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GUIDELINES FOR  
FRESH & MARINE  
WATER QUALITY

# Toxicant default guideline values for aquatic ecosystem protection

## Dissolved copper in marine water

Technical brief

May 2023

DRAFT

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New Zealand Government



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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](http://www.waterquality.gov.au/anz-guidelines)).

Copper is widely distributed in the Earth's crust and is an essential trace element for micro-organisms, plants and animals. It is commonly used in roofing, plumbing, electrical wiring and electronics. Copper is also used as a biocide in antifouling paints, wood preservatives, fungicides and herbicides. The major anthropogenic sources of copper for marine environments include antifouling paints, stormwater, municipal wastewater discharges, mining and mineral processing, and metal and electrical manufacturing.

Since the last revision of the marine copper DGVs in 2000, new data have become available, including high quality local species data. Furthermore, there is increased understanding of the effect of dissolved organic carbon (DOC) on copper toxicity. Increases in DOC reduce the aquatic toxicity of copper, as copper binds to DOC, decreasing the available free copper ion concentrations. Consequently, the current DGVs incorporate a correction for DOC.

High reliability DGVs for dissolved copper in marine water were derived from chronic (long-term) toxicity data for 32 species comprising four diatoms, four brown microalgae, one blue-green alga, three green microalgae, two green macroalgae, two brown macroalgae, four cnidarians, two echinoderms, one annelid, one crustacean, six molluscs and two fish. The DGVs (at salinity 25–36‰ and DOC ≤0.5 mg/L) for 99%, 95%, 90% and 80% species protection are 0.12 µg/L, 0.40 µg/L, 0.72 µg/L and 1.4 µg/L, respectively. Adjustments to the DGVs can be made when DOC is >0.5 mg/L, although no further adjustment should be made when DOC is >6 mg/L. The 95% species protection values for copper in marine water are recommended for adoption in the assessment of slightly-to-moderately disturbed ecosystems.

The Australian and New Zealand guidelines for fresh and marine water quality ([ANZG 2018](#)) provides guidance for evaluating monitoring data against DGVs and recommends a decision scheme that includes consideration of the bioavailable fraction. Such guidance for metals, presented as a decision tree, is provided in Appendix A: Actions to assess the bioavailable fraction of a metal.

Appendix B: Toxicity data that passed the screening and quality assessment and were used to derive the default guideline values lists all chronic toxicity data used in the derivation. Equations for the adjustment of the DGVs based on DOC are provided in Appendix C: DOC correction for dissolved copper default guideline values.

# 1 Introduction

Copper is a naturally occurring metallic element. It is an abundant trace element, present in the Earth's crust at approximately 50 ppm (Landner & Reuther 2004). It is found as the native metal but predominantly in the form of: sulfide minerals chalcopyrite ( $\text{CuFeS}_2$ ), chalcocite ( $\text{Cu}_2\text{S}$ ) and bornite ( $\text{Cu}_5\text{FeS}_4$ ); copper carbonates azurite ( $\text{Cu}_3(\text{CO}_3)_2(\text{OH})_2$ ) and malachite ( $\text{Cu}_2\text{CO}_3(\text{OH})_2$ ); and the  $\text{Cu}^+$  oxide mineral cuprite ( $\text{Cu}_2\text{O}$ ) (Stumm & Morgan 1996, European Copper Institute 2008). The major copper mines are in the Americas (particularly Chile), Australia and Europe (particularly Russia, Poland and Sweden) (Landner & Reuther 2004, European Copper Institute 2008).

Copper has been used for centuries in jewellery, vessels, currency and tools (Landner & Reuther 2004). Current uses include architectural structures, roofing, plumbing, electrical wiring and in electronics (European Copper Institute 2008). Copper is also used as a biocide in antifouling paints, wood preservatives, fungicides and herbicides (European Copper Institute 2008). One of the major anthropogenic sources of copper in marine environments is antifouling paints, as  $\text{Cu}^+$  oxide is the most commonly used biocide in these paints internationally (Turner 2010, Ytreberg et al. 2010) and in Australia and New Zealand (Gadd et al. 2011). Measurements in water in marinas indicate dissolved copper concentrations up to 20  $\mu\text{g}/\text{L}$  (Gadd & Cameron 2012). Other sources of copper include municipal wastewater discharges, mining and mineral processing, metal and electrical manufacturing, and stormwater (from copper in architectural structures and vehicle brake linings (Timperley et al. 2005, Kennedy & Sutherland 2008)).

Copper is a D-block element and a transition metal—as such, it has more than one oxidation state (Stumm & Morgan 1996, European Copper Institute 2008). The principal states are cuprous ( $\text{Cu}^+$ ) and cupric ( $\text{Cu}^{2+}$ ) (Stumm & Morgan 1996). These are found as salts such as  $\text{Cu}^{2+}$  sulfate pentahydrate ( $\text{CuSO}_4 \cdot 7\text{H}_2\text{O}$ ) and  $\text{Cu}^+$  oxide ( $\text{Cu}_2\text{O}$ ) (Stumm & Morgan 1996).  $\text{Cu}^+$  is unstable in aqueous media and will oxidise to  $\text{Cu}^{2+}$ , which typically binds to inorganic and organic ligands, such as iron oxides or humic acids. In water, sediment and soil, the binding affinities of  $\text{Cu}^{2+}$  with inorganic and organic matter depend on pH, the oxidation-reduction potential in the local environment, and the presence of competing metal ions and inorganic anions. The solubility limit of copper is approximately 500  $\mu\text{g}/\text{L}$  in saline water (Krauskopf 1956).

Background concentrations of copper in marine water are low and can be difficult to measure accurately, and ultra-trace techniques and special precautions are required to avoid sample contamination. In the North Pacific Ocean, total copper was 0.03–0.1  $\mu\text{g}/\text{L}$  (Bruland 1980). In the Tasman Sea, dissolved copper concentrations were 0.030–0.036  $\mu\text{g}/\text{L}$  in surface water, increasing to ~0.2  $\mu\text{g}/\text{L}$  at depths of 3 000 m or more (Thompson et al. 2014). In most of these samples, the dissolved copper was more than 99% complexed by organic ligands (Thompson et al. 2014). Closer to shore, in measurements off the coast of New South Wales, Australia, dissolved copper was 0.025–0.036  $\mu\text{g}/\text{L}$  (Apte et al. 1998). In New Zealand coastal waters, total copper concentrations were 0.15–0.5  $\mu\text{g}/\text{L}$  in the outer Otago Harbour (Hunter & Tyler 1987) away from sources of pollution. Dissolved copper measured 0.38  $\mu\text{g}/\text{L}$  in Milford Sound (NIWA unpublished data) at locations influenced by marinas and vessel moorings. Total copper measured 1.5–1.8  $\mu\text{g}/\text{L}$  in Waitemata Harbour (Gadd & Cameron 2012), an area influenced by urban land use and vessels.

The ANZECC/ARMCANZ (2000) default guideline values (DGVs) for dissolved copper in marine water for 99%, 95%, 90% and 80% species protection were 0.3 µg/L, 1.3 µg/L, 3 µg/L and 8 µg/L, respectively. The DGVs were derived using the species sensitivity distribution method, from 70 chronic toxicity values for 26 species from five taxonomic groups (fish, crustaceans, molluscs, annelids and algae), and were considered high reliability values. The lowest toxicity value at the time was for the mollusc *Mytilus edulis* (0.4 µg/L, converted from 30 day EC50 for reproduction of 2 µg/L).

This technical brief provides updated DGVs for dissolved copper in marine water, which supersede the ANZECC/ARMCANZ (2000) DGVs. The current derivation has added new data published since 2000, including chronic data for Australasian species, and has added an adjustment based on dissolved organic carbon (DOC). The derivation has also drawn upon relevant aspects of copper risk assessments for the European Union (European Copper Institute 2008) and copper guidelines for the United Kingdom (Maycock et al. 2011) and the United States (USEPA 2016a). The DGV derivation process and the data included are described in Section 4.

## 2 Aquatic toxicology

Copper is an essential trace element for micro-organisms, plants and animals because it is a cofactor for numerous enzymes (IPCS 1998), and insufficient concentrations can lead to impaired metabolic functions and reduced growth. Copper deficiency has been observed in marine diatoms due to the naturally low concentrations in open oceans (Peers & Price 2006). Although marine organisms can regulate copper to some extent via excretion and/or storage in a detoxified form, when intake exceeds the capacity for regulation, copper is both acutely and chronically toxic (European Copper Institute 2008).

### 2.1 Mechanisms of toxicity

The mechanisms of copper toxicity in marine systems are reviewed in detail in the European Union Risk Assessment Report for copper (European Copper Institute 2008). In marine fish and invertebrates, copper toxicity is thought to be due to disruption of the osmo-regulatory and iono-regulatory systems and plasma ammonia metabolism (Grosell et al. 2003, Lewis et al. 2016). Chronic exposure to excess copper can result in alterations of brain function, enzyme activity, blood chemistry, and metabolism, which lead to adverse effects on growth, reproduction and survival. In microalgae, copper toxicity is thought to be due to reactive oxygen species, disruption of antioxidant defence mechanisms and consequent disruption of cell division (Stauber & Florence 1987, Adams et al. 2016, Jiang et al. 2016). As there are different mechanisms of copper toxicity between fish, invertebrates and plants, and algae, copper toxicity datasets have potential to exhibit bimodality or multimodality.

### 2.2 Toxicity

The acute toxicity of copper to marine species ranges over more than an order of magnitude based on North American data, from an EC50 of 17 µg/L for a mysid shrimp (*Holmesimysis costata*) to EC50 values of ~500 µg/L for several fish and some invertebrate species (USEPA 2003). However, EC50 values similar to or in excess of the copper solubility limit for marine water (500 µg/L) are not



reliable, as these would have included test concentrations where copper was not completely soluble. Toxicity values are typically lower for longer test durations, though chronic EC50 values are not frequently more than 10-fold lower than the acute EC50 values (European Copper Institute 2008, JRC-IHCP 2010). For example, for the saltwater cladoceran *Moina monogolica*, the 24 hour LC50 value was 154 µg/L (acute), compared to a 21 day EC50 of 20 µg/L for total reproduction (chronic) (Wang et al. 2007a).

NOEC values for chronic toxicity tests reviewed in the European Union copper risk assessment ranged from 3 µg/L to >3 000 µg/L (European Copper Institute 2008). Microalgal species (diatoms and green microalgae) were amongst the most sensitive species, with NOEC/EC10 values <10 µg/L reported for 48–72 hour tests (European Copper Institute 2008). These data are also included in the current derivation. Macroalgal growth and germination are also affected by copper, with NOEC values of <10–50 µg/L reported for 14–19 day tests (Anderson et al. 1990, Brooks et al. 2008).

The early life stages of molluscs, echinoderms and corals are generally much more sensitive than adult stages, and also when compared to annelids and crustaceans (Markich et al. 2002, Langdon et al. 2009). Development tests using mollusc embryos resulted in NOEC values of 6–10 µg/L for the pacific oyster (*Crassostrea gigas*) and mediterranean mussel (*Mytilus galloprovincialis*) in standard test waters (European Copper Institute 2008). However, there are some exceptions, for example, mussel larvae were less sensitive than adults when tested at DOC concentrations >2.5 mg/L (Deruytter et al. 2017). Several early life stage tests are included in the current derivation (see Section 4.2).

There are comparatively few chronic toxicity data for marine fish, with some studies reporting effects on juvenile fish growth (length and/or weight) at copper concentrations of ~100 µg/L (Wang et al. 2014, 2015). Studies of topsmelt (*Atherinops affinis*) suggest that larval development is a more sensitive endpoint than larval mortality or embryo development (Anderson et al. 1991, McNulty et al. 1994).

## 3 Factors affecting toxicity

### 3.1 Copper speciation

In saline water, dissolved copper is complexed by organic ligands (Stumm & Morgan 1996, Thompson et al. 2014, Sander et al. 2015). Inorganic species are dominated by  $\text{CuCO}_3$  and  $\text{Cu}(\text{CO}_3)_2^{2-}$ , and only a small fraction of copper is present in the free ionic form  $\text{Cu}^{2+}$  or as copper hydroxides ( $\text{CuOH}^+$  and  $\text{Cu}(\text{OH})_2$ ) or chlorides (e.g.  $\text{CuCl}_4^{2-}$ ) (Grosell 2011). The ionic  $\text{Cu}^{2+}$  form is generally acknowledged as the primary toxic form, and hydroxide forms may also be toxic, whereas carbonate complexes are typically not toxic (Hunt 1987) (NB studies examining this have been undertaken on freshwater species).

Organic matter has a greater influence on copper speciation than the pH and salinity of marine water. The pH is important for determining the complexation capacity of dissolved organic matter (DOM; typically and hereafter referred to as DOC, as it contains ~50% carbon by mass (Duarte et al. 2016)) and for determining the speciation of carbonate complexes, all of which influence metal speciation (Stumm & Morgan 1996, USEPA 2016a). In estuarine water, with its lower salinity and pH,



the concentrations of  $\text{Cu}^{2+}$  may be higher than in ocean water (Millero et al. 2009). The partitioning of copper to suspended particles is also a major removal route for copper (Stumm & Morgan 1996), and conditions need to become acidic (e.g. less than pH 6) before significant proportions of copper will desorb from particles and dissociate inorganic and organic complexes.

### 3.2 Toxicity modifying factors

The aquatic toxicity of copper is negatively correlated with the concentration of DOC (Arnold et al. 2006, Nadella et al. 2009, DePlama et al. 2011, Deruytter et al. 2015). For example, in the mussel *Mytilus* sp., the EC50 based on 48 hour embryo-larval development increased (i.e. toxicity was reduced) by a factor of approximately 5 as DOC concentration increased from 1 mg/L to 9 mg/L (i.e. EC50 ~12 µg/L at DOC 1–1.2 mg/L to EC50 60 µg/L at DOC 9 mg/L) (Arnold et al. 2006). The effect was similar for the estuarine copepod *Eurytemora affinis*, where the LC50 concentrations increased from 76 µg/L to 170 µg/L as DOC increased from 2 mg/L to 8 mg/L (Hall et al. 2008). The influence of DOC on copper toxicity has been reported for multiple taxonomic groups, including:

- molluscs such as mussels *Mytilus galloprovincialis* (Arnold et al. 2010a, Rosen et al. 2008, Zitoun et al. 2019) and *M. trossulus* (Nadella et al. 2009) and oysters *Crassostrea virginica* (Arnold et al. 2010) and *C. gigas* (Brooks et al. 2007)
- echinoderms such as the sand dollar *Dendraster excentricus* and the sea urchin *Strongylocentrotus purpuratus* (Arnold et al. 2010a, Rosen et al. 2008)
- the rotifer *Brachionus plicatilis* (Arnold et al. 2010b)
- macroalgae such as the brown alga *Fucus vesiculosus* (Brooks et al. 2008).

The concentration of DOC is not the only factor that affects metal binding (and hence toxicity)—the composition of DOC is also important (Baken et al. 2011, Mueller et al. 2012, Pearson et al. 2017). The complexation (binding) capacity of DOC depends on the prevalence of metal binding functional groups, including hydroxyl, aromatic, phenolic, amino and thiol groups (Smith et al. 2002). The prevalence of such groups depends on the DOC source (i.e. terrestrial or aquatic) and whether it comes from natural sources (e.g. leaf litter breakdown) or anthropogenic sources (e.g. wastewater treatment plant discharges). Generally, copper binding is highest in DOC from terrestrial sources that have a prevalence of humic-like components with high aromaticity (Chen et al. 2018). This is an area of ongoing research, and future models of DOC and toxicity may incorporate DOC quality as well as quantity.

The effects of salinity on copper toxicity are less clear. In the estuarine copepod *E. affinis*, salinity influenced toxicity within the range 2.5–25‰, with toxicity being higher at the higher salinity (Millero et al. 2009). Conversely, the estuarine fish *Fundulus heteroclitus* was most sensitive to copper in freshwater (96 hour LC50 value of 18 µg/L), followed by copper in high salinity water (35‰, LC50 value of 294 µg/L), and least sensitive to copper in intermediate salinity water (10‰, LC50 value of >963 µg/L) (Grosell et al. 2007). A slightly different pattern was shown for larval topsmelt (*A. affinis*), with copper toxicity decreasing as salinity increased (LC50s: 44 µg/L at 10‰; 72 µg/L at 17‰; 134 µg/L at 25‰; and 205 µg/L at 34‰) (Anderson et al. 1995). However, in other studies, including those on the estuarine rotifer *B. plicatilis*, salinity (and pH) had no effect on copper toxicity (Arnold et al. 2010b). Overall, the studies suggest that salinity affects copper speciation and, therefore, toxicity,

but these effects are non-linear and are species-dependent. Consequently, it is difficult to incorporate corrections for salinity into DGVs.

As water temperature affects metabolism and speciation, it could be expected to influence metal toxicity and lead to higher toxicity at higher temperatures. However, there are few studies that show this conclusively. For the protist *Euplotes crassus*, copper toxicity increased as water temperature increased (Gomiero & Viarengo 2014). However, for the tropical brittle star *Amphipholis squamata* (Delle Chiaje 1828), toxicity was higher at a lower temperature (15°C) than at a higher temperature (25°C) (Black et al. 2015). The effects of temperature on copper toxicity are likely dependent on each species' thermal tolerance range.

### 3.3 Biotic ligand model

In response to these potential influences on copper toxicity, the Biotic Ligand Model (BLM), initially developed for freshwater (Di Toro et al. 2001, Santore et al. 2002), has been extended to predicting copper toxicity in marine water. The BLM is a metal bioavailability model that simulates speciation as an equilibrium system that includes the complexation of inorganic ions and DOC (Chadwick et al. 2008, USEPA 2016a). The BLM also includes reactions that describe the chemical interactions of copper and other cations with physiologically active sites, termed biotic ligands (BLs), which correspond to the proximate site of action of toxicity in aquatic organisms, such as fish gills. USEPA (2016a) uses a BLM to calculate a water quality criterion (WQC) at site-specific concentrations of DOC, pH, salinity and temperature. Of these four factors, DOC has the most influence in determining the WQC, and has an approximately 1:1 relationship, whereby the WQC approximately doubles as DOC doubles. By comparison, salinity and pH have minor effects (11% difference in the WQC between salinity of 10‰ and 30‰, at pH 8.1; and 4% difference in the WQC between pH 7.5 and 8.5, at salinity of 30‰) and temperature has a negligible effect (0.1% difference in the WQC between 5°C and 30°C, based on pH 8.1 and salinity 30‰).

## 4 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. Although some decisions on data selection/manipulation may reflect the Warne et al. (2015) method rather than the Warne et al. (2018) method, these were found to have no material effect on the final DGVs.

### 4.1 Collation of toxicity data

Toxicity data were collated from the ECOTOX database (USEPA 2016b), the ANZECC/ARMCANZ (2000) water quality guidelines, compilations of Australasian toxicity data (Markich et al. 2002, Langdon et al. 2009), and from the European Union risk assessment (European Copper Institute 2008) and the United Kingdom marine copper guideline derivation (Maycock et al. 2011). These were supplemented by searches using the journal abstracting service 'Web of Science' for studies published during 2015–2016 and not included in the ECOTOX database, and internet searches for Australian and New Zealand toxicity data contained within grey literature, theses or unpublished reports.

The toxicity dataset was restricted to chronic studies (following details in Warne et al. (2018)). Data were only included for studies that had measured the copper concentrations either in the test solutions, or in the stock solutions used to produce the test solutions, provided a clear concentration–response relationship was observed or stated. Although some studies reported concentrations as total copper, all copper was assumed to be in dissolved form in the test solutions given that laboratory toxicity test solutions typically have low particulate concentrations; therefore, the DGVs are representative of dissolved copper concentrations. Data were restricted to tests with salinity  $\geq 25\text{‰}$  and  $\leq 36\text{‰}$  following the guidance from Warne et al. (2018), although studies where the salinity at times exceeded 36‰ were included where the midpoint of the range was approximately 35‰. Antarctic species and any other testing conducted at  $< 1^\circ\text{C}$  were not included in the derivation. Because DOC may affect the toxicity of copper, the data were restricted to studies with DOC  $< 2$  mg/L. If a study did not report DOC, it was assumed to have low DOC, as would be the case for standard laboratory test control/diluent waters, and the study was included in the derivation.

These data exclusion rules resulted in several Australasian species not being included in the current derivation despite the existence of chronic data (see Appendix D: Chronic toxicity data for Australasian species not used to derive the default guideline values); this was predominantly due to not measuring the copper in the test medium or stock solutions. Of these species, the most sensitive were:

- the diatom *Thalassiosira weissflogii* (NOEC 0.08  $\mu\text{g/L}$  for a population test (Karner et al. 2006))
- the blue mussel *M. edulis* (NOEC 0.4  $\mu\text{g/L}$ , converted from an EC50 of 2  $\mu\text{g/L}$ , for a reproduction test (Stromgren & Nielsen 1991))
- the annelid *Galeolaria caespitose* (NOEC 0.9  $\mu\text{g/L}$ , converted from an EC50 of 4.6  $\mu\text{g/L}$ , for a development test (Ross & Bidwell 2001))
- the Pacific oyster *C. gigas* (NOEC  $\sim 1$   $\mu\text{g/L}$  for development tests (Martin et al. 1981, Worboys et al. 2002)).

The mussel, annelid and oyster species are expected to be protected by the DGVs for 95% species protection, and only the annelid and oyster are expected to be protected at the 90% species protection. The toxicity data suggest that the annelid and oyster would not be protected at the 80% species protection. The toxicity data suggest the diatom *T. weissflogii* would not be protected at any species protection level; the sensitivity of this species should be confirmed by further toxicity testing that incorporates the measurement of copper in the test solutions.

## 4.2 Toxicity data used in derivation

Data sourced from the ANZECC/ARMCANZ (2000) guidelines, the compiled Australasian toxicity datasets (Markich et al. 2002, Langdon et al. 2009), the European Union risk assessment (European Copper Institute 2008) and the United Kingdom guidelines (Maycock et al. 2011) were considered to have already been assessed for quality and considered to be acceptable, as recommended by Warne et al. (2018). All other data were quality assessed as per Warne et al. (2018) to ensure their suitability for inclusion.

Warne et al. (2018) advise that NOEC or LOEC data should not be used where there are preferred data (e.g. chronic no effect concentration (NEC), EC10, IC10) for  $\geq 8$  species that belong to  $\geq 4$  taxonomic groups, but that the exclusion of NOEC/LOEC data does not compromise the reliability and protectiveness of the DGVs. Because of the large pool of data for copper toxicity to marine organisms, only preferred data were included in an initial derivation. This resulted in data for 15 species, which is sufficient to derive DGVs using the species sensitivity distribution (SSD) (Warne et al. 2018). Based on the use of chronic data, the number of species included (15 species; classified as 'preferred') and the good fit of the model to the data (see SSD in Appendix E: Guideline value derivation with preferred toxicity estimates only), DGVs based on these data alone would have very high reliability. However, inclusion of only preferred data excludes several sensitive unicellular algae for which only NOEC data were available. The omission of NOEC data from the toxicity dataset resulted in higher DGVs. Therefore, additional data were included in the DGV derivation to ensure protection of sensitive species.

Specifically, the preferred data (NEC and EC10–EC20 data) were supplemented with NOEC data for species found in Australia or New Zealand. For four species, the NOEC data were reported as < values; these can be used in DGV derivation (Warne et al. 2018) and were treated and included as described below.

- For the diatom *Minutocellus polymorphus*, a value of 0.2  $\mu\text{g/L}$  was used, based on a reported LOEC of 0.2  $\mu\text{g/L}$  and a NOEC of <0.2  $\mu\text{g/L}$  (Levy et al. 2007). A more conservative approach of dividing the LOEC value of 0.2  $\mu\text{g/L}$  by the default conversion factor of 2.5 for a LOEC to a negligible effect equivalent was not justified on the basis that:
  - it would have resulted in a concentration close to background concentrations of copper
  - the effect size at the LOEC was only 16%, which is within the 20% effect size accepted for DGV derivation (i.e. up to EC20; Warne et al. (2018)).

Notably, the *M. polymorphus* toxicity value of 0.2  $\mu\text{g/L}$  was the lowest toxicity value in the dataset, and the apparent high sensitivity of this species was supported by a subsequent study that reported an IC50 of 1.0  $\mu\text{g/L}$  (Golding et al. 2015), similar to the IC50 of 0.6  $\mu\text{g/L}$  reported by Levy et al. (2007).

- For the brown microalga *Proteomonas sulcata*, where the NOEC was reported as <5  $\mu\text{g/L}$  (Levy et al. 2007), the IC50 value was 4.2  $\mu\text{g/L}$ . No LOEC was reported as it was greater than the IC50 (Levy et al. 2007). Therefore, it was more conservative to use the IC50 divided by 5 to estimate a negligible effect equivalent of 0.84  $\mu\text{g/L}$ .
- For the annelid *Hydroides elegans*, where the NOEC was reported as <6.2  $\mu\text{g/L}$ , the LC50 value was 52  $\mu\text{g/L}$  (Xie et al. 2005), which when converted to a negligible effect equivalent by dividing by 5 would have resulted in 10  $\mu\text{g/L}$ , a value larger than the reported NOEC. In this case, it was more conservative to use the LOEC value (6.2  $\mu\text{g/L}$ ) converted to a NOEC by dividing by 2.5 (2.5  $\mu\text{g/L}$ ).
- For the brown macroalga *Macrocystis pyrifera*, a 20 day NOEC of <10.2  $\mu\text{g/L}$  was reported for sporophyte production (Anderson et al. 1990). The effect size at this concentration was close to 50%, over the effect size accepted for DGV derivation without conversion (i.e. up to EC20; Warne et al. 2018). Therefore, this value was considered a LOEC and converted to a negligible effect equivalent of 4.1  $\mu\text{g/L}$  by dividing by 2.5. This value is supported by a 48 hour test, with 100% effect on sporophyte production reported at the lowest test concentration of 18  $\mu\text{g/L}$  (Anderson et al. 1990).

As there were no preferred data for any fish species, NOEC data were accepted for fish to ensure that this taxonomic group was included in the DGV derivation.

There were 63 chronic toxicity values assessed as being of suitable quality for use in the DGV derivation (Appendix B: Toxicity data that passed the screening and quality assessment and were used to derive the default guideline values). Of these, four were of the highest preference (NEC) from one species, 33 were of the next highest preference (EC/LC10–20) from 14 species, 24 were NOEC values from 15 species, and there was one converted IC50 for one species, and one converted LOEC for one species. The salinity range for the acceptable toxicity tests was 25–37‰, with the maximum being slightly higher than Warne et al. (2018) advise. Only one study reported a range that exceeded 36‰: tests on the giant kelp *M. pyrifera*, with salinity range reported as 35–37‰ (Anderson et al. 1990). Based on the midpoint of the salinity ranges being  $\leq 36$ ‰, the toxicity values from these tests were included in the derivation. The temperature range for the acceptable toxicity tests was 13–31°C; thus, the dataset included both temperate and (sub)tropical species. DOC was reported for 19 toxicity values representing 10 species, and ranged from <0.001 mg/L to 2 mg/L. For tests that did not report DOC, it was assumed to be <2 mg/L.

The 63 acceptable toxicity values (Appendix B: Toxicity data that passed the screening and quality assessment and were used to derive the default guideline values) were summarised to 32 single species values (Table 1) for use in the SSD, by either calculating geometric means or selecting the value for the most sensitive endpoint, life-stage and toxicity test duration for each species, based on Warne et al. (2018). The DGV derivation used data from 32 species from 12 taxonomic groups. The toxicity values in the SSD range over 3 orders of magnitude. The lowest values were 0.2 µg/L for the diatom *M. polymorphus* and 0.3 µg/L for the green microalga *Micromonas pusilla* (Levy et al. 2007). The most sensitive fauna species was the New Zealand paua (mollusc) *Haliotis iris*, at 0.7 µg/L (Rouchon 2015). The least sensitive species was a blue–green alga *Cyanobium* sp., at 300 µg/L (Alquezar & Anastasi 2013). Details of the data quality assessment and all the data that were deemed to have passed the quality assessment are provided as supporting information.

The ANZECC/ARMCANZ (2000) DGVs for copper in marine water were based on 25 species, seven fewer than the current derivation. This is the result of substantial new data becoming available in the past 20 years. Data for 27 of the 32 included species are from studies published since 2003. Several species that were in the ANZECC/ARMCANZ (2000) derivation are not included in the current derivation, primarily due to the:

- new classifications for chronic toxicity tests, which require longer test durations than in the ANZECC/ARMCANZ (2000) derivation (e.g. adult fish tests now require  $\geq 21$  days (Warne et al. 2018) compared to  $\geq 7$  days (ANZECC/ARMCANZ 2000))
- preference for EC10/NOEC data over converted EC50 data
- the avoidance of data reported before 1980.

As the different mechanisms of copper toxicity suggest the potential to exhibit bimodality or multimodality, the toxicity dataset was assessed for this following the advice in Warne et al. (2018). There was some visual evidence of bimodality but little evidence from statistical tests and limited evidence of taxa-specific sensitivity, with the possible exception of diatoms (Appendix F: Modality assessment for copper). It is considered that the apparent visual bimodality is simply an artefact of

the data included and the use of NOEC data in the distribution. Consequently, all the acceptable data were used in the DGV derivation.

**Table 1 Summary of single chronic toxicity values, all species used to derive the default guideline values for dissolved copper in marine water**

Taxonomic group	Species	Life stage	Exposure duration (d)	Toxicity measure (test endpoint)	Reported toxicity value (µg/L)	Final toxicity value (µg/L)
Diatom	<i>Entomoneis punctulata</i>	Exponential growth	3	IC10 (Population growth rate)	1.4	1.4
	<i>Minutocellus polymorphus</i>	Exponential growth	3	NOEC (Population growth rate)	<0.2	0.2 <sup>a</sup>
	<i>Ceratoneis closterium</i>	Exponential growth	3	EC10 (Population growth rate)	1.7	1.7
	<i>Phaeodactylum tricoratum</i>	Exponential growth	3	IC20 (Population growth rate)	0.7	0.7
Brown microalga	<i>Proteomonas sulcata</i>	Exponential growth	3	IC50 (Population growth rate)	4.2	0.84 <sup>b</sup>
	<i>Coccolithus huxleyi</i>	Exponential growth	3	NOEC (Population growth rate)	8	8
	<i>Gephyrocapsa oceanica</i>	Exponential growth	3	NOEC (Population growth rate)	1.3	1.3
	<i>Isochrysis galbana</i>	Exponential growth	3	EC10 (Population growth rate)	2.6	2.6
Blue-green alga	<i>Cyanobium</i> sp.	Exponential growth	3	EC10 (Population abundance)	300	300
Green microalga	<i>Dunaliella tertiolecta</i>	Exponential growth	3	NOEC (Population growth rate)	8	8
	<i>Micromonas pusilla</i>	Exponential growth	3	NOEC (Population growth rate)	0.3	0.3
	<i>Tetraselmis</i> sp.	Exponential growth	3	NOEC (Population growth rate)	7	7
Green macroalga	<i>Ulva fasciata</i>	Zoospore	4	NOEC (Reproduction)	27	27
	<i>Ulva lactuca</i>	Zoospore	3	EC10 (settlement and growth inhibition)	56	56
Brown macroalga	<i>Macrocystis pyrifera</i>	Zoospore	20	LOEC (Sporophyte production)	10.2	4.1 <sup>c</sup>



Taxonomic group	Species	Life stage	Exposure duration (d)	Toxicity measure (test endpoint)	Reported toxicity value (µg/L)	Final toxicity value (µg/L)
	<i>Fucus vesiculosus</i>	Germiling	14	NEC (Growth)	17	17
Cnidarian	<i>Acropora tenuis</i>	Gamete	0.2	NOEC (Fertilisation)	34	34
	<i>Coelastrea aspera</i>	Gamete	0.2	NOEC (Fertilisation)	13	13
	<i>Platygyra daedalea</i>	Gamete	0.2	EC10 (Fertilisation)	1.4	1.4
	<i>Exaiptasia diaphana</i> <sup>d</sup>	Adult	28	EC10 (Reproduction)	9.8	9.8
Echinoderm	<i>Diadema savignyi</i>	Embryo	2	NOEC (Development)	9.6	9.6
	<i>Evechinus chloroticus</i>	Larva	3	EC10 (Development)	2.1	2.1
Annelid	<i>Hydroides elegans</i>	Gamete	8	LOEC (Metamorphosis)	6.2	2.5 <sup>c</sup>
Crustacean	<i>Tisbe furcata</i>	Life cycle	100	NOEC (Survival and reproduction)	19	19
Mollusc	<i>Amphibalanus amphitrite</i>	Larva	4	EC10 (Metamorphosis)	28	28
	<i>Haliotis iris</i>	Larva	3	EC10 (Development)	0.7	0.7
	<i>Mimachlamys asperrima</i>	Larva	2	NOEC (Development)	2	2
	<i>Mytilus galloprovincialis</i>	Embryo	2	EC10 (Development)	4.8	4.8
	<i>Mytilus trossulus</i>	Embryo	2	EC20 (Development)	5.6	5.6
	<i>Saccostrea glomerata</i>	Larva	14	LC10 (Mortality)	70	70
Fish	<i>Atherinops affinis</i>	Larva	12	NOEC (Development)	62	62
	<i>Epinephelus coioides</i>	Juvenile	25	NOEC (Growth)	21	21

<sup>a</sup> Less than (<) value; actual value used in SSD (Warne et al. 2018).

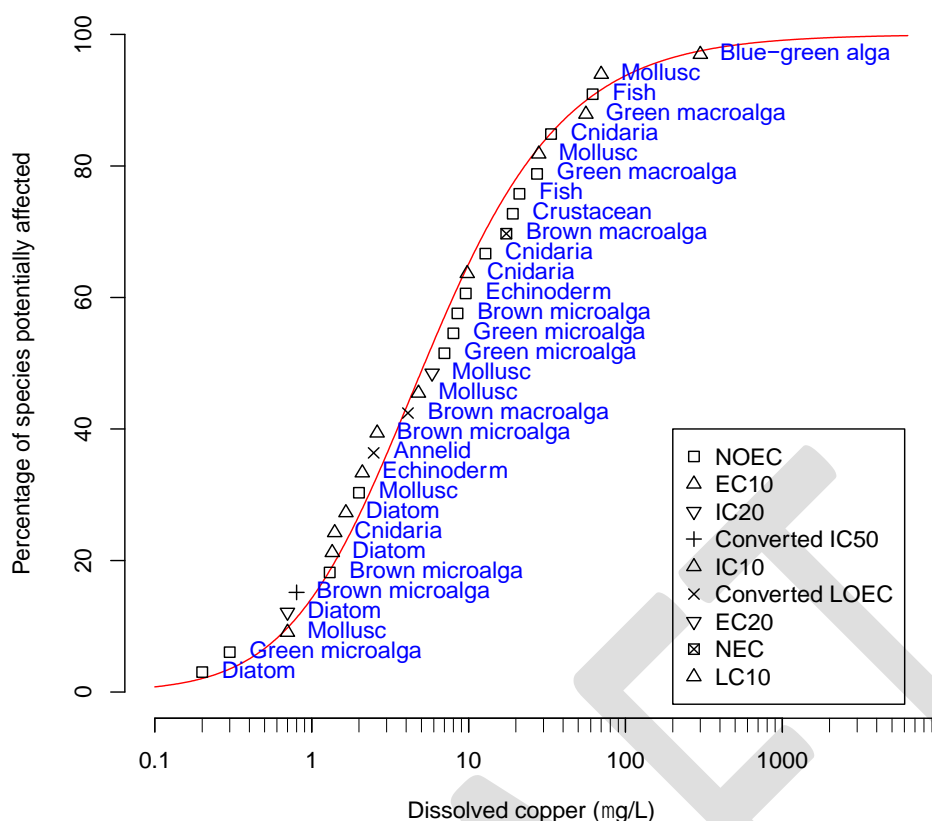
<sup>b</sup> Chronic IC50 converted to chronic NOEC/EC10 value by dividing by 5 (Warne et al. 2018).

<sup>c</sup> Chronic LOEC value converted to chronic NOEC/EC10 value by dividing by 2.5 (Warne et al. 2018).

<sup>d</sup> Formerly *Aiptasia pulchella* and *Exaiptasia pallida*.

### 4.3 Species sensitivity distribution

The cumulative frequency (species sensitivity) distribution (SSD) of the 32 marine copper chronic toxicity values reported in Table 1 is shown in Figure 1. The SSD was plotted using the Burrlioz 2.0 software. The model provided a good fit to the data, including the most sensitive species (Figure 1).



**Figure 1 Species sensitivity distribution, dissolved copper in marine water**

#### 4.4 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 (ANZG 2018).

The DGVs for 99%, 95%, 90% and 80% species protection are shown in Table 2. The DGVs apply to water with:

- DOC ≤0.5 mg/L
- salinity 25–36‰
- pH 6.5–8.0.

For marine water outside this range of salinity and pH, site-specific factors affecting toxicity should be considered, including modelling of metal speciation (see Appendix A: Actions to assess the bioavailable fraction of a metal).

**Table 2 Toxicant default guideline values, dissolved copper in marine water, very high reliability**

Level of species protection (%)	DGV for copper in marine water (µg/L) <sup>a, b</sup>
99	0.12
95	0.40
90	0.72
80	1.4

**a** The DGVs apply when DOC  $\leq 0.5$  mg/L. When DOC is  $>0.5$  mg/L, the DGVs should be adjusted based on the values in Table 3 or the equations in Appendix C: DOC correction for dissolved copper default guideline values; however, no further adjustments should be made when DOC  $>6$  mg/L.

**b** The DGVs were derived using the Burrlioz 2.0 software and rounded to two significant figures.

The DGVs can be adjusted for different DOC concentrations to produce DOC-modified DGVs (Table 3). Equations and tables for deriving DGVs at other DOC concentrations are provided in Appendix C: DOC correction for dissolved copper default guideline values, and the rationale for this is provided in Appendix G: Rationale for DOC correction. The adjustment is based on a linear slope of 1.24, obtained from a linear regression of the copper marine WQC USEPA (2016a) with varying DOC as modelled by its BLM, based on DOC from 0.5 mg/L to 6 mg/L. These DOC concentrations are expected to be representative of most New Zealand and Australian coastal marine waters. There is no adjustment above 6 mg/L DOC, irrespective of whether DOC is higher than this. For DOC concentrations well above 6 mg/L, either the adjustment to 6 mg/L should be used, or site-specific studies undertaken.

The USEPA (2016a) BLM and the chemical speciation modelling within it were based on copper complexometric titrations. Although this should in theory be applicable to all species, it appears that the protective effect of DOC does not apply to settled mussels, potentially due to DOC-Cu complexes being bioavailable to these mussels, or becoming bioavailable within the gill microenvironment (Deruytter et al. 2017). The reported 5% effect concentration (EC5) for blue mussels (*M. edulis*) is 8.8  $\mu\text{g/L}$  for clearance rate (Deruytter et al. 2017). Therefore, this species should be protected by all of the DGVs for 95% species protection up to the maximum DGV of 7.2  $\mu\text{g/L}$  for  $\geq 6$  mg/L DOC (Table 3).

**Table 3 Default guideline values for 95% species protection, dissolved copper in marine water with different dissolved organic carbon**

DOC (mg/L)	Dissolved copper DOC-modified guideline value at 95% species protection ( $\mu\text{g/L}$ )
0.5	0.40
1	1.0
2	2.3
4	4.7
$\geq 6$	7.2

Shading indicates the DGV for 95% species protection in the absence of information on DOC concentration.

## 4.5 Reliability classification

The dissolved copper marine water DGVs have a very high reliability classification (Warne et al. 2018) based on the outcomes for the following three criteria:

- Sample size—32 (preferred)
- Type of toxicity data—chronic
- SSD model fit—good (Burr type III model).

# Glossary

Term	Definition
acute toxicity	A lethal or adverse sub-lethal effect that occurs after exposure to a chemical for a short period relative to the organism's life span. Refer to Warne et al. [67] for examples of acute exposures.
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.
BLM	Biotic ligand model
chronic toxicity	A lethal or sub-lethal 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'.
DOC	Dissolved organic carbon.
DOM	Dissolved organic matter.
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.
ECx	The concentration of a substance in water or sediment that is estimated to produce an x% change in the response being measured or a certain effect in x% of the test organisms, 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 and site-specific guideline value.)
HCS (5% hazardous concentration)	The concentration that may result in effects to 5% of species. It is equivalent to the concentration that will protect 95% of species.
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.
ICx	The concentration of a substance in water or sediment that is estimated to produce an x% inhibition of the response being measured in test organisms relative to the control response, under specified conditions.
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.
LCx	The concentration of a substance in water or sediment that is estimated to be lethal to x% of a group of test organisms under specified conditions.
lowest observed effect concentration (LOEC)	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.
no effect concentration (NEC)	The maximum concentration of a toxicant that causes no adverse effect in a target organism, based on a threshold parameter in a concentration–response model.

<b>Term</b>	<b>Definition</b>
no observed effect concentration (NOEC)	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.
predicted no effect concentration (PNEC)	The concentration of a chemical that marks the limit at which below no adverse effects of exposure in an ecosystem are measured.
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.
speciation	The intimate chemical environment of the indicator, that is the compound or ion of which it forms a part.
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 (chemical)	Most commonly used for metals, chemical species are different forms of a particular chemical that may include different oxidation states, isotopes, complexes with organic ligands (in the case of metals) or with particulate matter.
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: Actions to assess the bioavailable fraction of a metal

A decision tree for evaluating monitoring data against default guideline values (DGVs) or site-specific guideline values for dissolved copper in marine water, which includes consideration of the bioavailable fraction, is shown in Figure A 1. The outcomes of the process shown here for water chemistry assessment should be used with other lines of evidence (e.g. biodiversity assessment or direct toxicity assessment) in a weight of evidence approach to assess overall water quality.

With respect to the modelling of bioavailable copper, it is necessary to consider simple ionic complexes; however, it is also known that there are colloidal forms and weak ionic complexes that can dissociate and cross biological membranes. Approaches such as the biotic ligand model (BLM) for copper in marine water (USEPA 2016a) are appropriate modelling options. Alternatively, speciation modelling is also an option, for example, the Windermere Humic Aqueous Model (WHAM7), which includes a solution speciation model as well as sub-models for ion binding to humic and fulvic acids, clay and oxides of iron, aluminium, manganese and silica (UKCEH 2021). Bioavailable copper can be measured using a range of techniques designed to measure the ‘labile’ fraction of metals that has been shown to correlate with the fraction that is biologically available (see Batley et al. 2004). Currently, the use of Chelex columns and diffusive gradients in thin films (DGT) are the most widely used approaches.

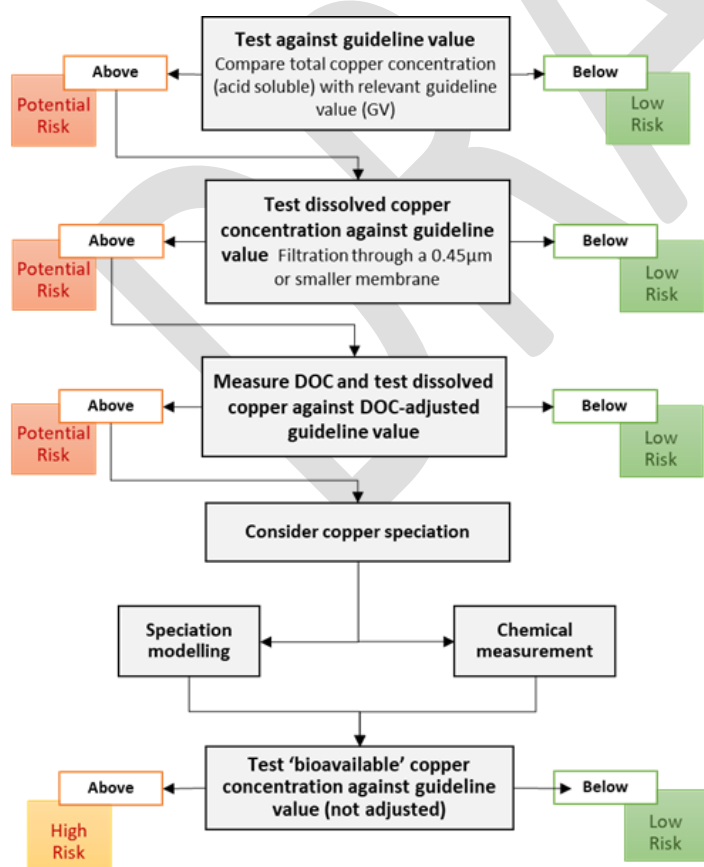


Figure A 1 Actions to assess bioavailable fraction of copper in marine water



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

**Table B 1 Summary, toxicity data that passed the screening and quality assurance processes, dissolved copper in marine water**

Taxonomic group	Species	Life stage	Exposure duration (d)	Toxicity measure (test endpoint)	Salinity (‰)	DOC (mg/L)	Temperature (°C)	Concentration (µg/L)	Reference
Diatom	<i>Entomoneis punctulata</i>	Exponential growth	3	IC10 (Population growth rate)	34	1	21	1.4	Stauber et al. (2008)
–	–	–	–	–	–	–	–	<b>1.4</b>	<b>Value used in SSD</b>
–	<i>Minutocellus polymorphus</i>	Exponential growth	3	NOEC (Population growth rate)	–	–	21	<0.2	Levy et al. (2007)
–	–	–	–	–	–	–	–	<b>0.2<sup>a</sup></b>	<b>Value used in SSD</b>
–	<i>Ceratoneis closterium</i> <sup>d</sup>	Exponential growth	3	IC10 (Population growth rate)	34	–	27	8	Johnson et al. (2007)
–	–	Exponential growth	3	EC10 (Population growth rate)	36	1.4	27	0.97	Adams et al. (2018)
–	–	Exponential growth	3	EC10 (Population growth rate)	36	1.4	27	0.87	Adams et al. (2018)
–	–	Exponential growth	3	EC10 (Population growth rate)	36	1.4	21	1.1	Adams et al. (2018)
–	–	–	–	–	–	–	–	<b>1.7</b>	<b>Value used in SSD (geometric mean)</b>
–	<i>Phaeodactylum tricornutum</i>	Exponential growth	3	IC20 (Population growth rate)	–	–	21	0.7	Angel et al. (2015)
–	–	–	–	–	–	–	–	<b>0.7</b>	<b>Value used in SSD</b>

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Taxonomic group	Species	Life stage	Exposure duration (d)	Toxicity measure (test endpoint)	Salinity (‰)	DOC (mg/L)	Temperature (°C)	Concentration (µg/L)	Reference
Brown microalga	<i>Coccolithus huxleyi</i>	Exponential growth	3	NOEC (Population growth rate)	–	–	21	8	Levy et al. (2007)
	–	–	–	–	–	–	–	<b>8</b>	<b>Value used in SSD</b>
	<i>Gephyrocapsa oceanica</i>	Exponential growth	3	NOEC (Population growth rate)	–	–	21	1.3	Levy et al. (2007)
	–	–	–	–	–	–	–	<b>1.3</b>	<b>Value used in SSD</b>
	<i>Isochrysis galbana</i>	Exponential growth	3	EC10 (Population growth rate)	35	–	28	3.1	Trenfield et al. (2015)
–	–	Exponential growth	3	EC10 (Population growth rate)	36	–	28	2.2	Adams et al. (2018)
	–	–	–	–	–	–	–	<b>2.6</b>	<b>Value used in SSD (geometric mean)</b>
	<i>Proteomonas sulcata</i>	Exponential growth	3	IC50 (Population growth rate)	–	–	27	4.2	Levy et al. (2007)
–	–	–	–	–	–	–	<b>0.84<sup>b</sup></b>	<b>Value used in SSD</b>	
Blue-green alga	<i>Cyanobium</i> sp.	Exponential growth	3	EC10 (Population abundance)	–	–	25	300	Alquezar & Anastasi (2013)
	–	–	–	–	–	–	–	<b>300</b>	<b>Value used in SSD</b>
Green microalga	<i>Dunaliella tertiolecta</i>	Exponential growth	3	NOEC (Population growth rate)	–	–	21	8	Levy et al. (2008)
	–	Exponential growth	3	NOEC (Population growth rate)	–	–	21	8	Levy et al. (2007)
	–	–	–	–	–	–	–	<b>8</b>	<b>Value used in SSD (geometric mean)</b>
	<i>Micromonas pusilla</i>	Exponential growth	3	NOEC (Population growth rate)	–	–	21	0.3	Levy et al. (2007)
–	–	–	–	–	–	–	<b>0.3</b>	<b>Value used in SSD</b>	

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Taxonomic group	Species	Life stage	Exposure duration (d)	Toxicity measure (test endpoint)	Salinity (‰)	DOC (mg/L)	Temperature (°C)	Concentration (µg/L)	Reference
	<i>Tetraselmis</i> sp.	Exponential growth	3	NOEC (Population growth rate)	–	–	21	7	Levy et al. (2008)
		Exponential growth	3	NOEC (Population growth rate)	–	–	21	7	Levy et al. (2007)
	–							<b>7</b>	<b>Value used in SSD (geometric mean)</b>
Green macroalga	<i>Ulva fasciata</i>	Zoospore	4	NOEC (Reproduction)	–	–	20	27	Hooten & Carr (1998)
	–							<b>27</b>	<b>Value used in SSD</b>
	<i>Ulva lactuca</i>	Zoospore	3	EC10 (Settlement and growth inhibition)	–	–	15	56	Wendt et al. (2013)
	–							<b>56</b>	<b>Value used in SSD</b>
Brown macroalga	<i>Macrocystis pyrifera</i>	Zoospore	20	LOEC (Sporophyte production)	35–37	–	13–15	10.2	Anderson et al. (1990)
	–							<b>4.1<sup>c</sup></b>	<b>Value used in SSD</b>
	<i>Fucus vesiculosus</i>	Germling	14	NEC (Growth)	31	<0.001	22	11	Brooks et al. (2008)
		Germling	14	NEC (Growth)	31	0.009	22	14	Brooks et al. (2008)
		Germling	14	NEC (Growth)	31	0.55	22	19	Brooks et al. (2008)
		Germling	14	NEC (Growth)	31	1.65	22	32	Brooks et al. (2008)
	–							<b>17</b>	<b>Value used in SSD (geometric mean)</b>

Toxicant default guideline values for aquatic ecosystem protection: Dissolved copper in marine water

Taxonomic group	Species	Life stage	Exposure duration (d)	Toxicity measure (test endpoint)	Salinity (‰)	DOC (mg/L)	Temperature (°C)	Concentration (µg/L)	Reference
Cnidarian	<i>Acropora tenuis</i>	Gamete	0.2	NOEC (Fertilisation)	–	–	28	34	Reichelt-Brushett & Harrison (2005)
	–							<b>34</b>	<b>Value used in SSD</b>
	<i>Coelastrea aspera</i>	Gamete	0.2	NOEC (Fertilisation)	–	–	28	13	Reichelt-Brushett & Harrison (2005)
	–							<b>13</b>	<b>Value used in SSD</b>
	<i>Platygyra daedalea</i>	Gamete	0.2	EC10 (Fertilisation)	–	–	–	1.4	Reichelt-Brushett & Hudspith (2016)
–							<b>1.4</b>	<b>Value used in SSD</b>	
Ecnidarian	<i>Exaiptasia diaphana</i> <sup>e</sup>	Adult	28	EC10 (Progeny: offspring)	–	–	25	12	Howe et al. (2014)
		Adult	28	EC10 (Progeny: juveniles)	–	–	25	8	Howe et al. (2014)
		Adult	14	EC10 (Reproduction rate)	36	1.1	24	12	Trenfield et al. (2017)
		Adult	14	EC10 (Reproduction rate)	36	1.1	28	8.8	Trenfield et al. (2017)
		Adult	14	EC10 (Reproduction rate)	36	1.1	31	11	Trenfield et al. (2017)
–							<b>9.8</b>	<b>Value used in SSD (geometric mean 28 day test)</b>	
Echinoderm	<i>Diadema savignyi</i>	Embryo	2	NOEC (Development)	34	–	25	9.6	Rosen et al. (2015)
	–							<b>9.6</b>	<b>Value used in SSD</b>
	<i>Evechinus chloroticus</i>	Larva	3	EC10 (Development)	34	–	16	2.1	Rouchon (2015)
–							<b>2.1</b>	<b>Value used in SSD</b>	

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Taxonomic group	Species	Life stage	Exposure duration (d)	Toxicity measure (test endpoint)	Salinity (‰)	DOC (mg/L)	Temperature (°C)	Concentration (µg/L)	Reference
Annelid	<i>Hydroides elegans</i>	Gamete	8	LOEC (Metamorphosis)	34	–	24	6.2	Xie et al. (2005)
	–	–	–	–	–	–	–	<b>2.5<sup>c</sup></b>	<b>Value used in SSD</b>
Crustacean	<i>Tisbe furcata</i>	Life cycle	100	NOEC (Survival and reproduction)	34	2	15	19	Diz et al. (2009)
	–	–	–	–	–	–	–	<b>19</b>	<b>Value used in SSD</b>
Mollusc	<i>Amphibalanus amphitrite</i>	Larva	4	EC10 (Metamorphosis)	32	–	30	28	van Dam et al. (2016)
	–	–	–	–	–	–	–	<b>28</b>	<b>Value used in SSD</b>
	<i>Haliotis iris</i>	Larva	3	EC10 (Development)	34	–	14	0.7	Rouchon (2015)
	–	–	–	–	–	–	–	<b>0.7</b>	<b>Value used in SSD</b>
	<i>Mimachlamys asperima</i>	Larva	2	NOEC (Development)	33	–	18	2	Krassoi et al. (1997)
	–	–	–	–	–	–	–	<b>2</b>	<b>Value used in SSD</b>
	<i>Mytilus galloprovincialis</i>	Embryo	2	EC10 (Development)	30	1.0	22	7.2	Zitoun et al. (2019)
	Embryo	2	EC10 (Development)	30	1.9	22	8.8	Zitoun et al. (2019)	
	Embryo	2	EC10 (Development)	36	0.7	22	4.1	Zitoun et al. (2019)	
	Embryo	2	EC10 (Development)	36	1.1	22	3.2	Zitoun et al. (2019)	
	Embryo	2	EC10 (Development)	25	1.2	16	8.5	Deruytter et al. (2015)	
	Embryo	2	EC10 (Development)	30	0.6	16	2.8	Deruytter et al. (2015)	

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Taxonomic group	Species	Life stage	Exposure duration (d)	Toxicity measure (test endpoint)	Salinity (‰)	DOC (mg/L)	Temperature (°C)	Concentration (µg/L)	Reference
		Embryo	2	EC10 (Development)	35	1.5	16	2.9	Deruytter et al. (2015)
	–							<b>4.8</b>	<b>Value used in SSD (geometric mean)</b>
	<i>Mytilus trossulus</i>	Embryo	2	EC20 (Development)	35	–	20	5.6	Nadella et al. (2009)
	–							<b>5.6</b>	<b>Value used in SSD</b>
	<i>Saccostrea glomerata</i>	Larva	14	LC10 (Mortality)	34	–	20	70	Markich et al. (2002)
	–							<b>70</b>	<b>Value used in SSD</b>
Fish	<i>Atherinops affinis</i>	Embryo	12	NOEC (Development)	33	–	21	92	Anderson et al. (1991)
		Larva	12	NOEC (Development)	33	–	21	62	Anderson et al. (1991)
		Larva	7	NOEC (Mortality)	–	–	–	134	McNulty et al. (1994)
	–							<b>62</b>	<b>Value used in SSD (lowest value)</b>
	<i>Epinephelus coioides</i>	Juvenile	25	NOEC (Growth)	28	–	24	21	Wang et al. (2014)
	–							<b>21</b>	<b>Value used in SSD</b>

**a** The actual value, rather than the less than value, was used in the SSD (Warne et al. 2018).

**b** Chronic IC50 converted to chronic NOEC/EC10 value by dividing by 5 (Warne et al. 2018).

**c** Chronic LOEC value converted to chronic NOEC/EC10 value by dividing by 2.5 (Warne et al. 2018).

**d** Formerly *Nitzschia closterium*.

**e** Formerly *Aiptasia pulchella* and *Exaiptasia pallida*.



## Appendix C: DOC correction for dissolved copper default guideline values

The equations for calculating default guideline values (DGV) at different dissolved organic carbon (DOC) concentrations are provided in Table C 1. The rationale for determining the DOC correction is provided in Appendix G: Rationale for DOC correction.

**Table C 1 Equations, default guideline values at different DOC concentrations**

Equation 1	<i>Guideline value at 99% protection level</i> = $1.24 \times (DOC_{site\ specific} - 0.5) + 0.13$
Equation 2	<i>Guideline value at 95% protection level</i> = $1.24 \times (DOC_{site\ specific} - 0.5) + 0.41$
Equation 3	<i>Guideline value at 90% protection level</i> = $1.24 \times (DOC_{site\ specific} - 0.5) + 0.74$
Equation 4	<i>Guideline value at 80% protection level</i> = $1.24 \times (DOC_{site\ specific} - 0.5) + 1.5$

Where  $DOC_{site\ specific}$  means the DOC (in mg/L) at the site of interest. These equations apply up to a maximum DOC of 6 mg/L.

DGVs at representative DOC concentrations were calculated from these equations; these are presented in Table C 2.

**Table C 2 Default guideline values, dissolved copper in marine water, varying DOC concentrations**

DOC (mg/L)	DGV for dissolved copper in marine water (µg/L)			
	99% species protection	95% species protection	90% species protection	80% species protection
0.5	0.12	0.40	0.72	1.4
1	0.74	1.0	1.3	2.0
2	2.0	2.3	2.6	3.3
3	3.2	3.5	3.8	4.5
4	4.5	4.7	5.1	5.7
5	5.7	6.0	6.3	7.0
6	6.9	7.2	7.5	8.2

## Appendix D: Chronic toxicity data for Australasian species not used to derive the default guideline values

**Table D 1 Toxicity data excluded from the default guideline value derivation, dissolved copper in marine water, in order of sensitivity**

Taxonomic group	Species	Life stage	Exposure duration (d)	Endpoint	Toxicity measure <sup>a</sup>	Salinity (‰)	Temp. (°C)	Toxicity value (µg/L)	Converted concentration (µg/L)	Reason for exclusion	Reference
Diatom	<i>Thalassiosira weissflogii</i>	Exponential growth	1.25	Population growth rate	NOEC	–	21	0.075	0.075	No copper measurement	Karner et al. (2006)
Mollusc	<i>Mytilus edulis</i>	Gamete	30	Reproduction	EC50	32	–	2	0.4	No copper measurement	Stromgren & Nielsen (1991)
Annelid	<i>Galeolaria caespitosa</i>	Gamete	2	Development	EC50	32–35	16–18	4.6	0.92	No EC10/NOEC data	Ross & Bidwell (2001)
Mollusc	<i>Crassostrea gigas</i>	Egg/Larva	2	Development	EC10	35	20	1	1	No copper measurement	Worboys et al. (2002)
Mollusc	<i>Crassostrea gigas</i>	Egg/Larva	2	Development	EC50	34	20	5.3	1.1	No EC10/NOEC data	Martin et al. (1981)
Mollusc	<i>Mytilus edulis</i>	Gamete	2	Development	EC50	34	20	5.8	1.2	EC50 data only	Martin et al. (1981)
Cnidarian	<i>Pocillopora damicornis</i>	–	35	Growth	LOEC	36	26	4	1.6	No EC10/NOEC data	Bielmyer et al. (2010)
Echinoderm	<i>Heliocidaris tuberculata</i>	Zygote	3	Development	NOEC	34	18	2	2	No copper measurement	Doyle et al. (2003)
Green macroalga	<i>Ulva intestinalis</i>	Germling	5	Mortality	LC50	–	20	9.9	2	No copper measurement	Girling et al. (2015)
Mollusc	<i>Mytilus edulis</i>	Embryo	2	Development	EC50	30	15	11	2.1	EC50 data only	Arnold et al. (2009)

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Taxonomic group	Species	Life stage	Exposure duration (d)	Endpoint	Toxicity measure <sup>a</sup>	Salinity (‰)	Temp. (°C)	Toxicity value (µg/L)	Converted concentration (µg/L)	Reason for exclusion	Reference
Diatom	<i>Ceratoneis closterium</i>	Exponential growth	3	Population growth rate	EC10	35	21	2.2	2.2	No copper measurement	Hook et al. (2014)
Echinoderm	<i>Diadema setosum</i>	Embryo	0.042	Fertilisation	EC50	30	–	12	2.4	No copper measurement	Ramachandran et al. (1997)
Mollusc	<i>Mytilus edulis</i>	Embryo	2	Development	EC50	26–34	14–18	12	2.4	EC50 data only	Tucker (1998)
Crustacean	<i>Allorchestes compressa</i>	Juvenile	27	Population	NOEC	22	–	3	3	Salinity <25‰	Stauber et al. (1996)
Mollusc	<i>Nassarius dorsatus</i>	Larvae	4	Growth	EC10	34	24–31	3.1	3.1	DOC >2 (2.4 mg/L)	Trenfield et al. (2016)
Mollusc	<i>Haliotis rubra</i>	Embryo	2	Development	EC10	–	20	3.7	3.7	No copper measurement	Gorski & Nugegoda (2006)
Crustacean	<i>Moina mongolica</i>	Neonate	21	Reproduction	EC20	10	20	3.8	3.8	Salinity <25‰	Wang et al. (2007a)
Echinoderm	<i>Centrostephanus rodgersii</i>	Sperm	0.055	Fertilisation	NOEC	35.5	20	4	4	No copper measurement	King (1999)
Red macroalga	<i>Ceramium tenuicorne</i>	–	7	Growth	EC20	7	22	4	4	Salinity <25‰	Ytreberg et al. (2010)
Diatom	<i>Nitzschia paleacea</i>	Exponential growth	2	Population growth rate	EC50	–	21	23	4.6	No EC10/NOEC data	Franklin et al. (2001)
Annelid	<i>Galeolaria caespitosa</i>	Gamete	2	Development	EC50	32–35	16–18	23	4.6	No EC10/NOEC data	Ross & Bidwell (2001)
Red macroalga	<i>Champia parvula</i>	–	14	Reproduction	MATC	30	20–22	5.1	5.1	No EC10/NOEC data	Steele & Thursby (1983)
Amphipod	<i>Allorchestes compressa</i>	Juvenile	28	Growth	MEC	31	19	5.2	5.2	No EC10/NOEC data	Ahsanullah & Williams (1991)
Mollusc	<i>Mytilus edulis</i>	Embryo	2	Development	IC10	20	–	16	6.6	Salinity <25‰	CH2M HILL (1999)
Green macroalga	<i>Ulva reticulata</i>	–	7	Population	NOEC	20–40	20	8.7	8.7	No copper measurement	Mamboya et al. (2009)

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Taxonomic group	Species	Life stage	Exposure duration (d)	Endpoint	Toxicity measure <sup>a</sup>	Salinity (‰)	Temp. (°C)	Toxicity value (µg/L)	Converted concentration (µg/L)	Reason for exclusion	Reference
Diatom	<i>Skeletonema costatum</i>	Exponential growth	4	Population growth rate	EC10	33	25	9.1	9.1	No copper measurement	Fisher & Froud (1980)
Platyhelminth	<i>Stylochus pygmaeus</i>	Adult	10	Reproduction	LOEC	34	23–24	10	9.5	No EC10/NOEC data	Lee & Johnston (2007)
Brown microalga	<i>Rhodomonas salina</i>	Exponential growth	3	Population	EC50	–	20	48	9.6	No EC10/NOEC data	Debelius et al. (2009)
Sponge	<i>Halichondria panacea</i>	–	80	Population	NOEC	–	–	9.9	9.9	DOC 2.4–6.8 mg/L	Foekema et al. (2015)
Diatom	<i>Entomoneis punctulata</i>	Exponential growth	3	Population	NOEC	34	21	10	10	No copper measurement	Stauber (1995)
Diatom	<i>Nitzschia bilobata</i>	Exponential growth	3	Population	NOEC	34	21	10	10	No copper measurement	Stauber (1995)
Cnidarian	<i>Goniastrea retiformis</i>	Gamete	0.23	Fertilisation	NOEC	–	–	10	10	No copper measurement	Reichelt-Brushett & Harrison (2005)
Crustacean	<i>Moina mongolica</i>	Neonate	21	Population	EC20	10	20	12	12	Salinity <25‰	Wang et al. (2007a)
Echinoderm	<i>Heliocidaris erythrogramma</i>	Larva	6	Development	NOEC	35.5	20	13	13	No copper measurement	King (1999)
Crustacean	<i>Moina mongolica</i>	Neonate	21	Mortality	EC20	10	20	15	15	Salinity <25‰	Wang et al. (2007a)
Ascidian	<i>Ciona intestinalis</i>	Gamete	0.833	Development	NOEC	33	20	16	16	No copper measurement	Bellas et al. (2004)
Mollusc	<i>Mytilus edulis</i>	Gamete	2	Development	EC50	32–35	16–18	84	17	EC50 data only	Ross & Bidwell (2001)
Crustacean	<i>Amphibalanus amphitrite</i>	Larva	2	Development	NOEC	34	24	17	17	No copper measurement	Qiu et al. (2005)
Diatom	<i>Chaetoceros</i> sp.	Exponential growth	3	Population	EC50	–	20	88	18	No EC10/NOEC data	Debelius et al. (2009)

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Taxonomic group	Species	Life stage	Exposure duration (d)	Endpoint	Toxicity measure <sup>a</sup>	Salinity (‰)	Temp. (°C)	Toxicity value (µg/L)	Converted concentration (µg/L)	Reason for exclusion	Reference
Arthropod	<i>Allorchestes compressa</i>	Juvenile	28	Mortality	MEC	31	19	24	24	No EC10/NOEC data	Ahsanullah & Williams (1991)
Arthropod	<i>Artemia franciscana</i>	Adult	>40–<60	Mortality	NOEL	90	23	25	25	No copper measurement	Browne et al. (2002)
Echinoderm	<i>Tripneustes gratilla</i>	Gamete	0.25	Fertilisation	LOEC	31	28	25	10	No copper measurement	Edullantes & Galapate (2014)
Ochrophyta	<i>Nannochloropsis gaditana</i>	Exponential growth	3	Population	EC50	–	20	137	27	No EC10/NOEC data	Debelius et al. (2009)
Fish	<i>Morone</i> sp.	Juvenile	42	Growth	NOEC	13–14	19–25	33	33	Salinity <25‰	Bielmyer et al. (2005)
Arthropod	<i>Callinassa australiensis</i>	Adult	14	Immobilisation	EC50	34–38	18–20	190	38	No EC10/NOEC data	Ahsanullah et al. (1981)
Annelid	<i>Capitella capitata</i>	Adult	28	Mortality	LC50	–	–	200	40	No copper measurement; data from before 1980	Reish et al. (1976)
Fish	<i>Morone</i> sp.	Juvenile	21	Growth	NOEC	15	19	53	53	Salinity <25‰	Bielmyer et al. (2006)
Fish	<i>Synechogobius hasta</i>	–	30	Growth	NOEC	19–20	19–21	57	57	Salinity <25‰	Chen et al. (2013)
Crustacean	<i>Tigriopus angulatus</i>	Larva	28	Population	NOEC	34	20	60	60	No measurement of copper	Medina et al. (2008)
Green microalga	<i>Chlorella vulgaris</i>	Exponential growth	9	Population	EC10	7.6	24	94	94	No copper measurement	Latala & Surosz (1999)
Diatom	<i>Nitzschia palea</i>	Exponential growth	5	Population	NOEC	–	23	95	95	No copper measurement	Nguyen-Deroche et al. (2009)
Green microalga	<i>Stichococcus bacillaris</i>	–	9	Population	EC10	7.6	24	95	95	No copper measurement	Latala & Surosz (1999)
Green microalga	<i>Chlorella pyrenoidosa</i>	Exponential growth	4	Population	EC50	10	20	510	101	No copper measurement	Wang et al. (2007b)

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Taxonomic group	Species	Life stage	Exposure duration (d)	Endpoint	Toxicity measure <sup>a</sup>	Salinity (‰)	Temp. (°C)	Toxicity value (µg/L)	Converted concentration (µg/L)	Reason for exclusion	Reference
Green microalga	<i>Oocystis submarina</i>	–	9	Population	EC10	7.6	24	123	123	No copper measurement	Latala & Surosz (1999)
Mollusc	<i>Scutus breviculus</i>	Adult	2	Growth	NOEC	41	13	221	221	Salinity >36‰	Lee et al. (2010)
Brown macroalga	<i>Hormosira banksia</i>	Gamete	3	Growth	NOEC	34	15	500	500	No copper measurement	Kevekordes (2001)
Diatom	<i>Thalassiosira pseudonana</i>	–	4	Population	EC10	33	25	550	550	No copper measurement	Nguyen-Deroche et al. (2009)
Diatom	<i>Cylindrotheca</i> sp.	Exponential growth	3	Population	IC50	–	22	7 700	1 540	No copper measurement	Satoh et al. (2005)

a Toxicity measures:

- EC10: 10% effect concentration
- EC20: 20% effect concentration
- EC50: median effect concentration
- IC10: 10% inhibition concentration
- IC50: median inhibition concentration
- LC50: median lethal concentration
- LOEC: lowest observed effect concentration
- LOEL: lowest observed effect level
- MATC: maximum acceptable toxicant concentration
- MEC: minimum effect concentration
- NOEC: no observed effect concentration
- NOEL: no observed effect level.

## Appendix E: Guideline value derivation with preferred toxicity estimates only

Chronic data using the preferred toxicity estimates of EC/IC/LCx where x is ≤10, NEC and EC/IC/LC15–20 are summarised in Table E 1. The species sensitivity distribution (SSD) based on these data is shown in Figure E 1. The fit of the Burr III distribution to these data was good.

Based on the use of chronic data, the number of species included (15 species; classified as 'preferred') and the good fit of the distribution, protective concentration (PC) values based on these data alone would have very high reliability. These PC values were 0.35 µg/L, 0.62 µg/L, 0.9 µg/L and 1.5 µg/L for 99%, 95%, 90% and 80% species protection, respectively.

However, the use of only preferred toxicity estimates excludes three sensitive unicellular algae for which only NOEC data were available. The PC values derived from only the preferred data would protect one of these species based on 95% level of protection or above (*Proteomonas sulcate*, converted NOEC 0.8 µg/L) and would provide no protection for two of the species (*Minutocellus polymorphus*, NOEC 0.2 µg/L; *Micromonas pusilla*, NOEC 0.3 µg/L). Therefore, it was deemed necessary to include non-preferred data in the DGV derivation to ensure that sensitive species would be sufficiently protected.

**Table E 1 Summary of preferred chronic toxicity values for dissolved copper in marine water**

Taxonomic group	Species	Life stage	Exposure duration (d)	Toxicity measure (test endpoint)	Toxicity value (µg/L)
Diatom	<i>Ceratoneis closterium</i>	Exponential growth	3	EC10 (Population growth rate)	1.7
	<i>Entomoneis punctulata</i>	Exponential growth	3	IC10 (Population growth rate)	1.4
	<i>Phaeodactylum tricornutum</i>	Exponential growth	2–3	IC20 (Population growth rate)	0.7
Blue–green alga	<i>Cyanobium</i> sp.	Exponential growth	3	EC10 (Population growth rate)	300
Brown microalga	<i>Isochrysis galbana</i>	Exponential growth	3	EC10 (Population growth rate)	2.6
Green macroalga	<i>Ulva lactuca</i>	Zoospore	3	EC10 (Settlement and growth)	56
Brown macroalga	<i>Fucus vesiculosus</i>	Germling	14	NEC (Growth)	17
Cnidarian	<i>Exaiptasia diaphana</i> <sup>a</sup>	Adult	28	EC10 (Reproduction)	9.8
	<i>Platygyra daedalea</i>	Gamete	0.2	EC10 (Fertilisation)	1.4
Echinoderm	<i>Evechinus chloroticus</i>	Larva	3	EC10 (Development)	2.1
Mollusc	<i>Amphiblanus amphitrite</i>	Larva	4	EC10 (Metamorphosis)	28
	<i>Haliotis iris</i>	Larva	3	EC10 (Development)	0.7
	<i>Mytilus galloprovincialis</i>	Embryo	2	EC10 (Development)	4.8
	<i>Mytilus trossulus</i>	Embryo	2	EC20 (Development)	5.6
	<i>Saccostrea glomerata</i>	Larva	7	LC10 (Mortality)	70

<sup>a</sup> Formerly *Aiptasia pulchella* and *Exaiptasia pallida*.

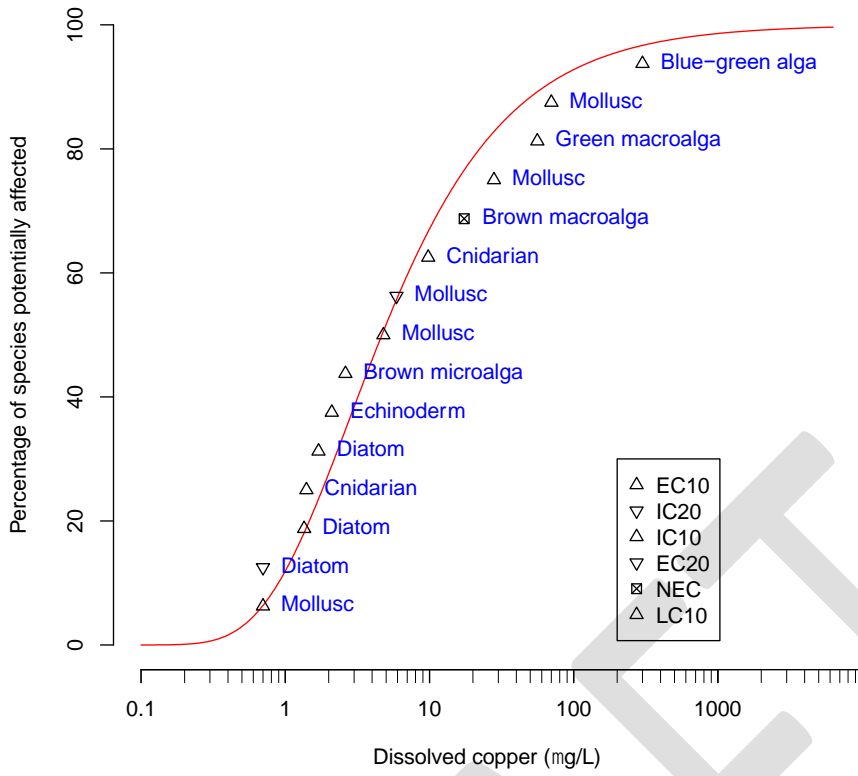


Figure E 1 Species sensitivity distribution, preferred chronic data only, dissolved copper in marine water



## Appendix F: Modality assessment for copper

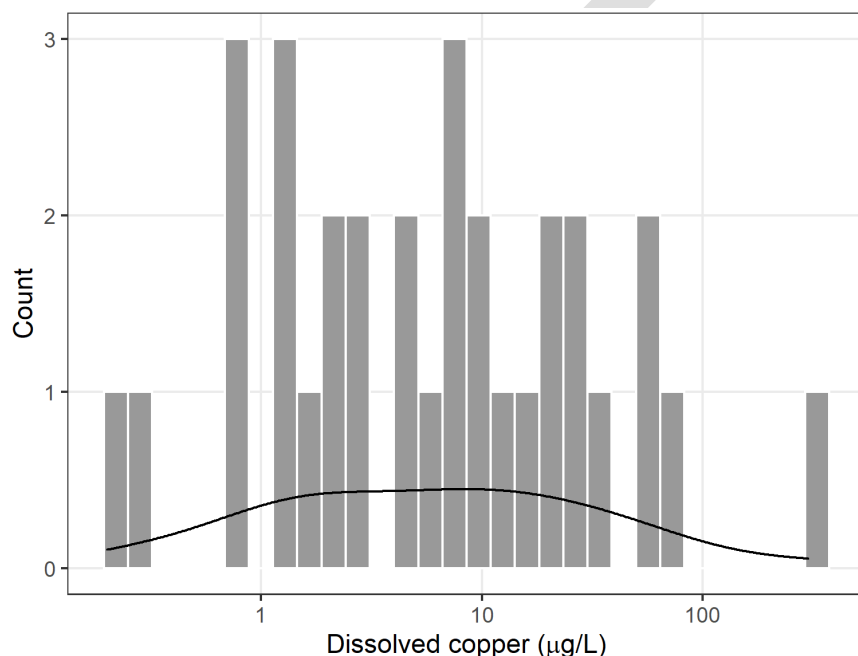
A modality assessment was undertaken for copper according to the four questions stipulated in Warne et al. (2018). These questions and their answers are listed as follows.

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

Yes, there are differences in the mode of action for copper between fish/invertebrates and plants/algae. Copper is a well-known fungicide and herbicide.

### Does the dataset suggest bimodality?

A visual assessment of the SSD for the final toxicity dataset (32 species, Section 4) suggested there was bimodality, with a break in the data between 2.6  $\mu\text{g/L}$  and 4.1  $\mu\text{g/L}$ ; this is more visible in a histogram (Figure F 1).



**Figure F 1 Histogram and density plot of final toxicity dataset**

A bimodality coefficient of 0.34 was calculated for the log-transformed dataset. Values between 0.555 and 1 represent bimodal distributions, so the value of 0.34 did not indicate bimodality in this dataset.

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

There was some indication of taxa-specific sensitivity when broad taxonomic groupings were compared, with microalgae typically more sensitive than macroinvertebrates, macroalgae or fish (Figure F 2). This is primarily due to the highly sensitive diatom species, as shown in Figure F 3 where toxicity is compared across the 12 taxonomic groups. There are also some mollusc species that are highly sensitive.

The data are also shown in the SSD in Figure F 4, and indicate that microalgae are in the bottom two-thirds of the final dataset (though there is one data point for a blue-green alga at the top of the curve). However, the distribution of the microalgae data does not correlate with the bimodality observed in the histogram, indicating that this is not the cause of the apparent bimodality.

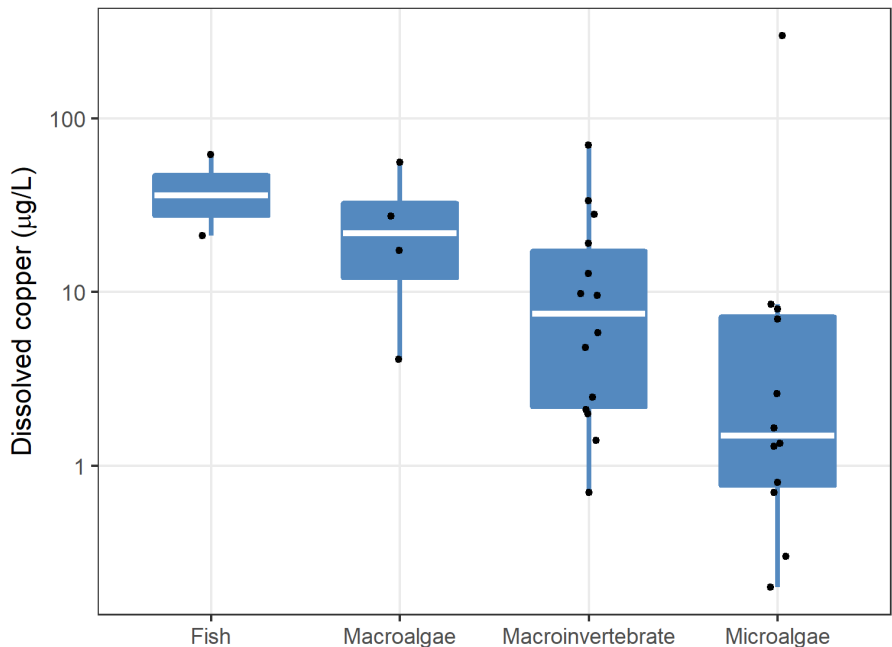


Figure F 2 Comparison of dissolved copper toxicity between taxa

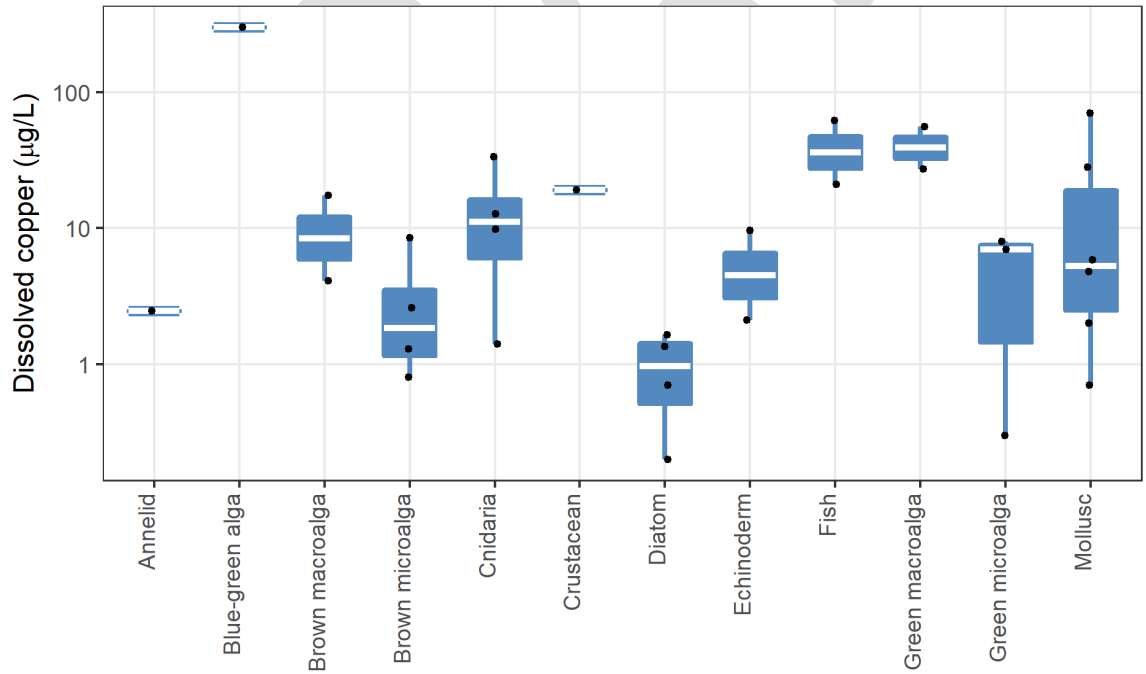
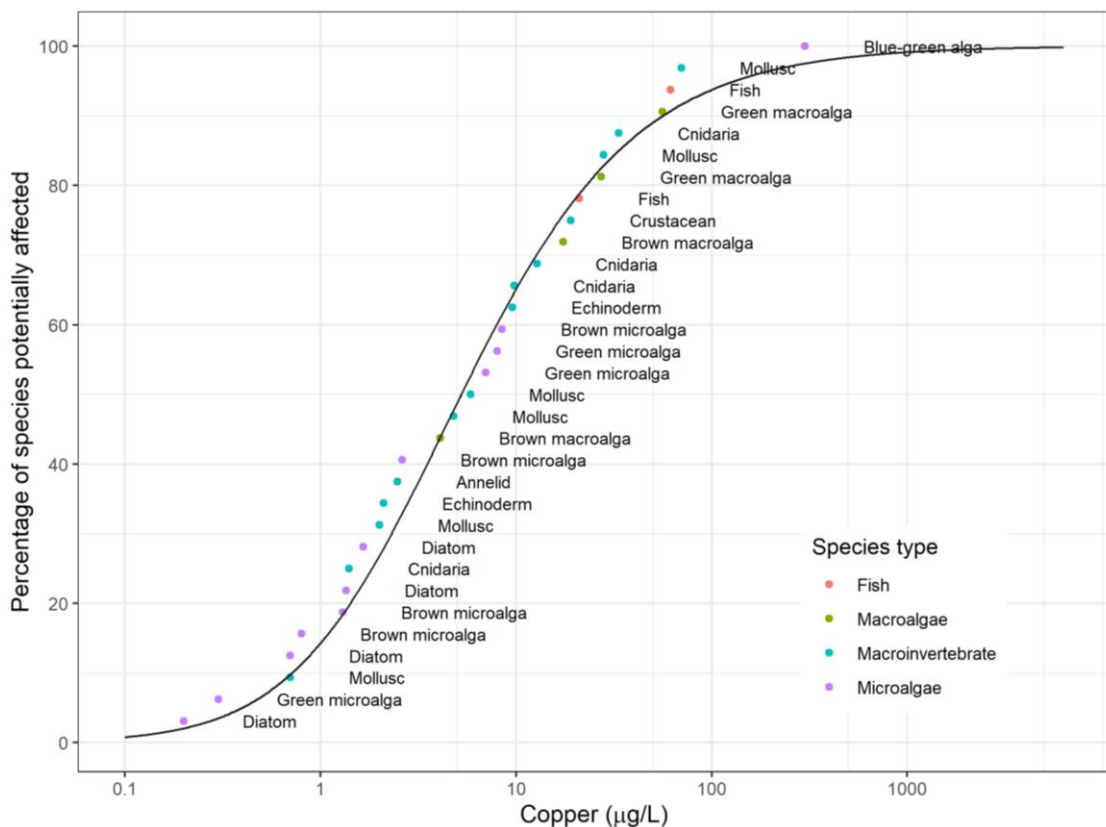


Figure F 3 Comparison of dissolved copper toxicity between taxa



**Figure F 4 Species sensitivity distribution, final dataset for dissolved copper**

**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?**

It is possible that the apparent bimodality of the dataset is due to an artefact of the test conditions. Many of the values just above the apparent break between 2.6 µg/L and 4.1 µg/L are NOEC data, and three (for *C. huxleyi*, *D. tertiolecta* and *Tetraselmis* sp.) are from the same study (Levy et al. 2007) and were likely tested at the same nominal concentrations. If tests were repeated and NEC or EC10 data calculated, it is likely that there would be a difference in these concentrations and the apparent break in the dataset would be reduced. Thus, it is probable that the break in the dataset is an artefact of the data included as well as the inclusion of NOEC data.

The lack of strong evidence for bimodality and the high degree of overlap in taxa-specific sensitivity, with the possible exception of diatoms, supports the use of all the acceptable data to derive the DGVs.

## Appendix G: Rationale for DOC correction

Bioavailability corrections for dissolved copper default guideline values (DGVs) in marine water are being used (or have been proposed) in several jurisdictions. These are discussed as follows.

The European Union risk assessment (European Copper Institute 2008) investigated relationships between dissolved organic carbon (DOC) and toxicity data based on six species:

- *Mytilus galloprovincialis*
- *Mytilus edulis*
- *Crassostrea gigas*
- *Dendraster excentricus*
- *Strongylocentrotus purpuratus*
- *Fucus vesiculosus*.

Copper toxicity was investigated at multiple DOC concentrations for each of these species in chronic tests (48 hour embryo-larval development tests for the invertebrates, and zoospore growth for the algae). Although NOEC data were used in the risk assessment SSD, EC50 data were used to develop statistical relationships between DOC and toxicity, as EC50 data are stronger descriptors of toxicity than NOEC data. There was no significant difference between the slopes for each species; therefore, a single power relationship was developed for all six species after normalising EC50 data to the most sensitive species (*Mytilus edulis*). All NOEC data included in the SSD were then normalised based on this relationship, with an assumption that added DOC was 100% active and natural DOC was 50% active. An HC5 of 5.2 µg/L of copper was calculated based on 2 mg/L of DOC (equivalent to 1 mg/L active DOC). A predicted no effect concentration (PNEC) of 2.6 µg/L of copper was recommended based on the HC5 and an assessment factor of 2, due to the absence of high quality mesocosm data. An adjustment based on DOC is provided from the power equation in Figure G 1.

$$PNEC_{site\ specific} = 2.64 \times \left(\frac{DOC}{2}\right)^{0.6139}$$

**Figure G 1 Adjustment based on DOC**

For the United Kingdom guidelines (Maycock et al. 2011), a linear model was derived to describe the relationship between DOC and toxicity, based on EC10 data for *M. galloprovincialis* from 48 hour embryo-larval development tests (Arnold et al. 2005, 2006, 2009, 2010a)s. The dataset used in the European Union risk assessment (European Copper Institute 2008) was updated with additional data and all data were adjusted to a DOC of 1 mg/L (natural DOC, equivalent to 0.5 mg/L active DOC). The HC5 calculated from this was 2.64 µg/L and an assessment factor of 1 was applied, resulting in a PNEC of 2.64 µg/L. Where natural DOC exceeds 1 mg/L, a site-specific PNEC can be calculated with the equation in Figure G 2.

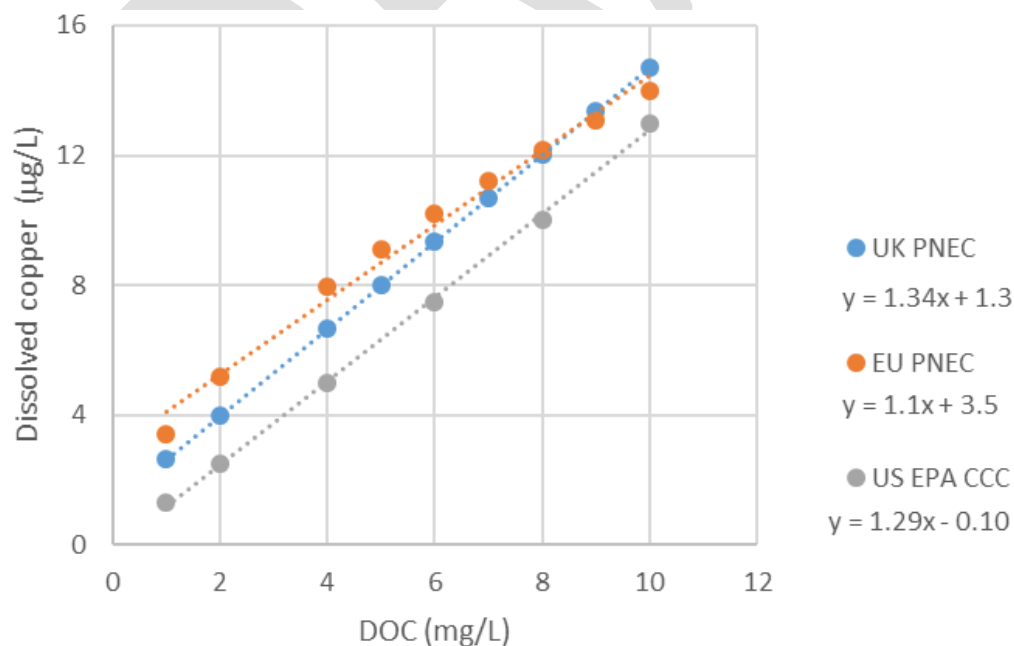
$$PNEC_{site\ specific} = 2.64 + 2.677 \times \left( \frac{DOC}{2} - 0.5 \right)$$

**Figure G 2 Calculating PNEC when natural DOC >1 mg/L**

The United States implemented a DOC correction within its marine biotic ligand model (BLM) (USEPA 2016a). A BLM includes models for copper speciation, based on the chemical equilibrium model WHAM, which describes metal interactions with natural organic matter. The chemical speciation part of the marine water BLM was developed from copper complexometric titrations of water samples collected in San Diego and Pearl Harbour (Chadwick et al. 2008). Three DOC binding sites were included in the model (Chadwick et al. 2008). The BLM also included models of the interaction of copper and competing cations with physiologically active sites (biotic ligands), using binding constants. The copper and cation binding constants were based on *M. galloprovincialis* and *S. purpuratus* 48 hour embryo-larval development tests. The BLM was implemented with the United States water quality criterion for copper in marine water, thus enabling adjustment of the criterion based on the temperature, pH, salinity and DOC of water.

The United States chronic criterion for copper in marine/estuarine water, known as a criterion continuous concentration (CCC), is based on acute data that have been adjusted based on a geometric mean of acute-chronic ratios for five sensitive freshwater species and two estuarine/marine species. This method was used as there were only two species for which there was acceptable chronic values according to the guideline development principles (which use EC20 for chronic criteria derivation). The CCC was obtained from the draft water quality criteria document (USEPA 2016a) for a salinity of 30‰, temperature of 20°C and pH of 8.2.

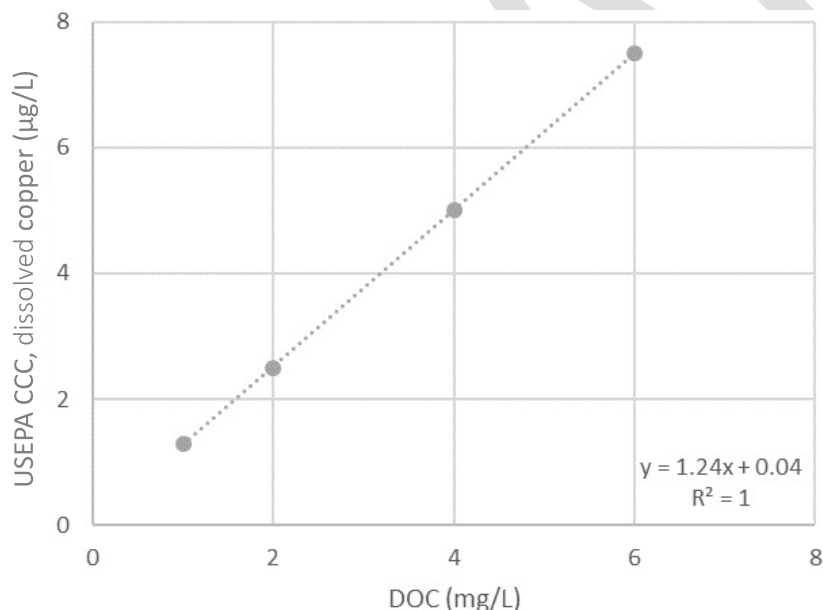
The relationships between copper guideline values and DOC concentrations from these three jurisdictions are shown in Figure G 3 with linear regression lines. These show very similar slopes, especially for the United States and United Kingdom guidelines. The European Union data are derived from a power relationship and, therefore, the linear model does not fit as well.



**Figure G 3 Copper and DOC concentration, bioavailability corrections of the European Union, United Kingdom and United States**

For the ANZG (2018) DGVs for dissolved copper in marine water, the BLM was selected as a suitable model to correct the DGVs for varying levels of DOC because it is based on chemical speciation modelling rather than toxicity data, and therefore should be applicable to all species. However, there is a recent study that suggests copper toxicity to settled mussels is not affected by DOC and that DOC corrections should not be used for this life stage (Deruytter et al. 2017). The BLM showed a generally linear relationship between DOC and dissolved copper; however, the fit at low DOC concentrations was improved when the regression was based on DOC from 1 mg/L to 6 mg/L. This linear regression provided a slope of 1.24 (Figure G 4). This slope results in most accurate predictions of values at the lower DOC concentrations, which are representative of most New Zealand and Australian marine waters and where bioavailability and toxicity would be highest.

The lack of a protective effect of DOC on settled mussels is potentially due to DOC-Cu complexes either being bioavailable to these mussels or becoming bioavailable within the gill microenvironment (Deruytter et al. 2017). Therefore, the DOC adjustment should not result in DGVs that exceed toxicity estimates reported for settled mussels, to ensure that these species are protected. The 5% effect concentration (EC5) for (*M. edulis*) is 8.8 µg/L for clearance rate (Deruytter et al. 2017). Adjustments of the DGVs up to 6 mg/L DOC should be protective of this species, with a DOC-modified DGV of 8.3 µg/L for 80% level of protection. Therefore, there is no adjustment recommended above 6 mg/L DOC, irrespective of whether DOC is higher than this.



Note: grey dotted line shows fitted linear regression for data between a DOC of 1 mg/L and 6 mg/L.

**Figure G 4 Relationship between USEPA criterion continuous concentration (CCC) for copper and DOC concentration, as predicted by the USEPA BLM**

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