



# Proposed default guideline values for the protection of aquatic ecosystems: Diuron – marine

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### **Executive summary**

Diuron (3-(3,4-dichlorophenyl)-1,1-dimethylurea, CAS No. 330-54-1) is a urea herbicide, or more specifically a phenylurea herbicide. Diuron is a common photosynthesis-inhibiting herbicide used for the total control of weeds and mosses as well as selective control of germinating grass and broad-leaved weeds in a variety of crops (University of Hertfordshire 2013). Non-agricultural uses include its application to urban and industrial situations (i.e. sides of roads, railroads, areas around industrial buildings), as well for aquatic weed and algae control in flood mitigation channels and as a boat anti-foulant.

The previous Australian and New Zealand guideline value (GV) (formerly referred to as a trigger value) for diuron in marine environments was a low reliability value (using the ANZECC and ARMCANZ 2000 reliability scheme) as it was calculated using an assessment factor of 1000 applied to a chronic toxicity value for a marine mollusc (Warne 2000). More data on diuron toxicity are now available, including toxicity data to phototrophic species (species that photosynthesise e.g. plants and algae) resulting in high reliability values (Warne et al. 2015).

Diuron is significantly ( $p\leq0.05$ ) more toxic to phototrophic species (e.g. plants and algae) than to other organisms (non-phototrophs) and a species sensitivity distribution (SSD) that used all freshwater and marine species resulted in a bimodal distribution. For this reason the default GVs were derived using only phototroph toxicity data. The lowest reported chronic toxicity value to marine species is 0.54 µg/L (marine microalga, 3-day NOEC) and the lowest reported acute toxicity value to marine species is 1 µg/L (marine coral, 4-day NOEC).

Very high reliability default GVs for diuron in marine waters were derived based on chronic no observed effect concentration (NOEC), 10% effect concentration (EC10) and chronic estimated 50% effect (EC50) data for 20 marine species from six phyla and 11 classes, with a good fit of the SSD to the toxicity data. It should be noted that the default GVs presented here are expressed in terms of the active ingredient (diuron) rather than commercial formulations. The default GVs for a range of protection levels are:

Default guideline value type (ANZECC and ARMCANZ 2000)	Proposed diuron (marine) toxicity default guideline values (μg/L)						
Reliability	Very High						
High conservation value systems (99% species protection)	0.43						
Slightly to moderately disturbed systems (95% species protection)	0.67						
Highly disturbed systems (90% species protection)	0.86						
Highly disturbed systems (80% species protection)	1.2						

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#### **1** Introduction

Diuron is a herbicide ( $C_9H_{10}CI_2N_2O$  and Figure 1) that at room temperature is in the form of odourless, colourless crystals. It is the active ingredient of a variety of commercial herbicide formulations. Major metabolites of diuron are subsequentially demethylated diuron compounds: N'-(3-chlorophenyl)-N,N-dimethylurea (m-CPDMU), N'-(3,4-dichlorophenyl)-N-methylurea (DCPMU) and 3,4-dichlorophenylurea (DCPU) (APVMA 2011). The ecological effects of the minor metabolite 3,4-dichloroaniline (3,4-DCA) are not well known.



Figure 1 Structure of diuron (C = carbon, H = hydrogen, Cl = chlorine, N = nitrogen and O = oxygen).

Physicochemical properties of diuron that may affect its environmental fate and toxicity are presented in Table 1.

Physicochemical property	Value
Molecular weight	233.1 amu <sup>1</sup>
Aqueous solubility	37.4 mg/L @ temperature of $25^{\circ}C^{1}$ 35.6 mg/L @ temperature of $20^{\circ}C^{2}$
Logarithm of the octanol-water partition coefficient (log Kow)	2.85 $\pm$ 0.03, temperature of 25°C <sup>1</sup> 2.87 @ pH 7, temperature of 20°C <sup>2</sup>
Logarithm of the organic carbon water partition coefficient (log Koc)	2.60 <sup>1</sup> , 2.91 <sup>2</sup>
Logarithm of the bioconcentration factor (log BCF)	0.975 <sup>2</sup>
Half-life (t <sub>1/2</sub> ) in water	175 days (lagoon prediction) with majority of diuron (90%) residing in sediment <sup>3</sup>
Half-life (t <sub>1/2</sub> ) in soil	90 – 180 days <sup>1</sup> 75.5 days <sup>2</sup>

Table 1 Summary of selected physicochemical properties of diuron.

<sup>1</sup> BCPC (2012). <sup>2</sup> Pesticide Properties Database (University of Hertfordshire 2013). <sup>3</sup> Peterson and Batley (1991).

Diuron belongs to the phenylurea group within the urea class of herbicides, which also includes linuron and isoproturon. Diuron is extensively used in agriculture and forestry applications for the control of weeds as well as selective control of germinating grass and broad-leaved weeds in a variety of crops such as pineapples, bananas, asparagus, peas, cotton, sugarcane, wheat, barley oats, and ornamentals including tulips (BCPS 2012; University of Hertfordshire 2013). Diuron is also used to control weeds and algae in and around water bodies and is a component of marine antifouling paints (APVMA 2009). In Australia, diuron is one of the most heavily used herbicides, exceeded only by glyphosate, simazine and atrazine (AATSE 2002). It is a pre-emergence, residual herbicide as well as a post-emergence knockdown (University of Hertfordshire 2013) that exhibits some solubility in water (Table 1).

Diuron is absorbed principally through the roots of plants. It is then translocated acropetally (i.e. movement upwards from the base of plants to the apex) in the xylem and accumulates in the leaves (BCPC 2012). Diuron exerts its toxicity in aquatic plants (including algae) by inhibiting electron transport in the photosystem II (PSII) complex (University of Hertfordshire 2013), a key process in photosynthesis that occurs in the thylakoid membranes of chloroplasts. Urea herbicides bind to the plastoquinone B ( $Q_B$ ) protein binding site on the D1 protein in PSII. This prevents the transport of electrons to synthesise adenosine triphosphate (ATP, used for cellular metabolism) and nicotinamide adenine dinucleotide phosphate (NADPH, used in converting CO<sub>2</sub> to glucose), and therefore prevents CO<sub>2</sub> fixation (Wilson et al. 2000).

In addition to its main mode of action, PSII-inhibiting herbicides can lead to marked increases in the formation of reactive oxygen species (ROS) (Halliwell 1991). These include the synthesis of singlet oxygen (OH<sup>-</sup>), superoxide ( $O_2^{-}$ ) and hydrogen peroxide ( $H_2O_2$ ). Reactive oxygen species are highly reactive forms of oxygen that readily react with, and bind to, biomolecules including deoxyribonucleic acid (DNA) and ribonucleic acid (RNA). Reactive oxygen species are created during normal cellular functioning particularly in biochemical processes that involve the generation of energy, e.g. photosynthesis in chloroplasts and the Krebs cycle in the mitochondria of cells. In phototrophs, ROS are formed when the absorbed light energy exceeds the ability to convert  $CO_2$  to organic molecules, thus accumulating oxygen (Chen et al. 2012). Normal concentrations of ROS are involved in a number of cellular processes (Chen et al. 2012). However, prolonged exposure to elevated concentrations of ROS in plants, as a result of biotic (e.g. disease) and/or abiotic stressors (e.g. PSII-inhibiting herbicides), can cause irreversible cell damage and ultimately lead to cell death (apoptosis).

Diuron ultimately ends up in marine environments as a result of surface and/or subsurface runoff from agricultural applications following heavy or persistent rain events, as well as from antifouling paints (biocides) applied to the hull of marine vessels (APVMA 2009). Loss of diuron via volatilisation is minimal, it has a moderate solubility in water (Table 1) and low soil adsorption ability as indicated by its low log Koc value (Table 1) (Field et al. 2003). Diuron is relatively mobile and has been found to leach to groundwater and be transported in surface waters (Field et al. 2003; AVPMA 2011). Diuron has been commonly detected in estuarine and marine waters and sediments in countries including Japan, Netherlands, Portugal, Spain, Sweden and the UK (Konstantinour and Albanis 2004). This is at least partly due to its widespread use as a component of anti-fouling paints but other sources include urban and agricultural land. For example, diuron has been has been detected in approximately 66% of surface water samples collected between 2011–15 in waterways that drain agricultural land and discharge to the Great Barrier Reef (based on data in Turner et al. 2013a, 2013b; Wallace et al. 2014, 2015, 2016; Garzon-Garcia et al. 2015). It is therefore not surprising that diuron has been widely detected throughout the Great Barrier Reef and in fact is the most frequently detected pesticide in these waters between 2010 and 2015 (Kennedy et al. 2011; Bentley et al. 2012; Gallen et al. 2013; 2014; 2015).

In Australia, the APVMA suspended the registration of selected diuron products in late 2011 and enforced significant restrictions on the use of reaffirmed products. The main restriction prohibited the use of diuron during no-spray windows (from December 5, 2011 to March 31, 2012 onwards) for tropical crops including sugarcane, with restrictions being specific to the climatic and geographic conditions of each region. Other restrictions included specifying maximum application rates for different times of the year. Diuron is currently registered for use in Australia and many other countries, however, it has been reviewed in the United States (draft 2003), Canada (2007), United Kingdom (2007) and Europe (2007 and 2008) (APVMA 2009). Current restraints on diuron use in Australia can be found at <a href="http://apvma.gov.au/node/12511">http://apvma.gov.au/node/12511</a>.

### 2 Aquatic Toxicology

The review of the literature revealed that there were five published studies (Gagnon and Rawson 2009; Magnusson et al. 2008; Negri et al. 2005; Seery et al. 2006, Stauber et al. 2008) and one unpublished study (Seery and Pradella in prep) that determined the toxicity of diuron to Australasian marine organisms. These studies determined the toxicity of diuron to: the corals *Acropora millepora* and *Pocillopora damicornis* (Negri et al. 2005); the fish *Pagarus auratus* (Gagnon and Rawson 2009); the microalgae *Isochrysis galbana* (Seery and Pradella in prep), *Nephroselmis pyriformis* (Magnusson et al. 2008), *Nitzschia closterium* and *Entomoneis punctulata* (Stauber et al. 2008); and the macroalga *Hormosira banksii* (Seery et al. 2006). All of these studies generated data that passed the screening and quality assurance processes. However, only the data for *I. galbana, N. pyriformis*, *N. closterium* and *E. punctulata* were used to derive default GVs as they were chronic.

A summary of the high and moderate quality raw toxicity data for marine species is provided below.

#### Chronic data

There were marine chronic toxicity data for one coral, one crustacean, one fish, three macroalgae and 17 species of microalgae. The toxicity values for the corals had 90-day NOEC values for fecundity and size of 0.91 and 8.8  $\mu$ g/L, respectively. The toxicity values for crustaceans were 28-day NOEL and LOEC (mortality) values of 270 and 560  $\mu$ g/L, respectively. There was a single toxicity value of 440  $\mu$ g/L for a 38-day LOEC (mortality) for a fish. There were six 15-day values (EC10, LOEC and EC50) that ranged from 2.3 to 87.8  $\mu$ g/L, a 7-day EC50 (length) of 3.4  $\mu$ g/L and 10-day NOEC and LOEC (biomass) values of 2.5 and 5  $\mu$ g/L, respectively for macroalga. The toxicity data for microalgae were 3-day EC10, NOEC, LOEC, EC50 and IC50 values for a variety of endpoints (biomass yield, growth rate, area under the curve; cell density; cell number and biomass) that ranged from 0.54 to 95  $\mu$ g/L, 4-day LOEC and EC50 (cell density) values of 3.8 to 27  $\mu$ g/L and 10- and 14-day EC50 values that ranged from 10 to 76.9  $\mu$ g/L.

#### Acute data

There were marine acute ecotoxicity data for three corals, seven crustaceans, one echinoid, four fish, four macroalgae, two molluscs and one polychaete. The six toxicity values for corals consisted of 24-hour LC10 and LC50 (mortality) values of 91 and 4,800 µg/L, 96-hour NOEC values (fertilisation rate and survival) both of 1,000 µg/L and 96-hour NOEC (survival) values of 100 and 1000 µg/L (adult and larvae, respectively). The crustacean toxicity data consisted of 1-, 2- and 4day EC/LC50 (mortality) values that ranged from 1,000 to 21,000 µg/L and a 4-day NOEL (mortality) of 600 µg/L. The three toxicity values for echinoids were all for the same species and the 48-hour NOEC, LOEC and EC50 (fertilisation rate) values ranged from 500 to 5,090 µg/L. The 15 toxicity data for fish consisted of 36-hour to 6-day NOEC, NOEL, LOEC, LC10 and LC50 (mortality and hatching success) values that ranged from 50 to 7,826 µg/L. The macroalgae toxicity data were 4-day NOEC and EC50 (biomass – fresh weight) values that ranged from 1.3 to 20 µg/L, 2-day EC50 (germination) values of 4,650 and 6,290 µg/L, a 2-day EC50 (length) of 6750 µg/L and two 3-day NOEC (leaf length) values of 87.8 µg/L. The mollusc toxicity data consisted of 24- and 48-hour LC10 and LC50 (mortality) values of >1,000 µg/L, two 96-hour EC50 values (mortality/abnormality and growth) of 1,800 µg/L and 4,800 µg/L and a 96-hour NOEL (mortality/abnormality) of 2,400 µg/L. The single value for a polychaete was a 48-hour LC50 (mortality) of 16,000 µg/L.

#### **3 Factors Affecting Toxicity**

No factors have been reported as modifying the toxicity of diuron. As with many organic chemicals it might be expected that dissolved and particulate organic matter and suspended solids would affect its bioavailability and toxicity. However, any such effect would be relatively minor given the relatively low log Koc value of diuron (Table 1).

## **4** Guideline Derivation

The Australian and New Zealand default Guideline Values (GVs) for diuron in marine waters are provided in Table 2. Details of how the default GVs were calculated and the toxicity data that were used are provided below. As with all the other pesticides that have GVs, the GVs for diuron are expressed in terms of the concentration of the active ingredient.

Measured log bioconcentration factor (BCF) values for diuron are low (Table 1) and below the threshold at which secondary poisoning must be considered (i.e. threshold log BCF = 4, Warne et al. 2015). Therefore, the default GVs for diuron do not need to account for secondary poisoning.

Diuron default guideline values (marine) <sup>1</sup>			Re	liability classification <sup>2</sup>
Percent species protection	Concentration (µg/L)		Criterion	Result
99%	0.43		Sample size	20
95%	0.67		Type of toxicity data	Chronic NOEC/EC10 and chronic estimated NOEC/EC10 data
90%	0.86		SSD model fit	Good
80%	1.2		Reliability	Very High

Table 2 Default guideline values (µg/L) for diuron for the protection of marine ecosystems.

Guideline values were derived using the Burrlioz 2.0 (2016) software.

<sup>2</sup> See Warne et al. (2015) for definitions of guideline value "reliability".

## **5 Toxicity Data Used in Derivation**

The previous Australian and New Zealand GV (formerly referred to as a trigger value) for diuron in marine environments was a low reliability value (using the ANZECC and ARMCANZ 2000 reliability scheme) as it was based on one acute toxicity value for a marine mollusc species (Warne 2000). This value was calculated using the assessment factor (AF) method, dividing the lowest acute toxicity value of 1,800 µg/L by an assessment factor of 1000 (Warne 2000). Under the new method for deriving GVs (Warne et al. 2015) this value would be classified as having a *very low reliability*.

To obtain toxicity data for diuron to marine organisms, an extensive search of the scientific literature was conducted. In addition, the databases of the USEPA ECOTOX (USEPA 2015a), Office of the Pesticide Program (USEPA 2015b), the Australasian Ecotoxicology Database (Warne et al. 1998) and the ANZECC and ARMCANZ WQG toxicant databases (Sunderam et al. 2000) were searched. More data on diuron toxicity are now available, including data for phototrophic

species (species that photosynthesise, e.g. plants and algae) to derive default GVs of higher reliability, using the scheme of Warne et al. (2015).

In total, there were toxicity data for 45 marine species (12 phyla and 20 classes) that passed the screening and quality assessment processes (Attachment A, Table 4). The represented phyla were Annelida, Arthropoda, Bacillariophyta, Chlorophyta, Chordata, Cnidaria, Echinodermata, Haptophyta, Mollusca, Ochrophyta, Rhodophyta and Tracheophyta. The 20 classes were Actinopterygii (which accounts for approximately 99% of fish), Anthazoa (a class of cnidaria i.e. corals), Bacillariophyceae (a major grouping of diatoms), Bacillariophyceae incertae sedis (a group of diatoms), Bivalvia (a class of molluscs), Branchiopoda (a grouping of crustaceans), Chlorophyceae (a major grouping of green algae), Chrysophyceae (a class of golden algae), Coccolithophyceae (a class of yellow algae), Echinodea (a class of echinoderms), Entognatha (a class of arthropods), Florideophyceae (a class or sub-class of red algae), Liliopsida (monocots), Malacostraca (a large grouping of crustaceans), Maxillopoda (a class of crustaceans), Mediophyceae (another algae grouping), Nephrophyceae (a class of green algae), Phaeophyceae (a class of annelid worms) and Porphyridiophyceae (a class red algae).

Based on the mode of action of diuron, it would be expected that phototrophic species would be more sensitive than non-phototrophic species. The diuron ecotoxicity data for phototrophs and heterotrophs were then tested using the parametric two-sample *t* test to see if they were uni- or multi-modal. This indicated that the two groups had significantly different (p=<0.0001, Attachment B) sensitivities. Therefore, as recommended by Warne et al. (2015), only the ecotoxicity data for the more sensitive group of organisms (in this case, phototrophs) were used in calculating the default GVs.

There were chronic no observed effect concentration (NOEC) and 10% effect concentration (EC10) data for seven phototrophic marine species (that belonged to five phyla and five classes), which meets the minimum data requirements (i.e., at least five species belonging to at least four phyla) to use a SSD to derive a GV (Warne et al. 2015). However, the resulting protective concentration (PC) values were not recommended as the default GVs for diuron in marine waters since very high reliability GVs were able to be derived by including chronic estimated NOEC/EC10 values in the derivation. Further explanation is provided in Attachment C.

When the dataset was expanded to combine the chronic NOEC/EC10 with the chronic estimated NOEC (chronic LOEC/EC50 toxicity data that had been converted to estimates of chronic NOEC/EC10 by dividing by 5) values of marine phototrophic species, there were 20 species belonging to six phyla and 11 classes which met the minimum data requirements to use a SSD to derive default GVs (Warne et al. 2015). The number of species and taxa in the toxicity data used to derive the default GVs (Table 2) combined with the good fit of the distribution to these toxicity data (Figure 2) resulted in a very high reliability set of default GVs.

A summary of the toxicity data (one value per species) used to calculate the default GVs for diuron in marine environments is provided in Table 3. Further details about all the data for marine species that passed the screening and quality assurance schemes, including those used to derive the single species values used to calculate the default GVs are presented in Table 4, Attachment A.

# Table 3 Summary of the single toxicity value for each phototrophic species that was used to derive the default guideline values for diuron in marine waters. Data are arranged in alphabetical order of the test species.

Taxonomic group	Species	Phyla	Class	Duration (days)	Type <sup>1</sup>	Toxicity endpoint	Toxicity value (μg/L)
Microalgae	Achnanthes brevipes	Bacillariophyta	Bacillariophyceae	3	Chronic est. NOEC	Biomass yield, growth rate, AUC <sup>2</sup>	4.8
Microalgae	Amphora exigua	Bacillariophyta	Bacillariophyceae	3	Chronic est. NOEC	Biomass yield, growth rate, AUC <sup>2</sup>	6.2
Macroalgae	Ceramium tenuicorne	Rhodophyta	Florideophyceae	7	Chronic est. NOEC	Final length	0.68
Microalgae	Chaetoceros gracilis	Bacillariophyta	Mediophyceae	3	Chronic est. NOEC	Cell number	7.2
Microalgae	Dunaliella tertiolecta	Chlorophyta	Chlorophyceae	4	Chronic est. NOEC	Cell density	1.52
Microalgae	Emiliania huxleyi	Haptophyta	Coccolithophyceae	3	Chronic NOEC	Mortality	0.54
Microalgae	Entomoneis punctulata	Bacillariophyta	Bacillariophyceae	3	Chronic NOEC	Cell density	2.0
Microalgae	lsochrysis galbana	Haptophyta	Coccolithophyceae	3	Chronic EC10	Cell density	1.09
Microalgae	Monochrysis Iutheri	Ochrophyta	Chrysophyceae	3	Chronic est. NOEC	Biomass yield, growth rate, AUC <sup>2</sup>	3.6
Microalgae	Navicula forcipata	Bacillariophyta	Bacillariophyceae	4	Chronic est. NOEC	Cell density	5.4
Microalgae	Navicula incerta	Bacillariophyta	Bacillariophyceae	3	Chronic est. NOEC	Biomass yield, growth rate, AUC <sup>2</sup>	18.6
Microalgae	Nephroselmis pyriformis	Chlorophyta	Nephrophyceae	3	Chronic EC10	Cell density	2.2
Microalgae	Nitzschia closterium	Bacillariophyta	Bacillariophyceae	3	Chronic NOEC	Cell density	2.0
Microalgae	Phaeodactylu m tricornutum	Bacillariophyta	Bacillariophyta incertae sedis	10	Chronic est. NOEC	Biomass yield, growth rate, AUC <sup>2</sup>	2.0
Microalgae	Porphyridium cruentum	Rhodophyta	Porphyridiophyceae	3	Chronic est. NOEC	Biomass yield, growth rate, AUC <sup>2</sup>	4.8
Macroalgae	Saccharina japonica	Ochrophyta	Phaeophyceae	15	Chronic EC10	Fresh weight	2.3
Microalgae	Skeletonema costatum	Bacillariophyta	Mediophyceae	4	Chronic est. NOEC	Cell density	1.18
Microalgae	Thalassiosira fluviatilis	Bacillariophyta	Mediophyceae	3	Chronic est. NOEC	Biomass yield, growth rate, AUC <sup>2</sup>	19
Microalgae	Thalassiosira pseudonana	Bacillariophyta	Mediophyceae	4	Chronic est. NOEC	Cell density	0.86
Macrophyte	Zostera marina	Tracheophyta	Liliopsida	10	Chronic NOEC	Biomass (Old and new growth)	2.5

<sup>1</sup> Chronic NOEC/EC10 = no conversions applied; Chronic est. NOEC = chronic LOEC/EC50 values that were converted to chronic NOEC/EC10 values by dividing by 5 (Warne et al. 2015).<sup>2</sup> AUC = area under the growth curve.

#### **6** Species Sensitivity Distribution

The cumulative frequency (species sensitivity) distribution (SSD) of the 20 phototrophic marine species that were used to derive the default GVs is presented in Figure 2.



Figure 2 Cumulative frequency distribution generated using Burrlioz 2.0 (2016) of the sensitivity (chronic 10% effect concentration (EC10) and no observed effect concentration (NOEC) data with chronic estimated NOEC/EC10 data) values of marine phototrophic species to diuron. Chronic NOEC/EC10 = no conversions applied; Chronic est. NOEC = chronic LOEC/EC50 values that were converted to chronic NOEC/NOEL/EC10 values by dividing by 5 (Warne et al. 2015).

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# 8 Glossary, acronyms, abbreviations

Acute toxicity	An adverse effect that occurs as the result of a short-term exposure to a chemical relative to the organism's life span. Refer to Warne et al. (2015) for examples of acute exposures.
ANZECC	Australian and New Zealand Environment and Conservation Council.
ARMCANZ	Agricultural and Resource Management Council of Australia and New Zealand.
Bimodal	When the distribution of the sensitivity of species to a toxicant has two modes. This typically occurs with chemicals with specific modes of action. For example, herbicides are designed to affect plants at low concentrations but most animals are only affected at high concentrations.
CAS no.	Chemical Abstracts Service number. Each chemical has a unique identifying number that is allocated to it by the American Chemical Society.
Chronic toxicity	An adverse effect that occurs as the result of exposure to a chemical for a substantial portion of the organism's life span or an adverse sub-lethal effect on a sensitive early life stage. Refer to Warne et al. (2015) for examples of chronic exposures.
Default guideline value (Default GV)	A guideline value recommended for generic application in the absence of a more specific guideline value (e.g. site-specific), in the Australian and New Zealand Water Quality Guidelines.
ECx	The concentration of a chemical in water that is estimated to produce a x% effect on a sub-lethal endpoint. The magnitude of x can vary from 1 to 100, however values between 5 and 50 are more typical. The ECx is usually expressed as a time-dependent value (e.g. 24-hour or 96-hour ECx).
EC50 (Median effective concentration)	The concentration of a chemical in water that is estimated to produce a 50% effect on a sub-lethal endpoint. The EC50 is usually expressed as a time-dependent value (e.g. 24-hour or 96-hour EC50).
Endpoint	A measurable biological effect including, but not limited to, lethality, immobility, growth inhibition, immunological responses, organ effects, developmental and reproductive effects, behavioural effects, biochemical changes, genotoxicity, etc.
Guideline value (GV)	A measurable quantity (e.g. concentration) or condition of an indicator for a specific environmental 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 environmental value. Guideline values for more than one indicator should be used simultaneously in a multiple lines of

	evidence approach.
LC50 (Median lethal concentration)	The concentration of a chemical in water that is estimated to kill 50% of the test organisms. The LC50 is usually expressed as a time-dependent value (e.g. 24-hour or 96-hour LC50).
LOEC (Lowest observed effect concentration)	The lowest concentration of a chemical used in a toxicity test that has a statistically significant ( $p \le 0.05$ ) adverse effect on the exposed population of test organisms as compared with the controls. All higher concentrations should also cause statistically significant effects.
Mode of action	The means by which a chemical exerts its toxic effects. For example, triazine herbicides inhibit the photosystem II component of plants photosynthesis biochemical reaction.
NOEC (No observed effect concentration)	The highest concentration of a toxicant used in a toxicity test that does not have a statistically significant (p>0.05) effect, compared to the controls. The statistical significance is measured at the 95% confidence level.
Phototrophs	Organisms that photosynthesize as their main means of obtaining energy e.g. plants and algae.
PSII	Photosystem II of the photosynthetic biochemical pathway.
Site-specific	Relating to something that is confined to, or valid for, a particular place. Site-specific trigger values are relevant to the location or conditions that are the focus of a given assessment.
Species	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.
SSD	Species sensitivity distribution. A method that plots the cumulative frequency of species sensitivity and fits the best possible 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 concentration of chemical.

# Attachment A. Summary details of all marine toxicity data that passed the screening and quality assessment processes

Table 4 Summary of the key characteristics of the marine diuron toxicity data (acute and chronic) that passed the screening and quality assurance processes. It includes both phototrophic and non-phototrophic species for marine waters only.

Phyla	Class	Species	Life stage	Exposure duration (days)	Test type	Toxicity measure (test endpoint)	Test medium	Salinity (‰)	Temp . (°C)	рН	Concentration (µg/L)	Reference
Annelida	Polychaeta	Serpulid worm (Hydroides elegans)	Larvae	2	Acute	LC50 (Live animal count)	Marine water	33 ± 0.5	25 ± 1	8.1 – 8.4	16,000	Bao et al. (2011)
											16,000	GEOMETRIC MEAN
Arthropoda	Branchiopoda	Microinvertebrate (Artemia salina)	II - III instar	1	Acute	LC50 (Mortality)	Artificial seawater	35	25	Not stated	12,010	Koutsaftis and Aoyama (2007)
											12,010	GEOMETRIC MEAN
Arthropoda	Entognatha	Macroinvertebrate (Proisotoma minuta)	60 days old	7	Acute	LC50 (Mortality)	Artificial seawater	754 μS/cm	23	5.7	1,000	Park and Lees (2005)
											1,000	GEOMETRIC MEAN
Arthropoda	Malacostraca	Macroinvertebrate (Americamysis bahia)	Not stated	4	Acute	LC50 (Mortality)	Natural or artificial filtered seawater	20 ± 3	23 ± 1	Not stated	1,100	USEPA (2015)
											1,000	GEOMETRIC MEAN
Arthropoda	Malacostraca	Macroinvertebrate (Americamysis bahia)	Not stated	4	Acute	NOEL (Mortality)	Natural or artificial filtered seawater	20 ± 3	23 ± 1	Not stated	600	USEPA (2015)
											600	GEOMETRIC MEAN
Arthropoda	Malacostraca	Macroinvertebrate	Life cycle	28	Chronic	LOEC	Natural or artificial	20 ± 3	25 ± 2	Not	560	USEPA

		(Americamysis bahia)				(Mortality)	filtered seawater			stated		(2015)
											560	GEOMETRIC MEAN
Arthropoda	Malacostraca	Macroinvertebrate ( <i>Americamysis</i> <i>bahia</i> )	Life cycle	28	Chronic	NOEL (Mortality)	Natural or artificial filtered seawater	20 ± 3	25 ± 2	Not stated	270	USEPA (2015)
											270	GEOMETRIC MEAN
Arthropoda	Malacostraca	Microinvertebrate ( <i>Elasmopus rapax</i> )	Juvenile	4	Acute	LC50 (Mortality)	Marine water	33 ± 0.5	25 ± 1	8.1 – 8.4	3,000	Bao et al. (2011)
											3,000	GEOMETRIC MEAN
Arthropoda	Malacostraca	Macroinvertebrate (Penaeus aztecus)	Juvenile	2	Acute	LC50 (Mortality)	Natural, filtered or artificial seawater	20 ± 3	23 ± 1	Not stated	1,000	USEPA (2015)
											1,000	GEOMETRIC MEAN
Arthropoda	Maxillopoda	Macroinvertebrate ( <i>Balanus</i> <i>amphitrite</i> )	Larvae	1	Acute	LC50 (Mortality)	Marine water	33 ± 0.5	25 ± 1	8.1 – 8.4	21,000	Bao et al. (2011)
											21,000	GEOMETRIC MEAN
Arthropoda	Maxillopoda	Microinvertebrate ( <i>Nitocra spinipes</i> )	Not stated	4	Acute	LC50 (Mortality)	Artificial seawater	5	22 ± 2	Not stated	4,000	Karlsson et al. (2006)
											4,000	GEOMETRIC MEAN
Arthropoda	Maxillopoda	Microinvertebrate ( <i>Tigriopus</i> <i>japonicus</i> )	Adult	4	Acute	LC50 (Mortality)	Marine water	33 ± 0.5	25 ± 1	8.1 – 8.4	11,000	Bao et al. (2011)
											11,000	GEOMETRIC MEAN
Bacillariophyta	Bacillariophyceae	Microalgae (Achnanthes brevipes)	Not stated	3	Chronic	EC50 (Biomass yield, growth rate, AUC)	Synthetic salt water or filtered natural salt water	30 ± 5	20 ± 2	8.0 ± 0.1	24	USEPA (2015)
											24	GEOMETRIC MEAN
											4.8 <sup>@</sup>	VALUE USED

												IN SSD
Bacillariophyta	Bacillariophyceae	Microalgae (Amphora exigua)	Not stated	3	Chronic	EC50 (Biomass yield, growth rate, AUC)	Synthetic salt water or filtered natural salt water	30 ± 5	20 ± 2	8.0 ± 0.1	31	USEPA (2015)
											31	GEOMETRIC MEAN
											6.2 <sup>@</sup>	VALUE USED IN SSD
Bacillariophyta	Bacillariophyceae	Microalgae (Entomoneis punctulata)	Not stated	3	Chronic	EC50 (Cell density)	Filtered seawater	30	21	8.1 – 8.4	24	Stauber et al. (2008)
											24	GEOMETRIC MEAN
Bacillariophyta	Bacillariophyceae	Microalgae (Entomoneis punctulata)	Not stated	3	Chronic	LOEC (Cell density)	Filtered seawater	30	21	8.1 – 8.4	6	Stauber et al. (2008)
											6	GEOMETRIC MEAN
Bacillariophyta	Bacillariophyceae	Microalgae (Entomoneis punctulata)	Not stated	3	Chronic	NOEC (Cell density)	Filtered seawater	30	21	8.1 – 8.4	2	Stauber et al. (2008)
											2	GEOMETRIC MEAN
											2	VALUE USED IN SSD
Bacillariophyta	Bacillariophyceae	Microalgae ( <i>Navicula</i> forcipata)	Exponent- ial growth phase	4	Chronic	EC50 (Cell density)	F2 marine media	Not stated	20 ± 1	Not stated	27	Gatidou and Thomaidis (2007)
											27	GEOMETRIC MEAN
											5.4 <sup>@</sup>	VALUE USED IN SSD
Bacillariophyta	Bacillariophyceae	Microalgae ( <i>Navicula incerta</i> )	Not stated	3	Chronic	EC50 (Biomass yield, growth rate, AUC)	Synthetic salt water or filtered natural salt water	Not stated	20 ± 2	8.0 ± 0.1	93	USEPA (2015)
											93	GEOMETRIC MEAN

											18.6 <sup>@</sup>	VALUE USED IN SSD
Bacillariophyta	Bacillariophyceae	Microalgae ( <i>Nitzschia</i> <i>closterium</i> )	Not stated	3	Chronic	EC50 (Cell density)	Filtered seawater	30	21	8.1 – 8.4	17	Stauber et al. (2008)
											17	GEOMETRIC MEAN
Bacillariophyta	Bacillariophyceae	Microalgae ( <i>Nitzschia</i> <i>closterium</i> )	Not stated	3	Chronic	LOEC (Cell density)	Filtered seawater	30	21	8.1 – 8.4	6	Stauber et al. (2008)
											6	GEOMETRIC MEAN
Bacillariophyta	Bacillariophyceae	Microalgae ( <i>Nitzschia</i> <i>closterium</i> )	Not stated	3	Chronic	NOEC (Cell density)	Filtered seawater	30	21	8.1 – 8.4	2	Stauber et al. (2008)
											2	GEOMETRIC MEAN
											2	VALUE USED IN SSD
Bacillariophyta	Bacillariophyceae	Microalgae ( <i>Nitzschia</i> <i>closterium</i> )	Not stated	3	Chronic	EC50 (Biomass Yield, growth Rate, AUC)	Synthetic salt water or filtered natural salt water	30 ± 5	20 ± 2	8.0 ± 0.1	50	USEPA (2015)
											50	GEOMETRIC MEAN
Bacillariophyta	Bacillariophyta incertae sedis	Microalgae (Phaeodactylum tricornutum)	Not stated	3	Chronic	IC50 (Cell density)	Seawater	30	20	8.4	20.98	Clarkson et al. (1998)
											20.98	GEOMETRIC MEAN
Bacillariophyta	Bacillariophyta incertae sedis	Microalgae (Phaeodactylum tricornutum)	Not stated	14	Chronic	EC50 (Cell density)	Seawater	30	20	8.4	76.9	Clarkson et al. (1998)
											76.9	GEOMETRIC MEAN
Bacillariophyta	Bacillariophyta incertae sedis	Microalgae (Phaeodactylum tricornutum)	Not stated	10	Chronic	EC50 (Biomass yield, growth rate, AUC)	Synthetic salt water or filtered natural salt water	30 ± 5	20 ± 2	8.0 ± 0.1	10	USEPA (2015)

											10	GEOMETRIC MEAN
											2 <sup>@</sup>	VALUE USED IN SSD
Bacillariophyta	Mediophyceae	Microalgae (Chaetoceros gracilis)	Not stated	3	Chronic	IC50 (Cell number)	Provasoli medium	Not stated	25	Not stated	36	Koutsaftis and Aoyama (2006)
											36	GEOMETRIC MEAN
											7.2 <sup>@</sup>	VALUE USED IN SSD
Bacillariophyta	Mediophyceae	Microalgae ( <i>Skeletonema</i> <i>costatum</i> )	<7 days old	4	Chronic	EC50 (Cell density)	Marine water	33 ± 0.5	25 ± 1	8.1 – 8.4	5.9	Bao et al. (2011)
											5.9	GEOMETRIC MEAN
											1.18 <sup>@</sup>	VALUE USED IN SSD
Bacillariophyta	Mediophyceae	Microalgae (Thalassiosira fluviatilis)	Not stated	3	Chronic	EC50 (Biomass yield, growth rate, AUC)	Synthetic natural salt water or filtered natural salt water	30 ± 5	20 ± 2	8.0 ± 0.1	95	USEPA (2015)
											95	GEOMETRIC MEAN
											19 <sup>@</sup>	VALUE USED IN SSD
Bacillariophyta	Mediophyceae	Microalgae ( <i>Thalassiosira</i> pseudonana)	Not stated	4	Chronic	EC50 (Cell density)	Marine water	33 ± 0.5	25 ± 1	8.1 – 8.4	4.3	Bao et al. (2011)
											4.3	GEOMETRIC MEAN
											0.86 <sup>@</sup>	VALUE USED IN SSD
Chlorophyta	Chlorophyceae	Microalgae (Dunaliella tertiolecta)	Log growth phase	4	Chronic	EC50 (Cell density)	F2 marine media	20	25	Not stated	9.8	DeLorenzo et al. (2011)
Chlorophyta	Chlorophyceae	Microalgae	Exponent-	4	Chronic	EC50	F2 marine media	Not	20 ± 1	Not	5.9	Gatidou and

		(Dunaliella tertiolecta)	ial growth phase			(Cell density)		stated		stated		Thomaidis (2007)
											7.60	GEOMETRIC MEAN
											1.52 <sup>@</sup>	VALUE USED IN SSD
Chlorophyta	Chlorophyceae	Microalgae (Dunaliella tertiolecta)	Log growth phase	4	Chronic	LOEC (Cell density)	F2 marine media	20	25	Not stated	3.8	DeLorenzo et al. (2011)
											3.8	GEOMETRIC MEAN
Chlorophyta	Chlorophyceae	Microalgae (Dunaliella tertiolecta)	Not stated	10	Chronic	EC50 (Biomass yield, growth rate, AUC)	Synthetic natural salt water or filtered natural salt water	30 ± 5	20 ± 2	8.0 ± 0.1	20	USEPA (2015)
											20	GEOMETRIC MEAN
Chlorophyta	Nephrophyceae	Microalgae (Nephroselmis pyriformis)	Not stated	3	Chronic	EC10 (Biomass)	Filtered seawater	Not stated	24	Not stated	5.2	Magnusson et al. (2008)
											5.2	GEOMETRIC MEAN
Chlorophyta	Nephrophyceae	Microalgae (Nephroselmis pyriformis)	Not stated	3	Chronic	EC10 (Cell density)	Filtered seawater	Not stated	24	Not stated	2.2	Magnusson et al. (2008)
											2.2	GEOMETRIC MEAN
											2.2	VALUE USED IN SSD
Chlorophyta	Nephrophyceae	Microalgae (Nephroselmis pyriformis)	Not stated	3	Chronic	EC50 (Biomass)	Filtered seawater	Not stated	24	Not stated	8	Magnusson et al. (2008)
											8	GEOMETRIC MEAN
Chlorophyta	Nephrophyceae	Microalgae (Nephroselmis pyriformis)	Not stated	3	Chronic	EC50 (Cell density)	Filtered seawater	Not stated	24	Not stated	5.8	Magnusson et al. (2008)

											5.8	GEOMETRIC MEAN
Chordata	Actinopterygii	Sheepshead Minnow (Cyprinodon variegatus)	Not stated	4	Acute	NOEL (Mortality)	Seawater	20 ± 5	22 ± 2	7.5 – 8.5	3,600	USEPA (2015)
											3,600	GEOMETRIC MEAN
Chordata	Actinopterygii	Sheepshead Minnow (Cyprinodon variegatus)	Not stated	4	Acute	LC50 (Mortality)	Seawater	20 ± 5	22 ± 2	7.5 – 8.5	6,700	USEPA (2015)
											6,700	GEOMETRIC MEAN
Chordata	Actinopterygii	Sheepshead Minnow (Cyprinodon variegatus)	Early life	38	Chronic	LOEC (Mortality)	Dilution water	Not stated	25 ± 2	Not stated	440	USEPA (2015)
											440	GEOMETRIC MEAN
Chordata	Actinopterygii	Flathead Grey Mullet ( <i>Mugil cephalus</i> )	Juvenile	2	Acute	LC50 (Mortality)	Seawater	20 ± 5	23 ± 2	7.5 – 8.5	6,300	USEPA (2015)
											6,300	GEOMETRIC MEAN
Chordata	Actinopterygii	White mullet ( <i>Mugil curema</i> )	Not stated	2	Acute	LC50 (Mortality)	Not stated	Not stated	29	Not stated	6,300	Butler (1963)
											6,300	GEOMETRIC MEAN
Chordata	Actinopterygii	Australasian Snapper ( <i>Pagrus auratus</i> )	<2 hour fertilised eggs	1.5	Acute	NOEC (Hatching success)	Filtered seawater	40	24.5	Not stated	50	Gagnon and Rawson (2009)
											50	GEOMETRIC MEAN
Chordata	Actinopterygii	Turbot ( <i>Psetta maxima</i> )	Embryo	2	Acute	LC10 (Mortality)	Artificial seawater	34.2	18 ± 1	8.29 ± 0.1	1,396	Mhadhbi and Beiras (2012)
											1,396	GEOMETRIC MEAN

Chordata	Actinopterygii	Turbot ( <i>Psetta maxima</i> )	Embryo	2	Acute	LC50 (Mortality)	Artificial seawater	34.2	18 ± 1	8.29 ± 0.1	1,076	Mhadhbi and Beiras (2012)
											1,076	GEOMETRIC MEAN
Chordata	Actinopterygii	Turbot ( <i>Psetta maxima</i> )	Embryo	2	Acute	LOEC (Mortality)	Artificial seawater	34.2	18 ± 1	8.29 ± 0.1	1,250	Mhadhbi and Beiras (2012)
											1,250	GEOMETRIC MEAN
Chordata	Actinopterygii	Turbot ( <i>Psetta maxima</i> )	Embryo	2	Acute	NOEC (Hatching success)	Artificial seawater	34.2	18 ± 1	8.29 ± 0.1	5,000	Mhadhbi and Beiras (2012)
											5,000	GEOMETRIC MEAN
Chordata	Actinopterygii	Turbot ( <i>Psetta maxima</i> )	Embryo	2	Acute	NOEC (Mortality)	Artificial seawater	34.2	18 ± 1	8.29 ± 0.1	625	Mhadhbi and Beiras (2012)
											625	GEOMETRIC MEAN
Chordata	Actinopterygii	Turbot ( <i>Psetta maxima</i> )	Larvae	6	Acute	LC10 (Mortality)	Artificial seawater	34.2	18 ± 1	8.29 ± 0.1	1,617	Mhadhbi and Beiras (2012)
											1,617	GEOMETRIC MEAN
Chordata	Actinopterygii	Turbot ( <i>Psetta maxima</i> )	Larvae	6	Acute	LC50 (Mortality)	Artificial seawater	34.2	18 ± 1	8.29 ± 0.1	7,826	Mhadhbi and Beiras (2012)
											7,826	GEOMETRIC MEAN
Chordata	Actinopterygii	Turbot ( <i>Psetta maxima</i> )	Larvae	6	Acute	LOEC (Mortality)	Artificial seawater	34.2	18 ± 1	8.29 ± 0.1	1,250	Mhadhbi and Beiras (2012)
											1,250	GEOMETRIC MEAN
Chordata	Actinopterygii	Turbot ( <i>Psetta maxima</i> )	Embryo	6	Acute	NOEC (Hatching success)	Artificial seawater	34.2	18 ± 1	8.29 ± 0.1	5,000	Mhadhbi and Beiras (2012)
											5,000	GEOMETRIC MEAN
Chordata	Actinopterygii	Turbot (Psetta maxima)	Larvae	6	Acute	NOEC (Mortality)	Artificial seawater	34.2	18 ± 1	8.29 ± 0.1	625	Mhadhbi and Beiras (2012)

											625	GEOMETRIC MEAN
Cnidaria	Anthozoa	Coral (Acropora millepora)	Larvae	4	Acute	NOEC (Fertilisation rate)	Filtered seawater	Not stated	28	7	1,000	Negri et al. (2005)
											1,000	GEOMETRIC MEAN
Cnidaria	Anthozoa	Coral (Acropora millepora)	Larvae	4	Acute	NOEC (Survival)	Filtered seawater	Not stated	28	7	1,000	Negri et al. (2005)
											1,000	GEOMETRIC MEAN
Cnidaria	Anthozoa	Coral (Acropora tumida)	Larvae	1	Acute	LC10 (Live animal count)	Marine water	33 ± 0.5	25 ± 1	8.1 – 8.4	91	Bao et al. (2011)
											91	GEOMETRIC MEAN
Cnidaria	Anthozoa	Coral (Acropora tumida)	Larvae	1	Acute	LC50 (Live animal count)	Marine water	33 ± 0.5	25 ± 1	8.1 – 8.4	4,800	Bao et al. (2011)
											4,800	GEOMETRIC MEAN
Cnidaria	Anthozoa	Coral (Acropora valida)	Not stated	90	Chronic	NOEC (Fecundity)	Unfiltered oceanic seawater	Not stated	26 - 29	7.2	0.91	Cantin et al. (2007)
											0.91	GEOMETRIC MEAN
Cnidaria	Anthozoa	Coral ( <i>Acropora valida</i> )	Egg	90	Chronic	NOEC (Size)	Unfiltered oceanic seawater	Not stated	26 - 29	7.2	8.8	Cantin et al. (2007)
											8.8	GEOMETRIC MEAN
Cnidaria	Anthozoa	Coral (Pocillopora damicornis)	Adult	4	Acute	NOEC (Survival)	Filtered seawater	Not stated	28	7	100	Negri et al. (2005)
											100	GEOMETRIC MEAN
Cnidaria	Anthozoa	Coral ( <i>Pocillopora</i>	Larvae	4	Acute	NOEC (Survival)	Filtered seawater	Not stated	28	7	1,000	Negri et al. (2005)

		damicornis)										
											1,000	GEOMETRIC MEAN
Echinodermata	Echinoidea	Macroinvertebrate ( <i>Paracentrotus</i> <i>lividus</i> )	Not stated	2	Acute	EC50 (Fertilisation rate)	Natural filtered seawater (FSW)	38	18 ± 1	8.0 ± 0.2	5,090	Manzo et al. (2006)
											5,090	GEOMETRIC MEAN
Echinodermata	Echinoidea	Macroinvertebrate ( <i>Paracentrotus</i> <i>lividus</i> )	Not stated	2	Acute	LOEC (Fertilisation rate)	Natural filtered seawater (FSW)	38	18 ± 1	8.0 ± 0.2	1,000	Manzo et al. (2006)
											1,000	GEOMETRIC MEAN
Echinodermata	Echinoidea	Macroinvertebrate (Paracentrotus lividus)	Not stated	2	Acute	NOEC (Fertilisation rate)	Natural filtered seawater (FSW)	38	18 ± 1	8.0 ± 0.2	500	Manzo et al. (2006)
											500	GEOMETRIC MEAN
Echinodermata	Echinoidea	Macroinvertebrate (Paracentrotus lividus)	Not stated	0.021 (0.5 hours)	Acute	EC50 (Fertilisation rate)	Natural filtered seawater (FSW)	38	18 ± 1	8.0 ± 0.2	2,870	Manzo et al. (2008)
											2,870	GEOMETRIC MEAN
Haptophyta	Coccolithophycea e	Microalgae ( <i>Emiliania huxleyi</i> )	Exponent- ial growth phase	3	Chronic	EC50 (Cell number)	Seawater	33	17	8.3 - 8.4	2.26	Devilla et al. (2005)
											2.26	GEOMETRIC MEAN
Haptophyta	Coccolithophycea e	Microalgae ( <i>Emiliania huxleyi</i> )	Exponent- ial growth phase	3	Chronic	NOEC (Cell number)	Seawater	33	17	8.3 - 8.4	0.54	Devilla et al. (2005)
											0.54	GEOMETRIC MEAN
											0.54	VALUE USED IN SSD
Haptophyta	Coccolithophycea e	Microalgae (Isochrysis	Not stated	3	Chronic	EC10 (Cell density)	0.45 mm filtered seawater,	31 ± 2	29 ± 1	8.2 ± 0.2	1.09	Seery et al. (in prep)

		galbana)					autoclaved and f/2 Guillard's Marine					
											1.09	GEOMETRIC MEAN
											1.09	VALUE USED IN SSD
Haptophyta	Coccolithophycea e	Microalgae ( <i>Isochrysis</i> galbana)	Not stated	3	Chronic	EC50 (Cell density)	0.45 mm filtered seawater, autoclaved and f/2 Guillard's Marine	31 ± 2	29 ± 1	8.2 ± 0.2	2.77	Seery et al. (in prep)
											2.77	GEOMETRIC MEAN
Haptophyta	Coccolithophycea e	Microalgae ( <i>Isochrysis</i> galbana)	Not stated	10	Chronic	EC50 (Biomass yield, growth rate, AUC)	Synthetic salt water or filtered natural salt water	30 ± 5	20 ± 2	8.0 ± 0.1	10	USEPA (2015)
											10	GEOMETRIC MEAN
Mollusca	Bivalvia	Macroinvertebrate (Crassostrea virginica)	SPAT juvenile	4	Acute	EC50 (Mortality/Abnorm al development)	Good quality unfiltered seawater (natural or artificial with food added)	>12	25	Not stated	4,800	USEPA (2015)
Mollusca	Bivalvia	Macroinvertebrate (Crassostrea virginica)	Embryo/ larvae	4	Acute	EC50 (Mortality/Abnorm al development)	Good quality unfiltered seawater (natural or artificial with food added)	>12	25	Not stated	1,800	USEPA (2015)
											2,940	GEOMETRIC MEAN
Mollusca	Bivalvia	Macroinvertebrate (Crassostrea virginica)	SPAT juvenile	4	Acute	NOEL (Mortality/Abnorm al development)	Good quality unfiltered seawater (natural or artificial with food added)	>12	25	Not stated	2,400	USEPA (2015)
											2,400	GEOMETRIC MEAN
Mollusca	Bivalvia	Macroinvertebrate (Crassostrea virginica)	Not stated	4	Acute	EC50 (Growth)	Not stated	25	22	Not stated	1,800	Butler (1964)

											1,800	GEOMETRIC MEAN
Mollusca	Bivalvia	Macroinvertebrate ( <i>Crassostrea</i> gigas)	Mature fertilised eggs	1	Acute	LC10 (Mortality)	Daigo's Artificial Seawater	Not stated	25	Not stated	1,000	Tsunemasa and Okamura (2011)
											1,000	GEOMETRIC MEAN
Mollusca	Bivalvia	Macroinvertebrate ( <i>Crassostrea</i> gigas)	Mature fertilised eggs	1	Acute	LC50 (Mortality)	Daigo's Artificial Seawater	Not stated	25	Not stated	1,000	Tsunemasa and Okamura (2011)
											1,000	GEOMETRIC MEAN
Mollusca	Bivalvia	Macroinvertebrate ( <i>Crassostrea</i> gigas)	Mature fertilised eggs	2	Acute	LC10 (Mortality)	Daigo's Artificial Seawater	Not stated	25	Not stated	1,000	Tsunemasa and Okamura (2011)
											1,000	GEOMETRIC MEAN
Mollusca	Bivalvia	Macroinvertebrate ( <i>Crassostrea</i> gigas)	Mature fertilised eggs	2	Acute	LC50 (Mortality)	Daigo's Artificial Seawater	Not stated	25	Not stated	1,000	Tsunemasa and Okamura (2011)
											1,000	GEOMETRIC MEAN
Ochrophyta	Chrysophyceae	Microalgae (Monochrysis lutherî)	Not stated	3	Chronic	EC50 (Biomass yield, growth rate, AUC)	Synthetic salt water or filtered natural salt water	30 ± 5	20 ± 2	8.0 ± 0.1	18	USEPA (2015)
											18	GEOMETRIC MEAN
											3.6 <sup>@</sup>	VALUE USED IN SSD
Ochrophyta	Phaeophyceae	Macroalgae (Hormosira banksii)	Gametes	2	Acute	EC50 (Germination)	Seawater	30 - 32	21 - 22	8.0 – 8.5	4,650	Seery et al. (2006)
Ochrophyta	Phaeophyceae	Macroalgae (Hormosira banksii)	Not stated	2	Acute	EC50 (Germination)	Seawater	Not stated	18 ± 1	Not stated	6,290	Myers et al. (2006)
											5,408	GEOMETRIC MEAN

Ochrophyta	Phaeophyceae	Macroalgae (Hormosira banksii)	Not stated	2	Acute	EC50 (Length)	Seawater	Not stated	18 ± 1	Not stated	6,750	Myers et al. (2006)
											6,750	GEOMETRIC MEAN
Ochrophyta	Phaeophyceae	Macroalgae (Saccharina japonica)	Thalli	15	Chronic	EC10 (Disc area)	Artificial seawater	Not stated	Not stated	8.4	3.9	Kumar et al. (2010)
											3.9	GEOMETRIC MEAN
Ochrophyta	Phaeophyceae	Macroalgae (Saccharina japonica)	Thalli	15	Chronic	EC10 (Fresh weight)	Artificial seawater	Not stated	Not stated	8.4	2.3	Kumar et al. (2010)
											2.3	GEOMETRIC MEAN
											2.3	VALUE USED IN SSD
Ochrophyta	Phaeophyceae	Macroalgae (Saccharina japonica)	Thalli	15	Chronic	EC50 (Disc area)	Artificial seawater	Not stated	Not stated	8.4	40	Kumar et al. (2010)
											40	GEOMETRIC MEAN
Ochrophyta	Phaeophyceae	Macroalgae (Saccharina japonica)	Thalli	15	Chronic	EC50 (Fresh weight)	Artificial seawater	Not stated	Not stated	8.4	87.8	Kumar et al. (2010)
											87.8	GEOMETRIC MEAN
Ochrophyta	Phaeophyceae	Macroalgae (Saccharina japonica)	Thalli	15	Chronic	LOEC (Fresh weight)	Artificial seawater	Not stated	Not stated	8.4	25	Kumar et al. (2010)
											25	GEOMETRIC MEAN
Ochrophyta	Phaeophyceae	Macroalgae (Saccharina japonica)	Thalli	15	Chronic	LOEC (Growth rate - Chlorophyll a fluorescence)	Artificial seawater	Not stated	Not stated	8.4	6.25	Kumar et al. (2010)
											6.25	GEOMETRIC MEAN

Rhodophyta	Florideophyceae	Macroalgae (Ceramium tenuicorne)	Not stated	7	Chronic	EC50 (Final length)	Artificial seawater	5	22 ± 2	Not stated	3.4	Karlsson et al. (2006)
											3.4	GEOMETRIC MEAN
											0.68 <sup>@</sup>	VALUE USED IN SSD
Rhodophyta	Florideophyceae	Macroalgae (Gracilaria tenuistipitata)	Not stated	4	Acute	EC50 (Biomass - Fresh weight)	Filtered deep sea water and ultra- pure water	>5ppt	25	8	15	Hershner et al. (1982)
Rhodophyta	Florideophyceae	Macroalgae (Gracilaria tenuistipitata)	Not stated	4	Acute	EC50 (Biomass - Fresh weight)	Filtered deep sea water and ultra- pure water	>5ppt	25	8	20	Hershner et al. (1982)
											17.3	GEOMETRIC MEAN
Rhodophyta	Florideophyceae	Macroalgae (Gracilaria tenuistipitata)	Not stated	4	Acute	NOEC (Biomass - Fresh weight)	Filtered deep sea water and ultra- pure water	>5ppt	25	8	1.3	Hershner et al. (1982)
Rhodophyta	Florideophyceae	Macroalgae (Gracilaria tenuistipitata)	Not stated	4	Acute	NOEC (Biomass - Fresh weight)	Filtered deep sea water and ultra- pure water	>5ppt	25	8	2	Hershner et al. (1982)
											1.61	GEOMETRIC MEAN
Rhodophyta	Porphyridiophycea e	Microalgae (Porphyridium cruentum)	Not stated	3	Chronic	EC50 (Biomass yield, Growth rate, AUC)	Synthetic salt water or filtered natural salt water	30 ± 5	20 ± 2	8.0 ± 0.1	24	USEPA (2015)
											24	GEOMETRIC MEAN
											4.8 <sup>@</sup>	VALUE USED IN SSD
Tracheophyta	Liliopsida	Macrophyte (Halodule uninervis)	Not stated	3	Acute	NOEC (Leaf length)	Filtered seawater	Not stated	25.8 ± 0.3	Not stated	87.8	Nebeker and Schuytema (1998)
											87.8	GEOMETRIC MEAN
Tracheophyta	Liliopsida	Macrophytye (Zostera marina)	Not stated	10	Chronic	LOEC (Biomass - Old	Seawater	Not stated	Not stated	Not stated	5	Chesworth et al. (2004)

						and new growth)						
											5	GEOMETRIC MEAN
Tracheophyta	Liliopsida	Macrophytye (Zostera marina)	Not stated	10	Chronic	NOEC (Biomass - Old and new growth)	Seawater	Not stated	Not stated	Not stated	2.5	Chesworth et al. (2004)
											2.5	GEOMETRIC MEAN
											2.5	VALUE USED IN SSD
Tracheophyta	Liliopsida	Macrophytye ( <i>Zostera muelleri</i> )	Not stated	3	Acute	NOEC (Leaf length)	Filtered seawater	Not stated	25.8 ± 0.3	Not stated	87.8	Nebeker and Schuytema (1998)
											87.8	GEOMETRIC MEAN

<sup>@</sup> Values were chronic EC/LC50 values that were converted to chronic NOEC/EC10 values by dividing by 5 (Warne et al. 2015). <sup>&</sup> Value was the geometric mean of chronic LOEC and EC50 values that were converted to chronic NOEC/EC10 values by 2.5 and 5, respectively (Warne et al. 2015).

# Attachment B. Distribution of sensitivities for aquatic species used in SSD

The toxicity data for diuron to all freshwater and marine species that passed the screening and quality assessment schemes were combined to create a larger dataset to determine the modality of the data. All the data that were not chronic NOEC or EC10 values were first converted to this type of data using the methods recommended by Warne et al. (2015). A natural logarithmic (In) transformation was then applied to normalise the data. Visual examination of the histogram of the transformed data indicated that the distribution of the diuron ecotoxicity data may be bimodal (Figure 3).



# Figure 3 Histogram of the natural logarithm (In) of all diuron (freshwater and marine) toxicity data for phototrophic and non-phototrophic species (n = 103).

The diuron ecotoxicity data for phototrophic and non-phototrophic species were tested to see if they came from the same population. To test for significant differences (i.e. p-value  $\leq 0.05$ ) between the two groups, the parametric two-sample *t* test was used as the transformed diuron toxicity data had equal variances (Fisher's F-Test; p = 0.551) but did not follow a normal distribution (Anderson-Darling; p < 0.000). Results from the two-sample *t* test indicated that the two groups were significantly different (p = <0.0001), therefore it can be concluded that the distribution of the diuron concentration data is bi- or multi-modal.

# Attachment C. Rationale for the selected method for deriving the Default Guidelines Values for diuron in marine waters

The order of preference of using ecotoxicity data to derive protective concentration (PC) values and/or default GVs<sup>1</sup> for diuron to marine species is:

- chronic NOEC/EC10 ecotoxicity data for phototrophs;
- chronic NOEC/EC10 and chronic estimated NOEC/EC10 values for phototrophs;
- a combination of chronic, chronic estimated and converted acute ecotoxicity data for phototrophs (Warne et al. 2015).

In total, there were chronic NOEC/EC10 data for seven phototrophic marine species (five phyla and five classes) that passed the screening and quality assessment processes. The represented phyla were Bacillariophyta, Chlorophyta, Haptophyta, Ochrophyta and Tracheoyphyta. The represented classes were Bacillariophyceae (a major grouping of diatoms), Coccolithophyceae (a grouping of marine phytoplankton), Liliopsida (monocots), Nephrophyceae (an algae grouping) and Phaeophyceae (a brown marine algae grouping). These data just meet the minimum data requirements of the SSD method (Warne et al. 2015). The SSD and PC values generated using this data are presented in Figure 4 and Table 5, respectively.



Figure 4 Cumulative frequency distribution generated using Burrlioz 2.0 (2016) of the sensitivity of the chronic ecotoxicity data (chronic 10% effect concentration (EC10) and no observed effect concentration (NOEC) data values) of the seven marine phototrophic species to diuron. Chronic NOEC/NOEL = no conversions applied; Chronic Est. NOEC = chronic LOEC/EC50 values that were converted to chronic NOEC/NOEL/EC10 values by dividing by 5 (Warne et al. 2015).

<sup>&</sup>lt;sup>1</sup> The values generated from a SSD are termed protective concentration (PC) values (as they are the concentrations that provide specific levels of protection e.g. PC99, PC95, PC90 and PC80 aim to protect 99, 95, 90 and 80 percent of species, respectively). If the PC values are the best possible then they become the proposed default GVs.

Table 5 Protective concentration values ( $\mu$ g/L) of diuron for the protection of marine ecosystems generated from the species sensitivity distribution in Figure 4.

Diuron concenti (m	ration values parine) <sup>1</sup>	Re	eliability classification <sup>2</sup>
Percent species protection	Concentration (µg/L)	Criterion	Result
99%	0.51	Sample size	7
<mark>9</mark> 5%	0.8	Type of toxicity data	Chronic EC10 and NOEC data
90%	0.98	SSD model fit	Poor
80%	1.2	Reliability	Low

<sup>1</sup> Guideline values were derived using the Burrlioz 2.0 (2016) software.

<sup>2</sup> See Warne et al. (2015) for definitions of guideline value "reliability".

The resulting PC values were considered to be of *low reliability* (Table 5) according to the schema of Warne et al. (2015) because the data set used consisted of only seven species, chronic NOEC/EC10 values were used and the cumulative distribution had a poor fit to the data (Figure 4).

Given the large confidence intervals (Figure 4) and the low reliability of the above PC values (Table 5), it was decided to use of all the available chronic NOEC/EC10 data and chronic estimated NOEC/EC10 data (derived by converting chronic LOEC and EC/LC50 data to chronic NOEC/EC10 values using the methods stated in Warne et al. (2015)) to derive a second set of PC values. This resulted in ecotoxicity data for 20 species that belonged to six phyla and 11 classes, which is a considerably larger and more representative dataset than that used to derive the PC values presented in Table 5. In the resulting SSD the fit of the distribution to the data was good (Figure 2). This combined with the type of data and the number of species represented (Table 3) resulted in the PC values being classed as having *very high reliability*. Statistical methods, including the SSD methods, become more accurate and reliable as the amount of data available to analyse increases. All these factors combined led to the recommendation that the PC values derived using both chronic and chronic estimated ecotoxicity data (Table 2) be adopted as the default GVs for diuron in marine waters.

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