# Toxicant default guideline values for aquatic ecosystem protection

Alpha-cypermethrin in freshwater

Technical brief

July 2023

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## Summary

The default guideline values (DGVs) and associated information in this technical brief should be used in accordance with the detailed guidance provided in the Australian and New Zealand Guidelines for Fresh and Marine Water Quality website (www.waterquality.gov.au/anz-guidelines).

Alpha-cypermethrin is a contact and ingested pyrethroid insecticide applied to many agricultural crops to control chewing and sucking insects (WHO 1992, PPDB 2015). In Australia and New Zealand, it is used to control insects on a wide range of fruit, vegetable and cereal crops, as well as insects and spiders in domestic and commercial settings, and for the treatment of animal ectoparasites (ACVM 2020, APVMA 2020). Alpha-cypermethrin has a low aqueous solubility, is non-volatile and is persistent in water and sediment (WHO 1992, 2009, NCBI 2020).

Alpha-cypermethrin has a very fast knockdown action (i.e. it acts within minutes of application) (WHO 1992). Its mode of action is on the central nervous system, acting specifically on the sodium channels, causing paralysis and eventual death (WHO 1992). It also has a repellent and anti-feeding action on some insects. Based on a review of the aquatic toxicology, and consistent with its mode of action, alpha-cypermethrin is highly toxic to fish and aquatic invertebrates. Toxicity values for alpha-cypermethrin have been reported as low as 0.019 µg/L for the shrimp Paratya australiensis (acute LC50) and 0.025 µg/L for the cladoceran Ceriodaphnia dubia (chronic NOEC).

Alpha-cypermethrin is a racemic mixture of two isomers: (1R, cis)S and (1S, cis)R (WHO 2009). In the preparation of the default guideline values (DGVs) for alpha-cypermethrin, only data for these isomers were used. Data from toxicity tests that used insecticide formulations containing alpha-cypermethrin as the active ingredient were excluded from the DGV derivation because the toxicity of the carrier solvent (and other ingredients where stated) was not known.

Moderate reliability DGVs were derived based on chronic NOEC and acute LC50 (converted to chronic) data for 14 species from seven taxonomic groups, with a good fit of the distribution to the toxicity data. The DGVs for 99%, 95%, 90% and 80% species protection are 0.001 µg/L, 0.006 µg/L, 0.013 µg/L and 0.037 µg/L, respectively. The DGVs may be below current analytical limits of reporting. The 95%species protection level for alpha-cypermethrin (0.006 µg/L) is recommended for adoption in the assessment of slightly-to-moderately disturbed ecosystems.

## Introduction

Alpha-cypermethrin (CASRN 67375-30-8 and molecular formula C22H19Cl2NO3) is a pyrethroid insecticide and racemic (i.e. 1:1) mixture of two enantiomeric (i.e. mirror image) cis-isomers (WHO 2009):

* the (1R, cis)S isomer (or (S)-α-cyano-3-phenoxybenzyl-(1R,3R)-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropane-carboxylate)
* the (1S, cis)R isomer (or (R)-α-cyano-3-phenoxybenzyl-(1S,3S)-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropane-carboxylate).

These isomers represent two of eight isomers (four cis-isomers and four trans-isomers) present in the insecticide cypermethrin. Cypermethrin is formulated as four different insecticides: alpha-, beta-, theta- and zeta-cypermethrin (ATSDR 2003). Application rates of alpha-cypermethrin are lower than those of beta-, theta- and zeta-cypermethrin because the alpha-isomers are more biologically active than the beta-, theta- and zeta-isomers (WHO 1992).

Alpha-cypermethrin is approved for use in Australia (APVMA 2004) as a contact and ingested pyrethroid insecticide to control chewing and sucking insects, such as Lepidoptera, Coleoptera, and Hemiptera (WHO 1992, PPDB 2015). In Australia and New Zealand, it is used primarily to control insects on a wide range of fruit (e.g. apple, grape, avocado, kiwifruit, pear, stone fruit), vegetable (e.g. broccoli, cabbage, cauliflower, kale, lettuce, tomato) and other agricultural (e.g. canola, cotton, lucern, maize, rice) crops (ACVM 2020, APVMA 2020). It is also used domestically and commercially to control insects, such as ants, cockroaches, mosquitoes and flies (ACVM 2020, APVMA 2020), and in veterinary treatment applications, such as animal ectoparasiticides (APVMA 2020).

Alpha-cypermethrin’s mode of action is on the central nervous system, acting specifically by causing delayed closure of the sodium channels, causing paralysis and eventual death (WHO 1992). Alpha-cypermethrin has a very quick knockdown action (i.e. it acts within minutes of application) (WHO 1992).

In agricultural use, alpha-cypermethrin is often applied as a suspension concentrate or emulsifiable concentrate in a carrier solvent. Most emulsifiable concentrates contain between 25% and 75% of the active ingredient (Fishel 2013). Based on a review of readily available information for pesticide formulations containing alpha-cypermethrin registered for use in Australia, emulsifiable concentrate and suspension concentrate formulations of alpha-cypermethrin contain approximately 10% alpha-cypermethrin.

Alpha-cypermethrin is a crystalline powder with a weak aromatic odour (WHO 1992). It has low water solubility (0.005–0.01 mg/L at 25°C) (WHO 2013, Kegley et al. 2014, PPDB 2015, NCBI 2020) and is non-volatile (reported vapour pressure of 1.73x10-5 mm Hg at 20°C) (NCBI 2020). Estimated volatilisation half-lives were reported for a model river—8 days—and model lake—65 days (NCBI 2020). As it is stable in sunlight, photolysis in water is not an important fate process.

Alpha-cypermethrin is stable under acidic and neutral conditions but hydrolyses at pH 12–13 (WHO 1992). The hydrolysis half-life (DT50) has been reported to be 101 days at pH 7 and 20°C (WHO 2009, PPDB 2015, Kegley et al. 2014). With a LogKoc of 4.8 (Kegley et al. 2014), alpha-cypermethrin is expected to adsorb to sediments. Information reviewed on biodegradation potential of alpha-cypermethrin was limited and conflicting. Some sources indicated that alpha-cypermethrin is readily biodegraded by microorganisms based on its structural similarity to other pyrethroids (NCBI 2020). Other sources indicated greater persistence and slow rates of breakdown in soils (PPDB 2015) and in ready biodegradability tests (WHO 1992).

A LogKow value of 6.94 (NCBI 2020) suggests that alpha-cypermethrin may bioaccumulate. However, review of the available information for cypermethrin, which is expected to behave similarly to alpha-cypermethrin (WHO 1992), suggests that alpha-cypermethrin would not bioaccumulate. This is based on the experimentally-determined bioaccumulation in fish for cypermethrin, which was lower than might have been anticipated from the Kow (WHO 1992). Concentrations of cypermethrin in fish decreased when exposure ceased, presumably because cypermethrin is rapidly metabolised.

Although specific methods exist for quantifying alpha-cypermethrin in water matrices, they require complex sample manipulation that cannot be performed as part of a routine analyte screen. Routine analyses cannot discriminate between the eight isomers of cypermethrin; thus, results are reported as ‘cypermethrin (sum of isomers)’. This has implications for assessing monitoring data against the alpha-cypermethrin default guideline values (DGVs), which are addressed in Section 4.3.

## Aquatic toxicology

A literature review of the effects of alpha-cypermethrin on freshwater organisms indicated less extensive research has been undertaken compared to other isomeric forms of cypermethrin. The review (and associated data quality assessment) identified toxicity data for 20 species—consisting of 13 chronic values for seven species, and 25 acute values for 16 species.

Some toxicity studies assessed formulations containing alpha-cypermethrin as the active ingredient. These formulations include a carrier solvent and, in some cases, other proprietary ingredients, for which the combined toxicity is not well understood. Accordingly, such studies are typically not appropriate for deriving guideline values for active ingredients.

Of the chronic studies for alpha-cypermethrin of purity >80%, crustaceans were the most sensitive and microalgae were the least sensitive taxonomic groups. Chronic NOECs for the cladocerans Ceriodaphnia dubia and Daphnia magna were approximately 0.03 µg/L (8-d growth) and 0.04 µg/L (21-d reproduction), respectively (Shen et al. 2012, USEPA 2019). In contrast, chronic NOECs for plant, algal and cyanobacterial species appear to be two to four orders of magnitude higher (i.e. less toxic); for example, 1.4 µg/L for the macrophyte Lemna gibba (7-d growth), 27 µg/L for the cyanobacterium Anabaena flos-aquae (4-d growth), and 72 µg/L for the diatom Navicula pelliculosa (4-d growth) (USEPA 2019). There are few chronic toxicity data available for fish. A chronic LOEC of 0.02 µg/L (30-d reproduction) was reported for the zebrafish Danio rerio; however, the purity of alpha-cypermethrin used for the testing was not stated (Ansari & Ansari 2012).

The majority of available aquatic toxicity studies for alpha-cypermethrin represent acute exposures, although approximately half of the studies assessed formulations containing alpha-cypermethrin and/or did not state the test chemical purity, making it difficult to draw conclusions about alpha-cypermethrin toxicity. Of the data based on alpha-cypermethrin of >80% purity, crustaceans were the most sensitive. Kumar et al. (2010) reported a 96-h LC50 of 0.019 µg/L for the freshwater shrimp Paratya australiensis. High acute sensitivity has also been reported for some fish species, with 96-h LC50s between 0.5 µg/L and 1 µg/L (Stephenson 1990, Jahanbakhshi et al. 2012, Shaluei et al. 2012), but up to approximately 10 µg/L has also been reported for other fish species (Yilmaz et al. 2004, Yilmaz 2005). Alpha-cypermethrin was less toxic to larvae of the mosquito Anopheles sinensis, with a 96-h LC50 of 60 µg/L (Chang et al. 2009).

## Factors affecting toxicity

Some evidence suggests that, in general, temperature, suspended solids and dissolved organic carbon affect the toxicity of cypermethrin and pyrethroids (Yang et al. 2006, Fojut et al. 2011). However, there are currently insufficient data to establish empirical relationships between abiotic factors and the toxicity of alpha-cypermethrin to freshwater organisms. Given the low solubility and high binding potential for pyrethroids and cypermethrin, only dissolved (filtered) concentrations of alpha-cypermethrin should be assessed against the DGVs.

## Default guideline value derivation

The DGVs were derived in accordance with the method described in Warne et al. (2018) and using Burrlioz 2.0 software.

### Toxicity data used in derivation

A summary of the toxicity data (one value per species) and conversions used to calculate the DGVs for alpha-cypermethrin in freshwater is provided in Table 1. Further details on the data that passed the screening and quality assurance schemes, including those used to derive the single species values used to calculate the DGVs, are presented in Appendix A: Toxicity data that passed the screening and quality assessment and used to derive the default guideline values, Table A 1. Details of the [data quality assessment](http://www.waterquality.gov.au/sites/default/files/documents/alpha-cypermethrin-fresh-dgvs-data-quality-assessment.xlsm) and the [data that passed the quality assessment](http://www.waterquality.gov.au/sites/default/files/documents/alpha-cypermethrin-fresh-dgvs-data-entry.xlsx) are provided as supporting information.

Only data for the alpha-isomers were used. Results from toxicity testing using insecticide formulations containing alpha-cypermethrin as the active ingredient were excluded from the DGV derivation because the toxicity of the carrier solvent (and other ingredients where stated) was not known (see Section 2). Additionally, results from studies where the alpha-cypermethrin purity was not known or was <80% were excluded. For example, chronic toxicity data for two species—the freshwater snail Lymnaea acuminate (hatchling mortality LOEC of 4 µg/L after 28-d exposure (Tripathi & Singh 2004)) and the zebrafish D. rerio (reproduction LOEC for the number of viable eggs, 0.02 µg/L after 30-d exposure (Ansari & Ansari 2012))—were excluded from the DGV derivation because the test chemical purity was not stated. In addition, acute toxicity data for six species were excluded because an alpha-cypermethrin formulation was tested or the purity of alpha-cypermethrin was not stated. The purity of alpha-cypermethrin in the studies used to calculate the DGVs was >90%.

Table  Summary of single chronic and estimated chronic toxicity data values, all species used to derive default guideline values for alpha-cypermethrin in freshwater

| Taxonomic group | Species | Life stage | Duration (h) | Type (acute/chronic) | Toxicity measure **a** | Reported toxicity value (µg/L) | Final toxicity value (µg/L) |
| --- | --- | --- | --- | --- | --- | --- | --- |
| Cyanobacterium | Anabaena flos-aquae | – | 96 | Chronic | NOEC | 27.2 | 27.2 **b** |
| Diatom | Navicula pelliculosa | – | 96 | Chronic | NOEC | 72 | 72 **b** |
| Macrophyte | Lemna gibba | – | 168 | Chronic | NOEC | 1.39 | 1.39 **b** |
| Crustacean | Paratya australiensis | Adults | 96 | Acute | LC50 | 0.019 | 0.002 **c** |
| Ceriodaphnia dubia | Neonates | 192 | Chronic | NOEC | 0.025 | 0.025 **b** |
| Daphnia magna | Neonates | 504 | Chronic | NOEC | 0.037 | 0.037 **b** |
| Insect | Culex tritaeniorhynchus | Larvae | 24 | Acute | LC50 | 1.43 | 0.143 **c** |
| Anopheles sinensis | Larvae | 24 | Acute  | LC50 | 60 | 6 **c** |
| Amphibian | Xenopus laevis | Larvae | 96 | Acute | LC50 | 6.9 | 0.69 **c** |
| Fish | Rutilus rutilus capsicus | Juveniles | 96 | Acute | LC50 | 0.627 | 0.063 **c** |
| Hypophthalmicthys molitrix | Juveniles | 96 | Acute | LC50 | 0.917 | 0.092 **c** |
| Huso huso | Juveniles | 96 | Acute  | LC50 | 0.952 | 0.095 **c** |
| Oreochromis niloticus | Larvae | 96 | Acute | LC50 | 3.42 | 0.342 **c** |
| Poecilia reticulata | Adults | 96 | Acute | LC50 | 9.43 | 0.943 **c** |

Note: Final toxicity values are reported to no more than three significant figures.

**a** The measure of toxicity being estimated/determined: LC50: median lethal concentration; NOEC: No observed effect concentration.

**b** Actual chronic NOEC.

**c** Default conversion from acute LC50 to chronic negligible effect (NOEC/EC10) concentration: Acute LC50/10 = Chronic negligible effect concentration.

Toxicity data from studies that reported nominal alpha-cypermethrin concentrations (or did not state whether the test chemical was measured) were included in the derivation. This was largely because several studies have assessed and reported toxicity at concentrations below current analytical reporting limits (e.g. Kumar et al. (2010) for P. australiensis, Shen et al. (2012) for C. dubia), and to exclude them from the derivation would have potentially resulted in under-protective DGVs.

Where only one value was available for a species, that value was used for the calculation of the species sensitivity distribution (SSD). For species with more than one value available, the data selected for the SSD was in accordance to Warne et al.(2018). Overall, 14 species from seven taxonomic groups were considered for the SSD. These species included: one cyanobacterium (A. flos-aquae); one diatom (N. pelliculosa); one macrophyte (L. gibba); one microinvertebrate crustacean (C. dubia); two macroinvertebrate crustaceans (P. australiensis and D. magna); two insects (Culex tritaeniorhynchus and A. sinensis); one amphibian (Xenopus laevis); and five fish (Rutilus rutilus caspicus, Hypophthalmicthys molitrix, Huso huso, Oreochromis niloticus and Poecilia reticulata). Of the toxicity data used for these 14 species, five were from chronic exposures (reported as NOECs), and nine were from acute exposures. The nine acute toxicity values were all LC50 values and were converted to estimated chronic negligible effect (e.g. NOEC/EC10) values using the default acute-to-chronic ratio of 10.

Modality checks were performed according to the four questions stipulated in Warne et al. (2018), with the details of the assessment provided in Appendix B: Modality assessment for alpha-cypermethrin. The weight of evidence assessment concluded that the dataset did not exhibit bimodality or multimodality and, hence, supported use of the data for the 14 species for derivation of the DGVs.

### Species sensitivity distribution

The cumulative frequency (species sensitivity) distribution (SSD) of the 14 chronic and converted acute alpha-cypermethrin toxicity data reported in Table 1 is shown in Figure 1. The SSD was plotted using the Burrlioz 2.0 software. The model was judged to provide a good fit to the data (Figure 1**Error! Reference source not found.**).

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Note: Units on the SSD are ng/L; elsewhere in this technical brief they are presented as µg/L.

Figure  Species sensitivity distribution, alpha-cypermethrin in freshwater

### Default guideline values

It is important that the DGVs (Table 2) and associated information in this technical brief are used in accordance with the detailed guidance provided in the Australian and New Zealand Guidelines for Fresh and Marine Water Quality [website](https://www.waterquality.gov.au/anz-guidelines) (ANZG 2018).

The alpha-cypermethrin DGVs for 99%, 95%, 90% and 80% species protection are shown in Table 2. The 95% species protection DGV of 0.006 µg/L is recommended for application for slightly-to-moderately disturbed ecosystems. It is important to note that the DGVs are below current analytical limits of reporting for alpha-cypermethrin (i.e. 0.2–0.5 µg/L). However, the available toxicity data indicate that toxic effects can occur below the current limits of reporting. ANZG (2018) provides guidance on what to do if DGVs are below analytical detection limits (see [Accounting for local conditions](https://www.waterquality.gov.au/anz-guidelines/guideline-values/default/water-quality-toxicants/local-conditions#default-guideline-values-and-analytical-detection-limits)).

Table  Toxicant default guideline values, alpha-cypermethrin in freshwater, moderate reliability

| Level of species protection (%) | DGV for alpha-cypermethrin in freshwater (μg/L)a |
| --- | --- |
| 99 | 0.001 |
| 95 | 0.006 |
| 90 | 0.013 |
| 80 | 0.037 |

**a The** DGVs were derived using the Burrlioz 2.0 software and rounded to either one or two significant figures.

Given that routine pesticide analyses cannot discriminate between the two isomers that make up alpha-cypermethrin from the other six isomers of cypermethrin, it is recommended that the cypermethrin concentration of the sample, which represents the sum of all isomers, is compared with the alpha-cypermethrin DGVs. If the cypermethrin concentration exceeds the relevant DGV, further investigation is required, which could include a more complex analysis specific to alpha-cypermethrin.

The DGVs were compared to the raw chronic toxicity data compiled from the literature review (i.e. 11 chronic values for five species). This check confirmed that the theoretical protection offered by the DGVs is expected to be adequate.

### Reliability classification

The alpha-cypermethrin freshwater DGVs have a moderate reliability classification (Warne et al. 2018) based on the outcomes for the following three criteria:

* Sample size—14 (good)
* Type of toxicity data—chronic and converted acute data
* SSD model fit—good (Burr Type III model).

## Glossary

| Term | Definition |
| --- | --- |
| acute toxicity | A lethal or adverse sublethal effect that occurs as the result of a short exposure period to a chemical relative to the organism’s life span. |
| acute-to-chronic ratio  | The species mean acute value (LC/EC50) divided by the chronic value (e.g. NOEC or EC10) for the same species. |
| bioaccumulation | The process by which chemical substances are accumulated by aquatic organisms by all routes of exposures (dietary and the ambient environment). |
| chronic toxicity | A lethal or sublethal adverse effect that occurs after exposure to a chemical for a period of time that is a substantial portion of the organism’s life span or an adverse effect on a sensitive early life stage. |
| default guideline value (DGV) | A guideline value recommended for generic application in the absence of a more specific guideline value (e.g. a site-specific guideline value) in the Australian and New Zealand Guidelines for Fresh and Marine Water Quality. Formerly known as ‘trigger values’. |
| EC50 (median effective concentration) | The concentration of a substance in water or sediment that is estimated to produce a 50% change in the response being measured or a certain effect in 50% of the test organisms relative to the control response, under specified conditions. |
| endpoint | The specific response of an organism that is measured in a toxicity test (e.g. mortality, growth, a particular biomarker). |
| guideline value (GV) | A measurable quantity (e.g. concentration) or condition of an indicator for a specific community value below which (or above which, in the case of stressors such as pH, dissolved oxygen and many biodiversity responses) there is considered to be a low risk of unacceptable effects occurring to that community value. Guideline values for more than one indicator should be used simultaneously in a multiple lines of evidence approach. (Also refer to [default guideline value](https://www.waterquality.gov.au/anz-guidelines/resources/glossary#default-guideline-value) and [site-specific guideline value](https://www.waterquality.gov.au/anz-guidelines/resources/glossary#site-specific-guideline-value).) |
| humic substances | Organic substances only partially broken down that occur in water mainly in a colloidal state. Humic acids are large-molecule organic acids that dissolve in water. |
| LC50 (median lethal concentration) | The concentration of a substance in water or sediment that is estimated to be lethal to 50% of a group of test organisms, relative to the control response, under specified conditions. |
| LOEC (lowest observed effect concentration) | The lowest concentration of a material used in a toxicity test that has a statistically significant adverse effect on the exposed population of test organisms as compared with the controls. |
| macrophyte | A member of the macroscopic plant life of an area, especially of a body of water; large aquatic plant. |
| NOEC (no observed effect concentration) | The highest concentration of a material used in a toxicity test that has no statistically significant adverse effect on the exposed population of test organisms as compared with the controls. |
| periphyton | The organisms attached to submerged plants. |
| site-specific guideline value | A guideline value that is relevant to the specific location or conditions that are the focus of a given assessment or issue. |
| species (biological) | A group of organisms that resemble each other to a greater degree than members of other groups and that form a reproductively isolated group that will not produce viable offspring if bred with members of another group. |
| species sensitivity distribution (SSD)  | A method that plots the cumulative frequency of species’ sensitivities to a toxicant and fits a statistical distribution to the data. From the distribution, the concentration that should theoretically protect a selected percentage of species can be determined. |
| toxicity | The inherent potential or capacity of a material to cause adverse effects in a living organism. |
| toxicity test | The means by which the toxicity of a chemical or other test material is determined. A toxicity test is used to measure the degree of response produced by exposure to a specific level of stimulus (or concentration of chemical) for a specified test period. |

## Appendix A: Toxicity data that passed the screening and quality assessment and used to derive the default guideline values

Table A  Summary, toxicity data that passed the screening and quality assurance processes, alpha-cypermethrin in freshwater

| Taxonomic group | Species | Life stage | Exposure duration (h) | Test type | Toxicity measure a(test endpoint) | Test medium | Temperature (°C) | pH | Concentration (µg/L)  | Reference |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Cyanobacterium | Anabaena flos-aquae | – | 96 | Chronic | NOEC (Growth) | – | – | – | 27.2 **b** | USEPA (2019) |
| Diatom | Navicula pelliculosa | – | 96 | Chronic | NOEC (Growth) | – | – | – | 72 **b** | USEPA (2019) |
| Macrophyte | Lemna gibba | – | 168 | Chronic | NOEC (Growth) | – | – | – | 1.39 **b** | USEPA (2019) |
| Crustacean | Paratya australiensis | Adults | 96 | Acute | LC50 (Survival) | Water | 22.6–24.2 | 7.7–7.9 | 0.019 **c** | Kumar et al. (2010) |
| Ceriodaphnia dubia | Neonates | 192 | Chronic | NOEC (Growth) | Moderately hard water | 23–25 | 8.0–8.1 | 0.025 **b** | Shen et al. (2012) |
| Daphnia magna | Neonates | 504 | Chronic | NOEC (Reproduction) | – | – | – | 0.037 **b** | USEPA (2019) |
| Insect | Culex tritaeniorhynchus | Larvae | 24 | Acute | LC50 (Survival) | Dechlorinated water | 22–24 | 7.0–7.4 | 1.43 **c** | Tripathi and Singh (2004) |
| Anopheles sinensis | Larvae | 24 | Acute | LC50 (Survival) | Dechlorinated tap water | – | – | 60 **c** | Chang et al. (2009) |
| Amphibian | Xenopus laevis | Larvae | 96 | Acute | LC50 (Survival) | FETAX Solution | 22–24 | 6.5–8.0 | 6.9 **c** | Yu et al. (2013) |
| Fish | Rutilus rutilus caspicus | Juveniles | 96 | Acute | LC50 (Survival) | Dechlorinated tap water | 22–24 | 7.0–7.4 | 0.627 **c** | Shaluei et al. (2012) |
| Hypophthalmicthys molitrix | Juveniles | 96 | Acute | LC50 (Survival) | Dechlorinated tap water | 18.5–21.5 | 7.1–8.6 | 0.917 **c** | Stephenson (1990) |
| Huso huso | Juveniles | 96 | Acute | LC50 (Survival) | Tap water | 22–24 | 7.8–8.0 | 0.952 **c** | Jahanbakhshi et al. (2012) |
| Oreochromis niloticus | Larvae | 96 | Acute | LC50 (Survival) | Aquarium water | 22–24 | 7.1–7.3 | 3.42 **c** | Yilmaz (2005) |
| Poecilia reticulata | Adults | 96 | Acute | LC50 (Survival) | Well water | 20–25 | 7.2 | 9.43 **c** | Yilmaz et al.(2004) |

**a** The measure of toxicity being estimated/determined: LC50: the lethal concentration for 50% of the test organisms; NOEC: No observed effect concentration.

**b** Value was used as is for the DGV derivation.

**c** Value was divided by default acute-to-chronic ratio of 10 to estimate the chronic negligible effect (NOEC/EC10) value for use in DGV derivation.

## Appendix B: Modality assessment for alpha-cypermethrin

A modality assessment was undertaken for alpha-cypermethrin according to the four questions stipulated in Warne et al. 2018. These questions and their answers are as follows.

##### Is there a specific mode of action that could result in taxa-specific sensitivity?

As discussed in Section 1, alpha-cypermethrin acts on the central nervous system, specifically by causing delayed closure of the sodium channels, paralysis and eventual death. Given ion channels in plants are also important in plant survival, it is unknown if there would be taxa-specific sensitivity to alpha-cypermethrin. Therefore, both plant and animal data were considered in the DGV derivation.

##### Does the dataset suggest bimodality?

Visual representation of the data, calculation of the bimodality coefficient (BC), and consideration of the range in the effect concentrations are recommended lines of evidence for evaluating whether bimodality or multimodality of the dataset is apparent. For this assessment:

* the histogram of the raw effect concentration SSD data (Figure B 1, the histogram on the left) could be interpreted as positively right skewed, typical of concentration-based data (Warne et al. 2018). The log transformed histogram generally follows a normal distribution (Figure B 1, the histogram on the right)
* data that span large ranges (>4 orders of magnitude) indicate potential for underlying bimodality or multimodality (Warne et al. 2018); the alpha-cypermethrin data span 4 orders of magnitude
* when the BC is greater than 0.555, it indicates that the data do not follow a typical normal distribution and may be bimodal; the BC for the log transformed data is 0.285 and does not support an assertion of bimodality.

Based on the lines of evidence described above, the distribution of the log transformed dataset is generally in accordance with a unimodal normal distribution.



Figure B 1 Histogram, raw data (left) and log transformed data (right)

##### Do data show taxa-specific sensitivity (i.e. through distinct groupings of different taxa types)?

As the mode of action of alpha-cypermethrin is likely to affect both plants and animals, the potential for taxa-specific sensitivity in the data was examined using box plots of the SSD data with the grouping variable phyla, and major organism types (e.g. plants, vertebrates) (Figure B 2).

As shown in Figure B 2, there is a trend for macroinvertebrates (Arthropoda) to be more sensitive to alpha-cypermethrin than other taxonomic groups. However, there are insufficient data for plants (Tracheophyta and Bacillariophyta) and Cyanobacteria to enable definitive conclusions.



Note: An asterisk represents an outlying value >1.5x the interquartile range. An open circle represents an outlying value >3x the interquartile range.

Figure B 2 Box plots, raw (left) and log transformed (right) data grouped by major types of organisms

Further investigation (Figure B 3), which considered organisms that are taxonomically distinct, indicated that crustaceans appear to be the most sensitive group. However, as there are only three species in this grouping, it is difficult to draw definitive conclusions.



Note: An asterisk represents an outlying value >1.5x the interquartile range. An open circle represents an outlying value >3x the interquartile range.

Figure B  Box plots, raw (left) and log transformed (right) data grouped by taxonomically distinct organisms

##### Is it likely that indications of bimodality or multimodality or distinct clustering of taxa groups are not due to artefacts of data selection, small sample size, test procedures, or other reasons unrelated to a specific mode of action?

The data do not show signs of bimodality or multimodality. Crustaceans appear to be the most sensitive group (Figure B 3). However, this is potentially due to artefacts associated with the small sample size of the dataset. Thus, on the basis of the available evidence, the dataset appears to be unimodal.

The weight of evidence supports use of the 14 species identified in the preparation of the SSD.

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