditions for four weeks prior to the start of the test.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	There were three seedlings per black pot (10.0cm diameter, 14.0cm height) with three replicates performed for each treatment.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	The plants were placed in the experimental green house with a relative humidity of 60-80%, a temperature of 25C +- 5 degrees, and a 12h light photoperiod with 800-1400 umol photons/m^2/s. Hoagland solution provided nutrients.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome(s) of interest–mechanistic outcomes including chlorophyll a and b, MDA, SOD, POD, and CAT. The outcome assessment methodology was reported clearly.
	Metric 18:	Consistency of Outcome Assessment	High	The outcome assessment was carried out consistently across study groups. Methods were reported clearly.
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	The statistical analysis was reported in section 2.6, and was adequate for the outcomes of interest.
	Metric 22:	Reporting of Data	High	Outcomes for both the controls and the exposure responses were reported in Figures 3-7.
	Metric 23:	Explanation of Unexpected Outcomes	High	Study authors did not report any unexpected outcomes.

Additional Comments: This evaluation was for the effect of TBBPA on mechanistic outcomes in K. obovata, including chlorophyll a and b, MDA, SOD, POD, and CAT, after exposure for 14 days. Samples were also taken at day 7.

Overall Quality Determination Medium

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Study Citation:	Jiang, Y., Lu, H., Wang, Y., Hong, H., Wang, Q., Liu, J., Yan, C. (2020). Uptake, biotransformation and physiological response of TBBPA in mangrove plants after hydroponics exposure. Marine Pollution Bulletin 151:110832.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Cell Culture Media; Root uptake
Taxa, Species, Age:	Vegetation; Vascular Plants; *Kandelia obovata*; Juvenile
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)-Photosynthesis
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	10113700

Domain	Metric	Rating	Comments

Study Citation:	Ltd,, W.I. (2002). Tetrabromobisphenol-A: A toxicity test to determine the effects of the test substance on seedling emergence of six species of plants.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; *Lolium perenne*; Manhattan3; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6391028

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 an independent laboratory, (Appendix 3).
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.17%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods (Appendix 4)
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups
	Metric 15:	Number of Organisms and Replicates per Group	Low	Replicates were not used
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health

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Study Citation:	Ltd,, W.I. (2002). Tetrabromobisphenol-A: A toxicity test to determine the effects of the test substance on seedling emergence of six species of plants.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; *Lolium perenne*; Manhattan3; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6391028

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 were no information presented 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: Emergence, weight, heightLolium perenne (rygrass) data on survival, emergence, and growth data are presented in table 5, page 24/122. NOEC/Ecx data estimates are listed on page 27 in Table 8.

Overall Quality Determination High

Study Citation:	Ltd,, W.I. (2002). Tetrabromobisphenol-A: A toxicity test to determine the effects of the test substance on seedling emergence of six species of plants.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; *Lolium perenne*; Manhattan3; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6391028

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 an independent laboratory, (Appendix 3).
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.17%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods (Appendix 4)
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups
	Metric 15:	Number of Organisms and Replicates per Group	Low	Replicates were not used
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups

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Study Citation:	Ltd,, W.I. (2002). Tetrabromobisphenol-A: A toxicity test to determine the effects of the test substance on seedling emergence of six species of plants.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; *Lolium perenne*; Manhattan3; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6391028

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 were no information presented 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: Lolium perenne (rygrass) data on survival, emergence, and growth data are presented in table 5, page 24/122. NOEC/Ecx data estimates are listed on page 27 in Table 8.

Overall Quality Determination High

Study Citation:	Ltd,, W.I. (2002). Tetrabromobisphenol-A: A toxicity test to determine the effects of the test substance on seedling emergence of six species of plants.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; *Lycopersicon esculentum*; Rutgers; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6391028

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 an independent laboratory, (Appendix 3).
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.17%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods (Appendix 4)
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups
	Metric 15:	Number of Organisms and Replicates per Group	Low	Replicates were not used
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health

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Study Citation:	Ltd,, W.I. (2002). Tetrabromobisphenol-A: A toxicity test to determine the effects of the test substance on seedling emergence of six species of plants.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; *Lycopersicon esculentum*; Rutgers; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6391028

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 were no information presented 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: Emergence, weight, heightLycopersicon esculentum (tomatoes) data on survival, emergence, and growth data are presented in table 7, page 26/122. NOEC/Ecx data estimates are listed on page 27 in Table 8.

Overall Quality Determination High

Study Citation:	Ltd,, W.I. (2002). Tetrabromobisphenol-A: A toxicity test to determine the effects of the test substance on seedling emergence of six species of plants.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; *Lycopersicon esculentum*; Rutgers; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6391028

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 an independent laboratory, (Appendix 3).
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.17%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods (Appendix 4)
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups
	Metric 15:	Number of Organisms and Replicates per Group	Low	Replicates were not used
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups

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Study Citation:	Ltd,, W.I. (2002). Tetrabromobisphenol-A: A toxicity test to determine the effects of the test substance on seedling emergence of six species of plants.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; *Lycopersicon esculentum*; Rutgers; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6391028

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 were no information presented 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: Lycopersicon esculentum (tomatoes) data on survival, emergence, and growth data are presented in table 7, page 26/122. NOEC/Ecx data estimates are listed on page 27 in Table 8.

Overall Quality Determination High

Study Citation:	Sverdrup, L. E., Hartnik, T., Mariussen, E., Jensen, J. (2006). Toxicity of three halogenated flame retardants to nitrifying bacteria, red clover (Trifolium pratense), and a soil invertebrate (Enchytraeus crypticus). Chemosphere 64(1):96-103.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake
Taxa, Species, Age:	Vegetation; Vascular Plants; *Trifolium pratense*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	938766

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Identified as Tetrabromobisphenol A (TBBPA; Cas no 79-94-7)
	Metric 2:	Test Substance Source	High	Purchased FLUKA (Germany) and analytically verified.
	Metric 3:	Test Substance Purity	Low	Purity and grade not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Solvent and negative controls reported.
	Metric 5:	Negative Control Response	High	Response of controls were acceptable.
	Metric 6:	Randomized Allocation	Medium	Seeds were not randomly allocated. Following initial exposure, all containers were placed in a randomized order, which was changed every week. This removes environmental bias.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	Experimental system and test media well described. concentrations were measured in sediment, however, the study reported effects based on nominal concentrations even though table 1 (page 4/8) reports deviations between nominal and actual concentrations. Differences would potentially reduce the reported toxicity of the compound in the results.
	Metric 8:	Consistency of Exposure Administration	High	Exposure administration was consistent across groups.
	Metric 9:	Measurement of Test Substance Concentration	High	TBBPA dissolved in acetone before application to soil. Results were reported with nominal concentrations. Analytical concentrations were separately reported and compared to nominal concentrations.
	Metric 10:	Exposure Duration and Frequency	High	Exposure duration and frequency were reported and acceptable.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Number of exposure groups and spacing were appropriate. A range finding test was conducted.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Sediment was the exposure media.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Species, life stage, and source were adequately described.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	Acclimation was not reported. Pretreatment conditions were equivalent across groups.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Number of organisms and replicates were appropriate for test.

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Study Citation:	Sverdrup, L. E., Hartnik, T., Mariussen, E., Jensen, J. (2006). Toxicity of three halogenated flame retardants to nitrifying bacteria, red clover (Trifolium pratense), and a soil invertebrate (Enchytraeus crypticus). Chemosphere 64(1):96-103.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake
Taxa, Species, Age:	Vegetation; Vascular Plants; *Trifolium pratense*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	938766

Domain	Metric		Rating	Comments
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Housing and environmental conditions were appropriate for test.
	Metric 17:	Outcome Assessment Methodology	High	Intended outcomes were reported.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	There were no reported differences between groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	Outcomes unrelated to exposures were not reported.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical analyses were well described.
	Metric 22:	Reporting of Data	High	Data for all exposures were reported.
	Metric 23:	Explanation of Unexpected Outcomes	High	No unexpected outcomes were reported.

Additional Comments: The data on plant growth is presented on page 6/8 in figure 2. ”No phytotoxicity was observed for TBBPA”

Overall Quality Determination High

Study Citation:	Dogan, M., Korkunc, M., Yumrutas, O. (2012). Effects of bisphenol A and tetrabromobisphenol A on bread and durum wheat varieties. Ekoloji Dergisi 21(85):114-122.
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; *Triticum aestivum*; Sagittario; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5728923

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 grade reported as analytical
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Low	Exposures were administered consistently across study groups but few details were provided
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	Medium	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	The number of exposure groups was lower than desired for a dose response
	Metric 12:	Testing at or Below Solubility Limit	N/A	All concentrations tested (10 and 50 mg/L) are above the water solubility limit of TBBPA listed in the Final Scope (4.15 mg/L). However, the exposure was conducted on filter paper which is acting as a soil surrogate, therefore this metric is marked N/A.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test seeds was not reported
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms,
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate

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Study Citation:	Dogan, M., Korkunc, M., Yumrutas, O. (2012). Effects of bisphenol A and tetrabromobisphenol A on bread and durum wheat varieties. Ekoloji Dergisi 21(85):114-122.
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; *Triticum aestivum*; Sagittario; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5728923

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

Additional Comments: This evaluation is for the assessment of germination.

Overall Quality Determination Medium

Study Citation:	Dogan, M., Korkunc, M., Yumrutas, O. (2012). Effects of bisphenol A and tetrabromobisphenol A on bread and durum wheat varieties. Ekoloji Dergisi 21(85):114-122.
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; *Triticum aestivum*; Sagittario; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Oxidative stress (including redox biology)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5728923

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 grade reported as analytical
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Low	Exposures were administered consistently across study groups but few details were provided
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	Medium	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	The number of exposure groups was lower than desired for a dose response
	Metric 12:	Testing at or Below Solubility Limit	N/A	All concentrations tested (10 and 50 mg/L) are above the water solubility limit of TBBPA listed in the Final Scope (4.15 mg/L). However, the exposure was conducted on filter paper which is acting as a soil surrogate, therefore this metric is marked N/A.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test seeds was not reported
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms,
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest

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Study Citation:	Dogan, M., Korkunc, M., Yumrutas, O. (2012). Effects of bisphenol A and tetrabromobisphenol A on bread and durum wheat varieties. Ekoloji Dergisi 21(85):114-122.
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; *Triticum aestivum*; Sagittario; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Oxidative stress (including redox biology)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5728923

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

Additional Comments: None

Overall Quality Determination Medium

Study Citation:	Dogan, M., Korkunc, M., Yumrutas, O. (2012). Effects of bisphenol A and tetrabromobisphenol A on bread and durum wheat varieties. Ekoloji Dergisi 21(85):114-122.
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; *Triticum aestivum*; Sagittario; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5728923

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 grade reported as analytical
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Low	Exposures were administered consistently across study groups but few details were provided
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	Medium	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	The number of exposure groups was lower than desired for a dose response
	Metric 12:	Testing at or Below Solubility Limit	N/A	All concentrations tested (10 and 50 mg/L) are above the water solubility limit of TBBPA listed in the Final Scope (4.15 mg/L). However, the exposure was conducted on filter paper which is acting as a soil surrogate, therefore this metric is marked N/A.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test seeds was not reported
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms,
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest

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Study Citation:	Dogan, M., Korkunc, M., Yumrutas, O. (2012). Effects of bisphenol A and tetrabromobisphenol A on bread and durum wheat varieties. Ekoloji Dergisi 21(85):114-122.
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; *Triticum aestivum*; Sagittario; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5728923

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

Additional Comments: germination and growth

Overall Quality Determination Medium

Study Citation:	Li, Y., Zhou, Q., Li, F., Liu, X., Luo, Y.,i (2008). Effects of tetrabromobisphenol A as an emerging pollutant on wheat (Triticum aestivum) at biochemical levels. Chemosphere 74(1):119-124.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; *Triticum aestivum*; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Oxidative stress (including redox biology)-Photosynthesis
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3416156

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 97 to 99%, chemical grade reported as analytical
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reasonable
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable for a dose response
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms,
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The number of exposure groups and spacing of exposure levels were suitable for a dose response
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:	Li, Y., Zhou, Q., Li, F., Liu, X., Luo, Y.,i (2008). Effects of tetrabromobisphenol A as an emerging pollutant on wheat (Triticum aestivum) at biochemical levels. Chemosphere 74(1):119-124.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; *Triticum aestivum*; Embryo
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Oxidative stress (including redox biology)-Photosynthesis
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3416156

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 clearly described
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	Medium	responses did not follow a clear dose response although this was noted by authors

Additional Comments: enzymes and chlorophyll response

Overall Quality Determination High

Study Citation:	Dogan, M., Korkunc, M., Yumrutas, O. (2012). Effects of bisphenol A and tetrabromobisphenol A on bread and durum wheat varieties. Ekoloji Dergisi 21(85):114-122.
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; *Triticum durum*; Zenit; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5728923

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 grade reported as analytical
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Low	Exposures were administered consistently across study groups but few details were provided
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	Medium	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	The number of exposure groups was lower than desired for a dose response
	Metric 12:	Testing at or Below Solubility Limit	N/A	All concentrations tested (10 and 50 mg/L) are above the water solubility limit of TBBPA listed in the Final Scope (4.15 mg/L). However, the exposure was conducted on filter paper which is acting as a soil surrogate, therefore this metric is marked N/A.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test seeds was not reported
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms,
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate

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Study Citation:	Dogan, M., Korkunc, M., Yumrutas, O. (2012). Effects of bisphenol A and tetrabromobisphenol A on bread and durum wheat varieties. Ekoloji Dergisi 21(85):114-122.
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; *Triticum durum*; Zenit; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5728923

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

Additional Comments: This evaluation is for the assessment of germination.

Overall Quality Determination Medium

Study Citation:	Dogan, M., Korkunc, M., Yumrutas, O. (2012). Effects of bisphenol A and tetrabromobisphenol A on bread and durum wheat varieties. Ekoloji Dergisi 21(85):114-122.
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; *Triticum durum*; Zenit; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Oxidative stress (including redox biology)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5728923

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 grade reported as analytical
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The study provided only limited details on the measures taken to appropriately prepare test concentrations
	Metric 8:	Consistency of Exposure Administration	Low	Exposures were administered consistently across study groups but few details were provided
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	Medium	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	The number of exposure groups was lower than desired for a dose response
	Metric 12:	Testing at or Below Solubility Limit	N/A	All concentrations tested (10 and 50 mg/L) are above the water solubility limit of TBBPA listed in the Final Scope (4.15 mg/L). However, the exposure was conducted on filter paper which is acting as a soil surrogate, therefore this metric is marked N/A.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test seeds was not reported
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms,
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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
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest

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Study Citation:	Dogan, M., Korkunc, M., Yumrutas, O. (2012). Effects of bisphenol A and tetrabromobisphenol A on bread and durum wheat varieties. Ekoloji Dergisi 21(85):114-122.
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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; *Triticum durum*; Zenit; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Cell signaling/function-Oxidative stress (including redox biology)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
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Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions
	Metric 20:	Outcomes Unrelated to Exposure	Medium	There was no information in the study to suggest differences among groups
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	High	Statistical methods were adequately described
	Metric 22:	Reporting of Data	High	Data for exposure-related findings were presented for each treatment and control group
	Metric 23:	Explanation of Unexpected Outcomes	High	There were no unexpected outcomes

Additional Comments: None

Overall Quality Determination Medium

Study Citation:	Dogan, M., Korkunc, M., Yumrutas, O. (2012). Effects of bisphenol A and tetrabromobisphenol A on bread and durum wheat varieties. Ekoloji Dergisi 21(85):114-122.
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; *Triticum durum*; Zenit; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5728923

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 grade reported as analytical
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	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Low	Researchers did not report how organisms were allocated to study groups
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	Metric 8:	Consistency of Exposure Administration	Low	Exposures were administered consistently across study groups but few details were provided
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	Medium	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	The number of exposure groups was lower than desired for a dose response
	Metric 12:	Testing at or Below Solubility Limit	N/A	All concentrations tested (10 and 50 mg/L) are above the water solubility limit of TBBPA listed in the Final Scope (4.15 mg/L). However, the exposure was conducted on filter paper which is acting as a soil surrogate, therefore this metric is marked N/A.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	The source of the test seeds was not reported
	Metric 14:	Acclimatization and Pretreatment Conditions	High	all pretreatment conditions were the same for control and exposed organisms,
	Metric 15:	Number of Organisms and Replicates per Group	Medium	The numbers of test organisms and replicates were reported and sufficient to characterize toxicological effects
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Low	Environmental conditions were not sufficiently reported to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest

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Study Citation:	Dogan, M., Korkunc, M., Yumrutas, O. (2012). Effects of bisphenol A and tetrabromobisphenol A on bread and durum wheat varieties. Ekoloji Dergisi 21(85):114-122.
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; *Triticum durum*; Zenit; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5728923

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

Additional Comments: germination and growth

Overall Quality Determination Medium

Study Citation:	Ge, H. L., Zhang, F. (2015). The effects of composite photosynthetic bacterial inoculant PS21 on the biochemical characteristics of wheat seedlings under tetrabromobisphenol A stress. Biotechnology and Biotechnological Equipment 29(2):289-298.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake
Taxa, Species, Age:	Vegetation; Vascular Plants; *Triticum sp. (Wheat reported by author)*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Oxidative stress (including redox biology)-Photosynthesis
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3009733

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	High	Accepted common name used [tetrabromobisphenol A (TBBPA)].
	Metric 2:	Test Substance Source	Low	Test substance source was not reported or analytically verified.
	Metric 3:	Test Substance Purity	Low	Purity and grade were not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Use of negative control reported.
	Metric 5:	Negative Control Response	High	Response of negative control acceptable.
	Metric 6:	Randomized Allocation	Medium	Soil samples were randomly allocated into treatment groups prior to treatment.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	Method of concentration measurements at application not reported. No additional measurements reported.
	Metric 8:	Consistency of Exposure Administration	High	Exposures were consistent across groups
	Metric 9:	Measurement of Test Substance Concentration	Low	Measurements of exposure concentrations were not reported.
	Metric 10:	Exposure Duration and Frequency	High	Duration and frequency of exposures were acceptable.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	Exposure groups and spacing were acceptable.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Substance applied to soil for plant uptake.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Wheat seedlings were adequately described.
	Metric 14:	Acclimatization and Pretreatment Conditions	High	Acclimation and pretreatment were adequate.
	Metric 15:	Number of Organisms and Replicates per Group	Medium	Number of seedlings and replicates were acceptable.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions were appropriate for the test.
	Metric 17:	Outcome Assessment Methodology	High	Intended outcomes were reported.

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Study Citation:	Ge, H. L., Zhang, F. (2015). The effects of composite photosynthetic bacterial inoculant PS21 on the biochemical characteristics of wheat seedlings under tetrabromobisphenol A stress. Biotechnology and Biotechnological Equipment 29(2):289-298.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake
Taxa, Species, Age:	Vegetation; Vascular Plants; *Triticum sp. (Wheat reported by author)*; Not Applicable (e.g., fungi or algae studies) or Not Reported
Health Outcome:	Mechanistic-Biomarkers (exposure and effect)-Oxidative stress (including redox biology)-Photosynthesis
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	3009733

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	High	Outcome assessments were consistent across groups.
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	High	No differences reported among groups.
	Metric 20:	Outcomes Unrelated to Exposure	Medium	No outcomes unrelated to exposures reported.
Domain 7: Data Presentation and Analysis				
	Metric 21:	Statistical Methods	Low	Use of statistics and program used. However the type of statistic used was not reported.
	Metric 22:	Reporting of Data	High	Treatment results adequately reported.
	Metric 23:	Explanation of Unexpected Outcomes	High	No unexpected outcomes reported.

Additional Comments: None

Overall Quality Determination High

Study Citation:	Ltd,, W.I. (2002). Tetrabromobisphenol-A: A toxicity test to determine the effects of the test substance on seedling emergence of six species of plants.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; *Zea mays*; Mandan Bride; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6391028

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 an independent laboratory, (Appendix 3).
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.17%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods (Appendix 4)
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups
	Metric 15:	Number of Organisms and Replicates per Group	Low	Replicates were not used
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health

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Study Citation:	Ltd,, W.I. (2002). Tetrabromobisphenol-A: A toxicity test to determine the effects of the test substance on seedling emergence of six species of plants.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; *Zea mays*; Mandan Bride; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6391028

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 were no information presented 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: Emergence, weight, heightZea mays (corn) data on survival, emergance, and growth data are presented in table 20, page 20/122. NOEC/Ecx data estimates are listed on page 27 in Table 8.

Overall Quality Determination High

Study Citation:	Ltd,, W.I. (2002). Tetrabromobisphenol-A: A toxicity test to determine the effects of the test substance on seedling emergence of six species of plants.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; *Zea mays*; Mandan Bride; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6391028

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 an independent laboratory, (Appendix 3).
	Metric 3:	Test Substance Purity	High	Chemical purity reported as 99.17%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group.
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes.
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study groups.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system and methods for preparation of test media were described in adequate detail.
	Metric 8:	Consistency of Exposure Administration	High	exposures were administered consistently across study groups.
	Metric 9:	Measurement of Test Substance Concentration	High	Exposure concentrations were measured using appropriate analytical technologies and methods (Appendix 4).
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure was reported and suitable for the study type.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	High	The number of exposure groups and spacing of exposure levels were suitable.
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	The test organisms were adequately described and were obtained from a reliable source.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	The study did not report whether pretreatment conditions were the same for control and exposed groups.
	Metric 15:	Number of Organisms and Replicates per Group	Low	Replicates were not used
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	Environmental conditions of test system were conducive to maintenance of organism health.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest.
	Metric 18:	Consistency of Outcome Assessment	High	Outcomes were assessed consistently across study groups.

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Study Citation:	Ltd,, W.I. (2002). Tetrabromobisphenol-A: A toxicity test to determine the effects of the test substance on seedling emergence of six species of plants.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Not determined by study authors (i.e., chemical of interest in exposure water, but unable to determine exact uptake route)
Taxa, Species, Age:	Vegetation; Vascular Plants; *Zea mays*; Mandan Bride; Embryo
Health Outcome:	Mortality
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	6391028

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 were no information presented 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: Zea mays (corn) data on survival, emergance, and growth data are presented in table 20, page 20/122. NOEC/Ecx data estimates are listed on page 27 in Table 8.

Overall Quality Determination High

Study Citation:	Wang, Y., Zhang, M., Zhao, L., Zhang, W., Zhao, T., Chu, J., Qiu, Y., Gong, H., Li, R. (2019). Effects of tetrabromobisphenol A on maize (Zea mays L.) physiological indexes, soil enzyme activity, and soil microbial biomass. Ecotoxicology 28(1):1-12.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake
Taxa, Species, Age:	Vegetation; Vascular Plants; *Zea mays*; Embryo
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5123648

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 >97%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study group
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system was described in adequate detail, methods for preparation of test media were minimal
	Metric 8:	Consistency of Exposure Administration	Medium	The study provided few details on exposure administration
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	The number of exposure groups and spacing of exposure levels were suitable for a dose response but effects were observed in lowest concentration
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	Source and variety of seed 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	Only three replicates were used - three beakers per treatment.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Environmental conditions were not sufficiently reported to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest

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Study Citation:	Wang, Y., Zhang, M., Zhao, L., Zhang, W., Zhao, T., Chu, J., Qiu, Y., Gong, H., Li, R. (2019). Effects of tetrabromobisphenol A on maize (Zea mays L.) physiological indexes, soil enzyme activity, and soil microbial biomass. Ecotoxicology 28(1):1-12.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake
Taxa, Species, Age:	Vegetation; Vascular Plants; *Zea mays*; Embryo
Health Outcome:	Reproductive/Teratogenic
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5123648

Domain	Metric		Rating	Comments
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups
Domain 6: Confounding / Variable Control				
	Metric 19:	Confounding Variables in Test Design and Procedures	Low	The study did not provide enough information to allow a comparison of environmental conditions across 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	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 portion of the evaluation was for the germination rate reported in Table 1, which would fall under the reproductive outcome.

Overall Quality Determination Medium

Study Citation:	Wang, Y., Zhang, M., Zhao, L., Zhang, W., Zhao, T., Chu, J., Qiu, Y., Gong, H., Li, R. (2019). Effects of tetrabromobisphenol A on maize (Zea mays L.) physiological indexes, soil enzyme activity, and soil microbial biomass. Ecotoxicology 28(1):1-12.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake
Taxa, Species, Age:	Vegetation; Vascular Plants; *Zea mays*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5123648

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 >97%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study group
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system was described in adequate detail, methods for preparation of test media were minimal
	Metric 8:	Consistency of Exposure Administration	Medium	The study provided few details on exposure administration
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	The number of exposure groups and spacing of exposure levels were suitable for a dose response but effects were observed in lowest concentration
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	Source and variety of seed 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	Only three replicates were used - three beakers per treatment.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Environmental conditions were not sufficiently reported to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups

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Study Citation:	Wang, Y., Zhang, M., Zhao, L., Zhang, W., Zhao, T., Chu, J., Qiu, Y., Gong, H., Li, R. (2019). Effects of tetrabromobisphenol A on maize (Zea mays L.) physiological indexes, soil enzyme activity, and soil microbial biomass. Ecotoxicology 28(1):1-12.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake
Taxa, Species, Age:	Vegetation; Vascular Plants; *Zea mays*; Embryo
Health Outcome:	Development/Growth
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5123648

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 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	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: effects on soil microbial populations also reported

Overall Quality Determination Medium

Study Citation:	Wang, Y., Zhang, M., Zhao, L., Zhang, W., Zhao, T., Chu, J., Qiu, Y., Gong, H., Li, R. (2019). Effects of tetrabromobisphenol A on maize (Zea mays L.) physiological indexes, soil enzyme activity, and soil microbial biomass. Ecotoxicology 28(1):1-12.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake
Taxa, Species, Age:	Vegetation; Vascular Plants; *Zea mays*; Embryo
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)-Photosynthesis
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5123648

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 >97%
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported using an appropriate concurrent negative control group
	Metric 5:	Negative Control Response	High	The biological response of the negative control group was reported and reasonable for assessed outcomes
	Metric 6:	Randomized Allocation	Medium	The study reported that organisms were randomly allocated into study group
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	High	The experimental system was described in adequate detail, methods for preparation of test media were minimal
	Metric 8:	Consistency of Exposure Administration	Medium	The study provided few details on exposure administration
	Metric 9:	Measurement of Test Substance Concentration	Low	Exposure concentrations were not measured
	Metric 10:	Exposure Duration and Frequency	High	The duration of exposure and/or exposure frequency were reported and appropriate for the study type
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	Low	The number of exposure groups and spacing of exposure levels were suitable for a dose response but effects were observed in lowest concentration
	Metric 12:	Testing at or Below Solubility Limit	N/A	Exposure was via soil.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	Low	Source and variety of seed 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	Only three replicates were used - three beakers per treatment.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	Medium	Environmental conditions were not sufficiently reported to evaluate if adequate
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology reported the intended outcome of interest
	Metric 18:	Consistency of Outcome Assessment	High	outcomes were assessed consistently across study groups

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Study Citation:	Wang, Y., Zhang, M., Zhao, L., Zhang, W., Zhao, T., Chu, J., Qiu, Y., Gong, H., Li, R. (2019). Effects of tetrabromobisphenol A on maize (Zea mays L.) physiological indexes, soil enzyme activity, and soil microbial biomass. Ecotoxicology 28(1):1-12.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Soil; Root uptake
Taxa, Species, Age:	Vegetation; Vascular Plants; *Zea mays*; Embryo
Health Outcome:	Mechanistic-Oxidative stress (including redox biology)-Photosynthesis
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	5123648

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 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	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: effects on soil microbial populations also reported

Overall Quality Determination Medium

Study Citation:	Xu, X., Han, W. (2023). Analysis of tetrabromobisphenol A and bisphenol A in plant sample—method optimization and identification of the derivatives. Environmental Science and Pollution Research 30(34):82770-82779.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Water; Root uptake
Taxa, Species, Age:	Vegetation; Vascular Plants; *Zea mays*; Juvenile
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	11520674

Domain	Metric		Rating	Comments
Domain 1: Test Substance				
	Metric 1:	Test Substance Identity	Low	The TBBPA was identified by name only.
	Metric 2:	Test Substance Source	Low	The TBBPA was reported to be from Sigma-Aldrich Inc. It was not reported to be analytically verified.
	Metric 3:	Test Substance Purity	Low	The purity was not reported.
Domain 2: Test Design				
	Metric 4:	Negative Controls	High	Study authors reported the use of a concurrent negative control group.
	Metric 5:	Negative Control Response	High	It was reported in the text that TBBPA levels in all tissues from maize in the negative control group were below the detection limit, which is appropriate for a negative control.
	Metric 6:	Randomized Allocation	Low	The allocation method was not reported.
Domain 3: Exposure Characterization				
	Metric 7:	Experimental System/Test Media Preparation	Low	The preparation of the test substance was not reported. The test system was not described in detail. It was reported this was a hydroponic solution in Hoagland solution, but little else was reported.
	Metric 8:	Consistency of Exposure Administration	Low	Little information was reported on the exposure administration. Exposure environmental conditions were reported, but the test system was unclear, the test chambers were not reported, and the preparation of the test solution were not reported.
	Metric 9:	Measurement of Test Substance Concentration	Low	It was not reported if the exposure concentration was measured.
	Metric 10:	Exposure Duration and Frequency	High	It was reported that the exposure was conducted for 15d. This appeared adequate for the outcome of interest.
	Metric 11:	Number of Exposure Groups/ Spacing of Exposure Levels	N/A	There was only one exposure concentration (100ug/L), as the goal was not to observe a dose response.
	Metric 12:	Testing at or Below Solubility Limit	High	The test concentration was below the water solubility limit.
Domain 4: Test Organism				
	Metric 13:	Test Organism Characteristics	High	Maize seeds were obtained from the Chinese Academy of Agricultural Sciences in Beijing, China. Seeds were sterilized and washed. Then they were germinated on filter paper for three days before being placed into Hoagland solution for TBBPA exposure.
	Metric 14:	Acclimatization and Pretreatment Conditions	Low	It was not reported if the plants were acclimated in any way prior to the start of the study.

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Study Citation:	Xu, X., Han, W. (2023). Analysis of tetrabromobisphenol A and bisphenol A in plant sample—method optimization and identification of the derivatives. Environmental Science and Pollution Research 30(34):82770-82779.
Duration:	Overall Duration: 11 - 21 days; Exposure Duration: 11 - 21 days
Exposure Route, Media, Path:	Terrestrial; Water; Root uptake
Taxa, Species, Age:	Vegetation; Vascular Plants; *Zea mays*; Juvenile
Health Outcome:	ADME (biotransformation)
Chemical:	4,4-(1-Methylethylidene)bis[2,6-dibromophenol] (TBBPA)
HERO ID:	11520674

Domain	Metric		Rating	Comments
	Metric 15:	Number of Organisms and Replicates per Group	Low	There were three replicates per test exposure and control. The number of plants per test chamber was unclear.
Domain 5: Outcome Assessment				
	Metric 16:	Adequacy of Test Conditions	High	The test temperature was reported to be 20-25C, the relative humidity was 60-70%, the photoperiod was 14L:10D, and the light intensity was 132W/m^2. Hoagland solution was used to provide nutrients during testing.
	Metric 17:	Outcome Assessment Methodology	High	The outcome assessment methodology addressed or reported the intended outcome(s) of interest–uptake of TBBPA into roots, stems, and leaves of maize.
	Metric 18:	Consistency of Outcome Assessment	High	The outcome assessment was reported and carried out consistently.
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	It was reported that Origin Pro 9.1.1 was used to analyze the data, but little other information was provided.
	Metric 22:	Reporting of Data	Low	The results were described in the text for the exposure findings and the control results.
	Metric 23:	Explanation of Unexpected Outcomes	Low	This portion of the study did not report measures of variability.

Additional Comments: This evaluation was for the accumulation of TBBPA in maize roots, stems, and leaves after a 15 day hydroponic exposure. ADME was selected as the outcome of interest. The primary goal of this paper was optimize a method for determining TBBPA and BPA in plant samples, so the majority of the paper focused on this aspect. The uptake study was done as confirmation of methods.

Overall Quality Determination Medium