Proposed EQS for Water Framework Directive Annex VIII substances: Pendimethalin *(For consultation)*

by
Water Framework Directive - United Kingdom Technical Advisory Group (WFD-UKTAG)

Publisher: **Water Framework Directive - United Kingdom Technical Advisory Group (WFD-UKTAG)**
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Scotland
www.wfduk.org

May 2012

This report is the result of research commissioned and funded by the Environment Agency and the Scotland and Northern Ireland Forum for Environmental Research (SNIFFER).

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Research performed:
2008

Dissemination Status:
Publicly available

Keywords:
Pendimethalin, Water Framework Directive, specific pollutants, predicted no-effect concentrations, freshwater, saltwater

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Environment Agency Science Project Number:
SC080021/5a(ii)
(HOEP670085)

Use of this report

The development of UK-wide classification methods and environmental standards that aim to meet the requirements of the Water Framework Directive (WFD) is being sponsored by the UK Technical Advisory Group (UKTAG) for WFD on behalf of its member and partners.

This technical document has been developed through a project managed by the Environment Agency and has involved members and partners of UKTAG. It provides background information to support the ongoing development of the standards and classification methods.

While this report is considered to represent the best available scientific information and expert opinion available at the time of its completion, it does not necessarily represent the final or policy positions of UKTAG or any of its partner agencies.

Executive summary

The UK Technical Advisory Group (UKTAG) has commissioned a programme of work to derive Environmental Quality Standards (EQSs) for substances falling under Annex VIII of the Water Framework Directive (WFD). This report proposes predicted no-effect concentrations (PNECs) for pendimethalin using the methodology described in Annex V of the Directive.

The PNECs described in this report are based on a technical assessment of the available ecotoxicity data for pendimethalin, along with any data that relate impacts under field conditions to exposure concentrations. The data have been subjected to rigorous quality assessment so that decisions are based only on scientifically sound data. Following consultation with an independent peer review group, critical data have been identified and assessment factors selected in accordance with the guidance given in Annex V of the WFD.

Where possible, PNECs have been derived for freshwater and saltwater environments, and for long-term/continuous exposure and short-term/transient exposure. If they were to be adopted as EQSs, the long-term PNEC would normally be expressed as an annual average concentration and the short-term PNEC as a 95th percentile concentration. The feasibility of implementing these PNECs as EQSs has not been considered at this stage. However, this would be an essential step before a regulatory EQS can be recommended.

Properties and fate in water

Pendimethalin is in the dinitroaniline family of chemicals and is a selective herbicide used for the control of broadleaf and grassy weeds. It acts as a microtubule disruptor on pre-emergent plants. Pendimethalin works by inhibiting the steps in plant cell division responsible for chromosome separation and cell wall formation.

Pendimethalin has low water solubility, high hydrophobicity (log Kow = 5.2) and is stable under acidic and alkaline conditions. The limited amount of data available suggests that pendimethalin dissipates rapidly out of the water column. Pendimethalin residues are tightly bound to soil and sediment particles, the degree of sorption being dependent on the presence of organic matter. Pendimethalin is expected to moderately persist in sediment as it does not partition into the aqueous phase. Some dissipation will also occur due to biodegradation and photolysis.

Bioconcentration of pendimethalin in aquatic organisms is high with Bioconcentration Factors in fish ranging from an ‘environmentally realistic’ value of 1000 (in a mesocosm study) to a ‘worst case’ value of 5100 (in a laboratory study).

Availability of data

Very limited long- and short-term laboratory data are available for four different freshwater taxonomic groups including algae, crustaceans, fish and macrophytes. Based on the limited information available all taxa would appear to be similarly sensitive to chronic exposure to pendimethalin. However, for acute exposures algae and macrophytes appear to be the most sensitive taxa. This is consistent with the use of the substance as a herbicide. For marine organisms, single species short-term toxicity data are available for four different taxonomic groups (algae, crustaceans, fish and molluscs). However, long-term toxicity data are not available for the minimum of

three saltwater taxa (algae, crustaceans and fish) required under Annex V of the Water Framework Directive. The results from freshwater mesocosm studies confirm effects on algal species at very low exposure concentrations.

All the toxicity data and resulting Predicted No Effect Concentrations (PNECs) are given as concentrations of the active ingredient.

Pendimethalin has been shown to cause fluctuations in thyroid hormones in humans and in rats. However, chronic toxicity studies in three different animal species did not induce any oestrogenic or treatment-related effects on any other component of the endocrine system. *In vitro* assays using MCF-7 cells suggest that pendimethalin is a partial oestrogen receptor agonist.

Derivation of PNECs

Long-term PNEC for freshwaters

The lowest valid long-term toxicity value is for freshwater algae where a 5-day NOEC of 3.0 μg l^{-1} for effects on growth inhibition (using the growth rate endpoint) of the green alga *Pseudokirchneriella subcapitata* has been reported in the EU DAR (2003). Reliable long-term NOECs are available for algae, invertebrates and fish, and therefore an assessment factor of 10 can be applied resulting in a $PNEC_{freshwater\_lt}$ of 0.3 μg l^{-1}.

This value is lower than the previously proposed tentative non-statutory EQS of 1.5 μg l^{-1} which was derived by applying an assessment factor of 2 to the algal data.

Short-term PNEC for freshwaters

Reliable short-term data are available for algal, invertebrate and fish species. The lowest valid short-term toxicity value is a 5-day growth inhibition (using the growth rate endpoint) EC50 values of 5.8 μg l^{-1} for the diatom *Navicula pelliculosa* reported in the EU DAR (2003). The short-term toxicity database for freshwater organisms is not extensive, but does adequately indicate that algae and macrophytes are the most sensitive taxa to short-term exposure to pendimethalin. An assessment factor of 10 was therefore applied, resulting in a $PNEC_{freshwater\_st}$ of 0.58 μg l^{-1}.

This value is lower than the previously proposed tentative non-statutory EQS of 6.0 μg l^{-1} which was based on the EC50s for growth of 5.8 and 6.7 μg l^{-1} reported for the algae *Navicula pelliculosa* and *Pseudokirchneriella subcapitata*, respectively, without the application of an assessment factor.

Long-term PNEC for saltwaters

Since long-term single species toxicity data are only available for algae, a combined freshwater and saltwater dataset for the saltwater effects assessment was used to derive the PNEC. The most sensitive result is a 5-day growth inhibition (using the growth rate endpoint) NOEC for the diatom, *Skeletonema costatum*, of 2.7 μg l^{-1} reported in the EU DAR (2003). The combined dataset indicate that algae are evidently the most sensitive taxonomic group in both freshwater and saltwater with comparable species showing similar sensitivities. Therefore, it is recommended that the lowest marine toxicity value is used along with an assessment factor of 10 (but without the application of an additional assessment factor to account for the absence of data for groups such as echinoderms) as alga are thought to be the most sensitive taxa. This results in a $PNEC_{saltwater\_lt}$ of 0.27 μg l^{-1} which is essentially the same as the freshwater PNEC.

This value is lower than the previously proposed tentative non-statutory EQS of 1.5 μg l^{-1} which was based on the long-term PNEC for freshwaters because of insufficient marine data to derive an EQS.

Short-term PNEC for saltwaters

Reliable short-term saltwater toxicity data are available for algae, invertebrates and fish. The most sensitive result is a 5-day growth inhibition (using the growth rate endpoint) EC50 for the diatom, *Skeletonema costatum*, of 5.2 µg l^{-1} reported in the EU DAR (2003), which is consistent with toxicity values for freshwater species. The combined dataset indicate that algae are evidently the most sensitive taxonomic group in both freshwater and saltwater with comparable species showing similar sensitivities. Therefore, it is recommended that the lowest toxicity value for marine algae is adopted along with an assessment factor of 10 (but without the application of an additional assessment factor to account for the absence of data for groups such as echinoderms) as alga are thought to be the most sensitive taxa. This results in a $PNEC_{saltwater\_lt}$ of 0.52 µg l^{-1} which is consistent with the freshwater PNEC.

This value is more stringent than the previously proposed tentative non-statutory EQS of 6.0 µg l^{-1} which was also based on the short-term PNEC for freshwaters because of insufficient marine data to set an EQS.

PNEC for sediments

The TGD trigger value of a log Koc or log Kow of ≥3 is met, as the reported log Kow is 5.2 and reported Koc values range from 6700 to 29400. There are no reliable experimental data on sediment toxicity for pendimethalin and, therefore, no $PNEC_{sediment}$ can be derived.

PNEC for secondary poisoning

Bioconcentration data (as BCF values) for pendimethalin for fish range from 1000 (in a mesocosm study) to 5100 (in a laboratory study), hence the trigger of a BCF >100 is exceeded and derivation of PNECs for secondary poisoning of predators is required. The lowest relevant $NOEC_{food}$ is 50 mg kg^{-1} derived by extrapolation from a LOEC of 100 mg kg^{-1} from a 13-week study with rats.

Bioconcentration of pendimethalin in aquatic organisms is high with Bioconcentration Factors in fish ranging from 1000 (in a mesocosm study) to 5100 (in a laboratory study). The mesocosm study, which used a single dose and was carried out to GLP, was considered by the EU DAR (2003) to be valid and suitable to modify the BCF in fish and invertebrates to around 1000 with a clearance time of 5.1 days (>95% in 5 days) and to indicate a low potential for food chain biomagnification. However, there is a possibility that pendimethalin could be released on a more continuous basis and as a precautionary measure, it would be more appropriate to use the BCF of 5100 in the calculation of the $PNEC_{secpois.water}$.

Using the reported BCF of 5100 from the laboratory study in the US EPA RED (1997) for the calculation results in a corresponding water concentration of $PNEC_{secpois.water}$ = 0.55 mg kg^{-1} prey / BCF (5100) = 0.1 µg pendimethalin l^{-1}.

This concentration is lower than the proposed long-term PNEC for the protection of freshwater and saltwater organisms (i.e. 0.27 µg l^{-1}). Therefore, if quality standards are set on the basis of the proposed long-term water column it is probable that predators would not be protected from secondary poisoning.

Summary of proposed PNECs

Receiving medium/exposure scenario	Proposed PNEC ($\mu g\ l^{-1}$)	Existing EQS* ($\mu g\ l^{-1}$)
Freshwater/long-term	0.3	1.5
Freshwater/short-term	0.58	6.0
Saltwater/long-term	0.27	1.5
Saltwater/short-term	0.52	6.0
Sediments	Insufficient data	-
Secondary poisoning	0.1	-

*Tentative

Analysis

For water, the lowest proposed PNEC derived for pendimethalin is 0.1 $\mu g\ l^{-1}$. The data quality requirements are that, at a third of the EQS, total error of measurement should not exceed 50%. Using this criterion, it is evident that current analytical methodologies using solid phase extraction followed by gas chromatography-mass spectrometry, should offer adequate performance to analyse for pendimethalin.

Implementation issues

Based on consideration of the information collated within the report and the proposed PNECs the following comments are made re: implementation:-

- Current analytical methods should be adequate for compliance assessment.
- The PNECs derived are not subject to excessive uncertainty with assessment factors of 10 being applied in their derivation.
- Pendimethalin adsorbs rapidly to sediment and therefore its potential impact on sediment dwelling organisms will need to be considered as more data becomes available.

Contents

1 Introduction

The UK Technical Advisory Group (UKTAG) supporting the implementation of the Water Framework Directive (2000/60/EC)[1] is a partnership of UK environmental and conservation agencies. It also includes partners from the Republic of Ireland. UKTAG has commissioned a programme of work to derive Environmental Quality Standards (EQSs) for substances falling under Annex VIII of the Water Framework Directive (WFD). This report proposes predicted no-effect concentrations (PNECs) for pendimethalin using the methodology described in Annex V of the Directive.

The PNECs described in this report are based on a technical assessment of the available ecotoxicity data for pendimethalin, along with any data that relate impacts under field conditions to exposure concentrations. The data have been subjected to rigorous quality assessment so that decisions are based only on scientifically sound data.[2] Following consultation with an independent peer review group, critical data have been identified and assessment factors selected in accordance with the guidance given in Annex V of the WFD. The feasibility of implementing these PNECs as EQSs has not been considered at this stage. However, this would be an essential step before a regulatory EQS can be recommended.

1.1 Properties and fate in water

Pendimethalin is in the dinitroaniline family of chemicals and is a selective herbicide used for the control of broadleaf and grassy weeds. Pendimethalin acts as a microtubule disruptor on pre-emergent plants. It works by inhibiting the steps in plant cell division responsible for chromosome separation and cell wall formation. Pendimethalin is used in various formulations in terrestrial systems.

Pendimethalin has low water solubility, high hydrophobicity (log Kow = 5.2) and is stable under acidic and alkaline conditions. The limited amount of data available suggests that pendimethalin dissipates rapidly out of the water column. Pendimethalin residues are tightly bound to soil and sediment particles, the degree of sorption being dependent on the presence of organic matter. Pendimethalin is expected to moderately persist in sediment as it does not partition into the aqueous phase. Some dissipation will also be due to biodegradation and photolysis.

Bioconcentration of pendimethalin in aquatic organisms is high with Bioconcentration Factors in fish ranging from an ‘environmentally realistic’ value of 1000 (in a mesocosm study) to ‘worst case’ value of 5100 (in a laboratory study).

[1] *Official Journal of the European Communities* L327:1–72 (22/12/2000). Can be downloaded from http://www.eu.int/comm/environment/water/water-framework/index_en.html

[2] Data quality assessment sheets are provided in Annex I.

2 Results and observations

2.1 Identity of substance

Table 2.1 gives the chemical name and Chemical Abstracts Service (CAS) number for pendimethalin.

Table 2.1 Substance covered by this report

Name	CAS Number
Pendimethalin	40487-42-1

2.2 PNECs proposed for derivation of quality standards

Table 2.2 lists proposed PNECs obtained using the methodology described in the Technical Guidance Document (TGD) issued by the European Chemicals Bureau (ECB) on risk assessment of chemical substances (ECB 2003). *(NB: EU technical guidance for the derivation of EQSs (2011) has been produced since the finalisation of this report but used the same principles as the TGD)*

Section 2.6 summarises the effects data identified from the literature for pendimethalin. The use of these data to derive the values given in Table 2.2 is explained in Section 3.

Table 2.2 Proposed overall PNECs as basis for quality standard setting

PNEC	TGD deterministic approach (AFs)	TGD probabilistic approach (SSDs)	Existing EQS (Tentative)
Freshwater short-term	0.58 µg l^{-1}	–	6.0 µg l^{-1} (MAC)
Freshwater long-term	0.3 µg l^{-1}	–	1.5 µg l^{-1} (AA)
Saltwater short-term	0.52 µg l^{-1}	–	6.0 µg l^{-1} (MAC)
Saltwater long-term	0.27 µg l^{-1}	–	1.5 µg l^{-1} (AA)
Sediment	Insufficient data	–	-
Secondary poisoning	0.1 µg l^{-1}	–	-

AA = Annual Average
AF = Assessment Factor
MAC = Maximum Allowable Concentration
SSD = Species Sensitivity Distribution
TGD = Technical Guidance Document

2.3 Hazard classification

Table 2.3 gives the R-phrases (Risk-phrases) and labelling for pendimethalin.

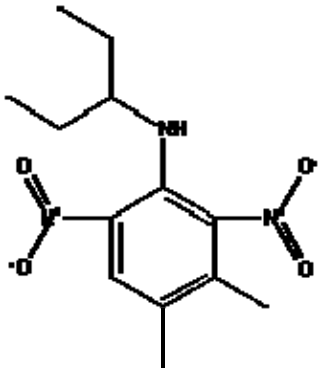
Table 2.3 Hazard classification

R-phrases and labelling	Reference
R43, R50/53 S2, 24, 29, 37,60, 61	ECB 2005

2.4 Physical and chemical properties

Table 2.4 summarises the physical and chemical properties of pendimethalin.

Table 2.4 Physical and chemical properties of pendimethalin

Property	Reference	Values
CAS number	ECB 2005	40487-42-1
Substance name	ECB 2005	N-(1-ethylpropyl)-2,6-dinitro-3,4-xylidine
Molecular formula	HSDB 2006	$C_{13}H_{19}N_3O_4$
Molecular structure	Chemfinder 2005	
Molecular weight	EU DAR 2003 HSDB 2006	281.3
Colour/form	EU DAR 2003 HSDB 2006	Orange-yellow crystalline solid Orange-brown solid
Odour	HSDB 2006	Fruit like
Melting point (°C)	US EPA RED1997 HSDB 2006	54-58°C 56-57°C
Boiling point (°C)	EU DAR 2003 HSDB 2006	330°C
Vapour pressure	EU DAR 2003 HSDB 2006	3×10^{-5} mm Hg at 25°C 1.94×10^{-3} Pa at 25°C
Density/ specific gravity	EU DAR 2003 HSDB 2006	1.19 at 25°C 0.85 g ml^{-1} at 25°C
Henry's Law constant	US EPA RED 1997 HSDB 2006 EU DAR 2003	2.2×10^{-5} atm m^3 mol^{-1} $8.6 \pm 5.4 \times 10^{-7}$ atm m^3 mol^{-1} 2.728×10^{-3} Kpa x m^3 mol^{-1} at 25°C

Property	Reference	Values
Water solubility	EU DAR 2003	0.54 mg l^{-1} at 20°C, pH 4 0.33 mg l^{-1} at 20°C, pH 7 0.44 mg l^{-1} at 20°C, pH 10
	HSDB 2006	0.3 mg l^{-1} at 20°C
Solubility in organic solvents	HSDB 2006	700 g l^{-1} at 26°C in acetone 628 g l^{-1} at 26°C in xylene 77 g l^{-1} at 26°C in isopropanol Readily soluble in benzene, toluene, dichloromethane

2.5 Environmental fate and partitioning

Table 2.5 summarises information obtained from the literature on the environmental fate and partitioning of pendimethalin.

Two laboratories simultaneously studied the biodegradation of radio-labelled pendimethalin in test systems simulating pelagic and sediment/freshwater habitats. The two laboratories observed different disappearance of pendimethalin in the pelagic systems: 60 – 70% after 100 days versus 0 – 40%. This was partly attributed to the different test concentrations with the highest degree of disappearance observed at the highest test concentration, and that pendimethalin serves as a carbon and energy source for the micro-organisms. The results from the water/sediment test systems were more similar – with disappearances of 30 – 40% and 0 – 35% after 100 days incubation, with the majority of the degradation taking place within the first 10 – 20 days (Rasmussen *et al.* 2002).

Pendimethalin was found to degrade rapidly in a model ecosystem. Isensee and Dubey (1983) studied the distribution of ^{14}C-labelled pendimethalin among sediment, water and aquatic organisms. Between 45-59% of pendimethalin was bound to sediment (Metapeake silt loam, pH 5.3; 0.87% organic matter; 38.4% sand; 49.4% silt, and 12.2% clay) by 30 days. The maximum concentration in water was obtained at 13 days, after which it remained stable at 11-13%. TLC analysis indicated that after 2 days only approximately 30% of the tagged chemical was found to be pendimethalin; this decreased to less than 1% after 30 days. Polar metabolites measured in the water column reached 54% by 2 days and >90% by 9-30 days. A total of 68% of soil residues was un-extractable and assumed to be bound.

Anaerobic aquatic metabolism half-lives range from 6 – 105 days (USEPA RED 1997). A half-life of 6 days was calculated for non-sterilized sandy loam pond sediment from Ontario, Canada (1.8% organic carbon) incubated at 25°C for 39 days under anaerobic conditions. In all conditions (5°C sterile and non-sterile and 25°C sterile and non-sterile) aqueous residues decreased rapidly to form soil-extractable residues, followed by a rapid increase in soil-bound residues. At both temperatures under non-sterile conditions aqueous residues declined to about 11.8% by 16 days and remained constant until the end of the study (39 days) (USEPA RED 1997). The half-life of 105 days was calculated in silt loam soil (1.2% organic carbon) that was incubated for 1-week under aerobic conditions and for 8-weeks under anaerobic conditions (USEPA RED 1997).

Helweg *et al.* (2003) conducted a field experiment in artificial ponds by spraying the pond water surface with pendimethalin and following the development in concentrations in the water body, at different depths, and in the sediment. The ponds were either well developed with both flora and fauna or macrophyte free. The dissipation half-life from the water phase was 1 – 2 days. Ponds with macrophytes initially had a higher concentration in the water phase whereas, after day 1, they had a lower concentration in the water phase compared to macrophyte-free ponds. Two

days after spraying a vertical concentration gradient could still be measured in the ponds. Concentrations of pendimethalin in the sediment on days 2, 15 and 31 after spraying were higher in ponds without macrophytes than in ponds with macrophytes. This was considered to be due to the reduction in vertical mixing and thus reduced transport from surface to bottom caused by the presence of macrophytes and the sorption of pendimethalin to macrophyte surfaces.

In summary pendimethalin can be expected to quickly dissipate out of the water column and become bound to sediment and other particulate matter. The degree of sorption is dependent on the presence of organic matter. Pendimethalin is expected to moderately persist in sediment as it does not readily desorb into the aqueous phase. Some dissipation will also be due to biodegradation and photolysis.

Bioconcentration of pendimethalin in aquatic organisms is high with Bioconcentration Factors in fish ranging from 1000 (in a mesocosm study) to 5100 (in a laboratory study).

Table 2.5 Environmental fate and partitioning of pendimethalin

Property	Values
Abiotic fate	Vapour-phase pendimethalin reacts with photochemically produced hydroxyl radicals with a rate constant of 3.03 x 10^{-11} cm^3 $molecule^{-1}$ sec^{-1}. Assuming a hydroxyl radical concentration of 5 x 10^5 radicals cm^3, the estimated half-life of pendimethalin in the atmosphere would be 12.7 h (Meylan and Howard, 1993).
Hydrolytic stability	Pendimethalin did not degrade in sterile aqueous buffer solutions (pH 5, 7, and 9) incubated in darkness at 25°C for 30 d (USEPA RED 1997). Stable from pH 4 to 9 (> 5 d at 50°C) (EU DAR 2003)
Photostability	Calculated half-lives were 16.5 d at pH 5, 7 and 9 and 21 d at pH 7 (USEPA RED 1997).
Volatilisation	Volatilisation from well-mixed surface waters may be an important transport process because greater volatilisation was observed under moist field conditions. Both the reported vapour pressure and Henry's Law Constant support the conclusion that volatility is a significant transport mechanism (USEPA RED 1997).
Distribution in water/sediment systems	Very rapid binding to sediment. Between 2 – 20% in the water phase: sediment – up to 84% at 0 d and 9.5% at 197 d (EU DAR 2003).
Degradation in soil	Pendimethalin is relatively stable and immobile in soil; DT50 (aerobic soil metabolism) ~ 172 d. Pendimethalin persists with decreased temperatures, decreased moisture and increased soil organic matter. Neither soil photolysis nor anaerobic metabolism are major degradation processes (USEPA RED 1997). Mineralization after 100 d – 1.7 – 2.4% (EU DAR2003).
Biodegradation in water	Not readily biodegradable (EC 2003). Calculated half-lives were 10-11 d in water containing different soil fungi and 354 d in sterile water (USEPA RED 1997).
Octanol–water coefficient (log Kow)	5.18 (SRC 2005) 5.2 at pH 7 (EU DAR 2003)
Log Koc	3.83 – 4.47 (EU DAR 2003)
Dissociation constant pKa	2.8 (EU DAR 2003)
Bioaccumulation BCF values	Pendimethalin accumulates readily in bluegill sunfish with BCF factors of 1400 in edible tissue, 5800 in non edible tissue and 5100 in whole fish. Depuration was rapid (US EPA RED 1997)[see 3.5.2]. BCF of 1000 (in fish and *Daphnia* in a mesocosm study [see 2.6.6]). Mesocosm study showed no potential for biomagnification. Elimination half-life (CT) = 5.1 days (EU DAR 2003)

BCF = Bioconcentration factor

2.6 Effects data

A summary of the mode of action of this substance can be found in Section 2.6.5.

Data collation followed a tiered approach. First, critical freshwater and saltwater data were compiled from existing EQS documents. Further data published after derivation of the current, tentative UK EQSs were then retrieved from the US Environmental Protection Agency (US EPA) ECOTOX database.[3]

Further data sources used included:

- ScienceDirect®;[4]
- Hazardous Substances Data Bank (HSDB®) database of the US National Library of Medicine;[5]
- US EPA Re-registration Eligibility Report (RED) for pendimethalin (US EPA 1997 – referred to in this report as USEPA RED 1997);
- European Commission Draft Assessment Report (Public Version) on pendimethalin prepared under the Plant Protection Products Directive 91/414/EEC (EU DAR 2003);
- OPP Pesticide Ecotoxicity Database - an online US EPA database held by the Office of Pesticide Programs that summarises ecotoxicological data used by the EPA for ecotoxicological assessments. This consists primarily of the endpoint data submitted in support of registration and re-registration of pesticide products (OPP 2007)[6].

Many of the most sensitive toxicity results are reported in unpublished studies carried out in support of pesticide registration. As only summary information on the tests is provided in compendium reports the Pesticide Safety Directorate was asked to provide additional information where possible to confirm the robustness of the key data used in the derivation of the freshwater and saltwater PNECs. Relevant additional extracts from the Draft Assessment Report (EU DAR 2003) prepared by the Rapporteur Member State (RMS) (Spain) for pendimethalin under the Directive 91/414/EEC Review process were made available as part of the assessment process.

The key data were also reported in the US EPA OPP Pesticide Ecotoxicity Database although the toxicity values for certain species were slightly different which was probably the result of re-interpretation of the data and/or re-calculation of toxicity values.

All concentrations of pendimethalin in this report are expressed as active ingredient and all key data were checked for accuracy as far as practicable using the available data. All PNEC values have been derived from studies using technical grade material.

2.6.1 Toxicity to freshwater organisms

Freshwater toxicity data on pendimethalin are available for various taxonomic groups including algae, invertebrates and fish as required for the application of the approach specified in the EU Technical Guidance Document (TGD) (ECB 2003). Long-term data are available for four taxonomic groups including algae, crustaceans, fish and macrophytes. Freshwater short-term toxicity data are available for the same four taxonomic groups (Table 2.6)

[3] http://www.epa.gov/ecotox/
[4] http://www.sciencedirect.com/
[5] http://toxnet.nlm.nih.gov/cgi-bin/sis/htmlgen?HSDB
[6] Http://www.ipmcenters.org/Ecotox/index.cfm

Table 2.6 Summary of available freshwater data for pendimethalin

Type of data	Taxonomic groups for which information is available
Long-term	Algae, crustaceans, fish and macrophytes
Short-term	Algae, crustaceans, fish and macrophytes

Based on the limited information available all taxa would appear to be of similar sensitivity to chronic exposure to pendimethalin. However, algae and macrophytes appear to be the most sensitive taxonomic groups in acute exposures. This is consistent with the use of the substance as a herbicide.

The data were also evaluated to assess whether differences in toxicity were due to different physical formulation effects (e.g., the use of the same chemical formulation but including either technical grade material or an emulsifiable concentrate). For all the data, with the exception of crustaceans, the majority of species showed relatively small differences in response after exposure to formulations or technical material, expressed as active ingredient (a.i.). It should be noted that the algal data are critical to the PNEC derivation and here the difference is just greater than 1. Theoretically, a lower toxicity when formulated indicates a lower bioavailability, whereas a higher toxicity value may indicate either a higher bioavailability or a contribution to toxicity from the formulating agents. The critical values for setting pendimethalin PNECs are all derived from studies on technical material. A comparison of the acute toxicity of pendimethalin and pendimethalin formulations to selected taxa is shown in Table 2.7.

Table 2.7 Comparison of acute toxicity to different taxa of pendimethalin and pendimethalin formulations

Species	Endpoint	Value (µg a.i. l^{-1})		
		Technical grade material	Formulation	Reference
Pseudokirchneriella subcapitata	72 h EC50	52	38	Bražėnaitė and Šakalienė (2006)
Lemna minor	7 d EC50	634	280	Cedergreen and Streibig 2005
Daphnia magna	48 h EC50	280	2325	OPP 2007
Onchorhynchus mykiss	96 h EC50	138	234	OPP 2007

Diagrammatic representations of the available freshwater data (cumulative distribution functions) for pendimethalin are presented in Figure 2.1 for long-term data and Figure 2.2 for short-term data.

These diagrams include all data regardless of quality and provide an overview of the spread of the available data. However, they are not species sensitivity distributions and have not been used to derive pendimethalin PNECs.

The lowest critical freshwater data are presented in Table 2.8 for long-term toxicity data and Table 2.9 for short-term toxicity data

These tables do not contain all the available toxicity data but only those which are considered most relevant to the derivation of PNECs.

The data in Tables 2.8 and 2.9 indicate that the toxicity of pendimethalin and its formulations occurs over a fairly limited concentration range.

Figure 2.1 Cumulative distribution function of freshwater long-term data (µg a.i. l^{-1}) for pendimethalin

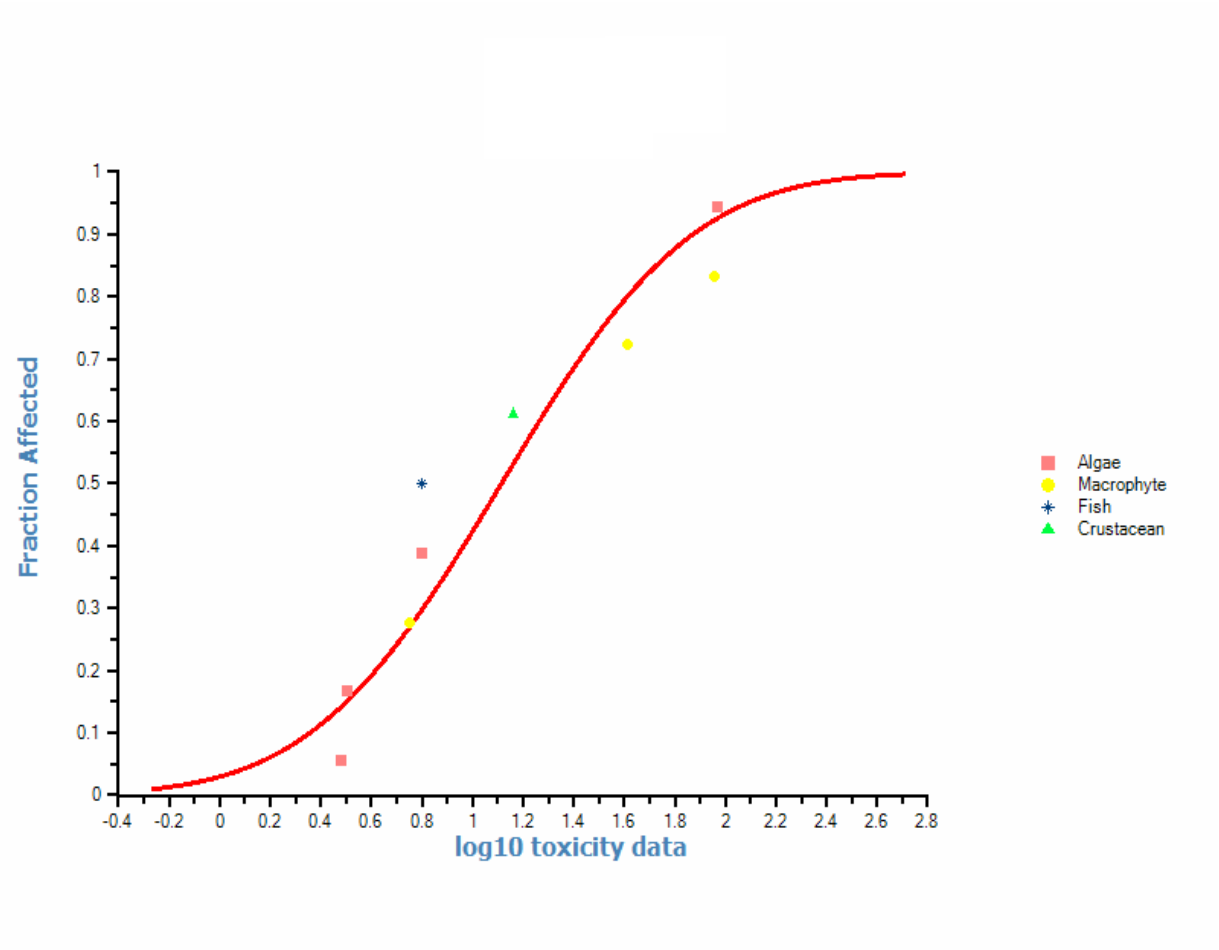


Figure 2.2 Cumulative distribution function of freshwater short-term data (μg a.i. l^{-1}) for pendimethalin

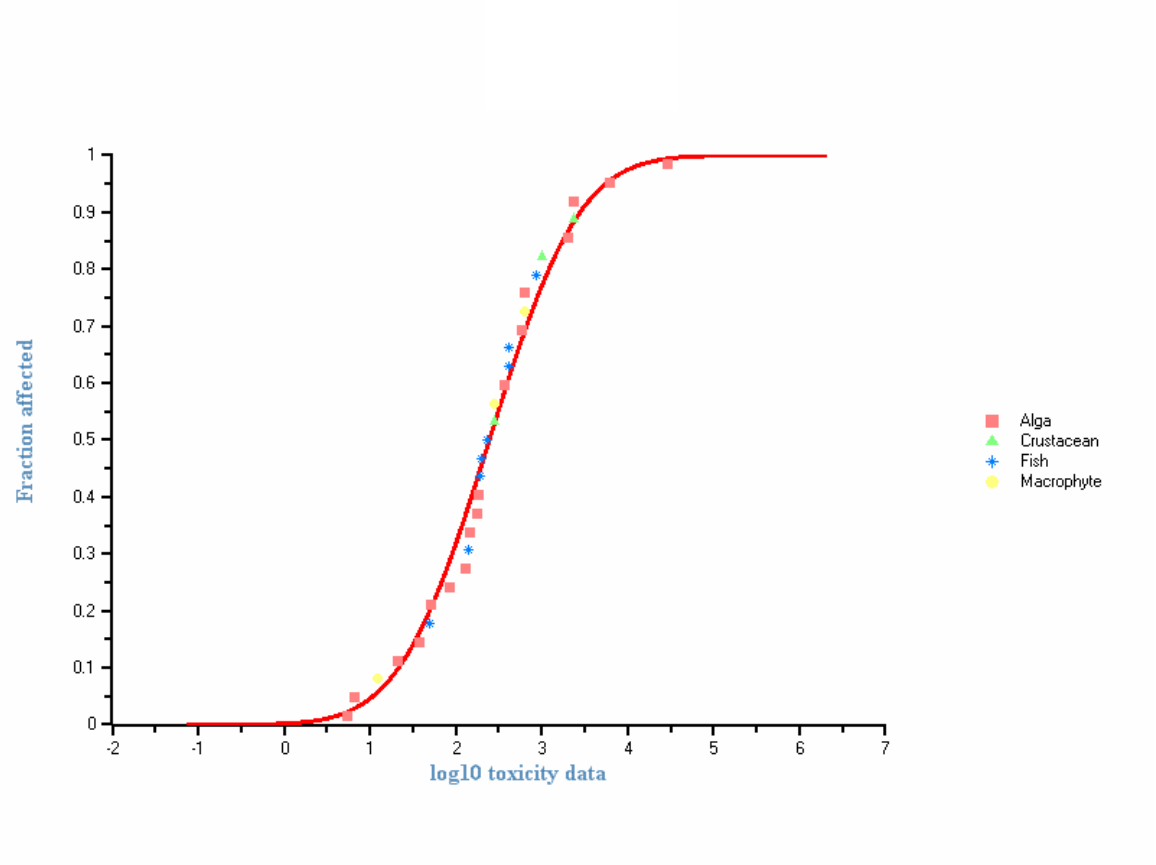


Table 2.8 Most sensitive long-term aquatic toxicity data for freshwater organisms exposed to pendimethalin

Chemical formulation (% purity)	Scientific name	Common name	Taxonomic group	Effect	Endpoint	Test duration	Conc. (μg a.i. l^{-1})	Exposure[1]	Toxicant analysis[2]	Comments	Reliability (Klimisch Code*)	Reference
Algae												
Technical grade (92.98%)	*Pseudokirchneriella subcapitata* (formerly *Selenastrum capricornutum*)	Green alga	ALG	Growth inhibition (growth rate)	NOEC	5 days	3.0	s	y	GLP study	1 (c)	Hughes *et al.* (1992a) cited in EU DAR (2003)
Technical grade (92.98%)	*Navicula pelliculosa*	Diatom	ALG	Growth inhibition (growth rate)	NOEC	5 days	3.2	s	y	GLP study	1 (c)	Hughes *et al.* (1992b) cited in EU DAR (2003
Stomp (33% a.i.)	*Pseudokirchneriella subcapitata*	Green alga	ALG	Growth inhibition (growth rate)	NOEC	3 days	6.3	s	y	GLP study	1	Barker *et al.* (2000) cited EU DAR (2003)
Higher plants												
Technical grade (92.8%)	*Lemna gibba*	Duckweed	MAC	Growth inhibition	NOEC	14 days	5.6	s	y	GLP study	1 (c)	Hughes *et al.* (1991) cited in EU DAR (2003)
Invertebrates												
Technical grade (92.2%)	*Daphnia magna*	Water flea	CRU	Reproduction	NOEC	21 days	14.5	ss	y	GLP study	1 (c)	Graney (1981) cited in EU DAR (2003)
Fish												
Technical grade (98.3%)	*Pimephales promelas*	Fathead minnow	FIS	Reproduction	NOEC	288 days	6.3	f	y	GLP study	1 (c)	EG & G Bionomics (1975) cited in EU DAR (2003)

* See Annex I and Annex II for explanation,
[1] Exposure: s = static; ss = semi-static, f = flow-through
[2] Toxicant analysis: y = measured
ALG = alga, CRU = crustacean, FIS = fish, MAC = macrophyte
NOEC = no observed effect concentration
c = Core study from US OPP 2007 (see Annex II)

Table 2.9 Most sensitive short-term aquatic toxicity data for freshwater organisms exposed to pendimethalin

Chemical formulation (% purity)	Scientific name	Common name	Taxonomic group	Effect	Endpoint	Test duration	Conc. (μg a.i. l^{-1})	Exposure[1]	Toxicant analysis[2]	Comments	Reliability (Klimisch Code*)	Reference
Algae												
Technical grade (92.98%)	*Navicula pelliculosa*	Diatom	ALG	Growth inhibition (Growth rate)	EC50	120 hours	5.8	s	y	GLP study	1 (c)	Hughes *et al*. (1992a) cited in EU DAR (2003)
Technical grade (92.98%)	*Pseudokirchneriella subcapitata* (formerly *Selenastrum capricornutum*)	Green alga	ALG	Growth inhibition (Growth rate)	EC50	120 hours	6.7	s	y	GLP study	1 (c)	Hughes *et al*. (1992b) cited in EU DAR (2003
Stomp (33% a.i.)	*Pseudokirchneriella subcapitata*	Green alga	ALG	Growth inhibition	EC50	72 hours	33.7	s	y	GLP study	1 (c)	Barker *et al.* (2000) cited EU DAR (2003)
Higher plants												
Technical grade (92.98%)	*Lemna gibba*	Duckweed	MAC	Growth inhibition	EC50	14 days	12.5	s	y	GLP study	1 (c)	Hughes *et al* (1991) cited in EU DAR (2003),
Stomp 40% a.i.	*Lemna minor*	Duckweed	MAC	Growth inhibition	EC50	168 hours	280	s	n	T=23°C	2	Cedergreen and Striebig (2005)
Invertebrates												
Technical grade (93.2%)	*Daphnia magna*	Water flea	CRU	Immobility	EC50	48 hours	280	s	y	GLP study	1 (c)	Forbis *et al* (1985) cited in EU DAR (2003)
Technical grade (94.2%)	*Procambarus simulans*	Crayfish	CRU	Survival	LC50	96 hours	1,000	s	y	GLP study	1 (c)	Thompson *et al* (1980) cited in EU DAR
Vertebrates (fish and amphibians)												
Technical grade (45% a.i.)	*Oncorhynchus mykiss*	Rainbow trout	FIS	Survival	LC50	96 hours	50	-	u		4	ECOTOX (2007) (Japanese/English abstract)

Chemical formulation (% purity)	Scientific name	Common name	Taxonomic group	Effect	Endpoint	Test duration	Conc. (μg a.i. l^{-1})	Exposure[1]	Toxicant analysis[2]	Comments	Reliability (Klimisch Code*)	Reference
Technical grade (93.2%)	*Oncorhynchus mykiss*	Rainbow trout	FIS	Survival	LC50	96 hours	138	s	y	GLP study	1 (c)	Sleight (1973) cited in EU DAR (2003)
Not stated	*Pimephales promelas*	Fathead minnow	FIS	Survival	LC50	96 hours	190 (170-220)	f	y	Temperature = 25.1°C, pH = 7.8, Hardness = 43.5 mg $CaCO_3$ l^{-1}, DO = 7.4 mg l^{-1}	4	Geiger, Call *et al*. (Eds) (1990) Cited HSDB (2006), OPP (2007)
Technical grade (93.2%)	*Lepomis macrochirus*	Bluegill sunfish	FIS	Survival	LC50	96 hours	199	s	y		1 (c)	Sleight (1973) cited in EU DAR (2003)

* Annex I and Annex II for explanation,
[1] Exposure: s = static; f = flow-through
[2] Toxicant analysis: y = measured; n = not measured; u = unknown
ALG = alga, CRU = crustacean, FIS = fish, MAC = macrophyte
EC50 = concentration resulting in a 50% effect, LC50 = concentration resulting in 50% mortality
c = Core study from US OPP 2007 (see Annex II)

2.6.2 Toxicity to saltwater organisms

Single species short-term toxicity data for pendimethalin for saltwater organisms are available for four different taxonomic groups: algae, crustaceans, fish and molluscs. Only one long-term toxicity datum is available, which is for the marine diatom *Skeletonema costatum* (Table 2.10). Short-term toxicity data are summarised in Table 2.11.

A diagrammatic representation of all the available short-term saltwater data (cumulative distribution function) for pendimethalin is presented in Figure 2.3. This diagram includes all data regardless of quality and provides an overview of the spread of the available data. The diagram is not a species sensitivity distribution and has not been used to set pendimethalin PNECs.

Figure 2.3 Cumulative distribution function of saltwater short-term data (μg a.i. l^{-1}) for pendimethalin

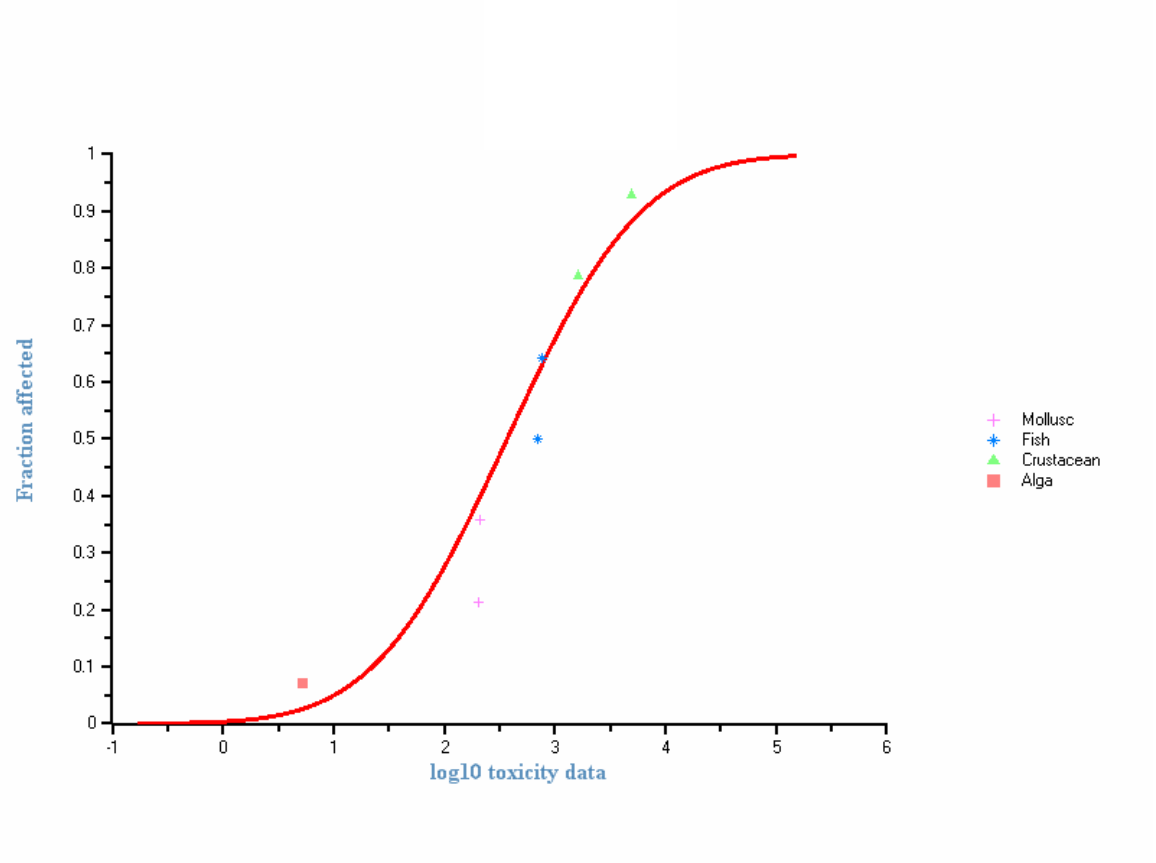


Table 2.10 Most sensitive long-term aquatic toxicity data for saltwater organisms exposed to pendimethalin

Chemical formulation (% purity)	Scientific name	Common name	Taxonomic group	Effect	Endpoint	Test duration	Conc. (μg a.i. l^{-1})	Exposure[1]	Toxicant analysis[2]	Comments	Reliability (Klimisch Code*)	Reference
Algae												
Technical grade (92.98%)	*Skeletonema costatum*	Diatom	ALG	Growth inhibition (Growth)	NOEC	5 days	2.7	s	y	GLP study	1 (c)	Hughes *et al.* (1992c) cited in EU DAR (2003)

* Annex I and Annex II for explanation,
[1] Exposure: s = static
[2] Toxicant analysis: y = measured
ALG = alga,
NOEC = no observed effect concentration
c = Core study from US OPP 2007 (see Annex II)

Table 2.11 Most sensitive short-term aquatic toxicity data for saltwater organisms exposed to pendimethalin

Chemical formulation	Scientific name	Common name	Taxonomic group	Effect	Endpoint	Test duration	Conc. (μg a.i. l^{-1})	Exposure[1]	Toxicant analysis[2]	Comments	Reliability (Klimisch Code*)	Reference
Algae												
Technical grade (92.98%)	*Skeletonema costatum*	Diatom	ALG	Growth inhibition (Growth)	EC50	120 hours	5.2	s	y	GLP study	1 (c)	Hughes *et al* (1992c) cited in EU DAR (2003)
Invertebrates												
Technical grade (45%)	*Crassostrea virginica*	Eastern oyster	MOL	Development	EC50	48 hours	202.5	s	u	-	4 (c)	USEPA RED (1997), OPP (2007)
Technical grade (92.2%)	*Crassostrea virginica*	Eastern oyster	MOL	Development	EC50	48 hours	210	s	u	-	4 (c)	USEPA RED (1997), OPP (2007)
Technical grade (92.2%)	*Penaeus duorum*	Pink shrimp	CRU	Survival	LC50	96 hours	1,600	s	u	-	4 (c)	USEPA RED (1997), OPP (2007)
Fish												
Technical grade (92.2%)	*Cyprinodon variegatus*	Sheepshead minnow	FIS	Survival	LC50	96 hours	707	s	u	-	4 (c)	USEPA RED (1997), OPP (2007)
Technical grade (45%)	*Cyprinodon variegatus*	Sheepshead minnow	FIS	Survival	LC50	96 hours	765	s	u	-	4 (c)	USEPA RED (1997), OPP (2007)

* Annex I and Annex II for explanation,
[1] Exposure: s = static
[2] Toxicant analysis: y = measured, u = unknown
ALG = alga, CRU = crustacean, FIS = fish, MOL = molluscs
EC50 = concentration resulting in a 50% effect, LC50 = concentration resulting in 50% mortality
c = Core study from US OPP 2007 (see Annex II)

2.6.3 Toxicity to sediment dwelling organisms

Only one study on the direct toxicity of pendimethalin to sediment-dwelling organisms was found in the open literature. The objectives of this study were to estimate the LC50 for water concentrations and the critical tissue concentrations for three herbicides, one of which was pendimethalin, and then to determine the subchronic toxicity of sediment-associated pendimethalin to 4th instar *Chironomus riparius*, a sediment-dwelling midge (Mäenpää *et al.* 2003). The authors reported that they could not achieve mortality in the water-only exposures in the range of the substance's water solubility, nor was any mortality seen in sediment spiked with high concentrations of pendimethalin (> 10 mg g^{-1} dw).

The EU DAR (2003) reports a NOEC of 138 µg l^{-1} from a 30-day long-term toxicity test with *C. riparius*. No other details are available and the EU DAR (2003) considers this result to be of poor quality. Therefore, it is not appropriate to use the data to derive a sediment PNEC.

2.6.4 Endocrine-disrupting effects

The list of purported endocrine disruptors, compiled by the Institute of Environment and Health (IEH 2005), lists pendimethalin as a group III substance. This categorisation covers chemicals for which it is considered there is insufficient evidence of endocrine disruption or for which there is only low concern with regard to exposure.

Pendimethalin is known to affect the pituitary-thyroid-axis in humans which means that this substance may be a potential endocrine disruptor. Although slight fluctuations in thyroid hormone levels have been noted in rats, chronic toxicity studies in three different animal species demonstrated no apparent oestrogenic effects or treatment-related effects on any other component of the endocrine system (Cornell University 1999).

Bohmler and Borowski (2004) tested pendimethalin for oestrogenic activity using the E-screen which examines the proliferative effect of oestrogens on their target cells as the endpoint. This assay uses MCF-7 cells and compares the cell number achieved in the absence of oestrogens (negative control), in the presence of 17β-oestradiol (positive control) and in the presence of the substance under investigation. Pendimethalin was found to be a partial oestrogen receptor agonist. Further studies may be required to determine whether pendimethalin has endocrine-disrupting effects *in vivo* at environmental concentrations.

2.6.5jhu Mode of action of pendimethalin

Plants

Pendimethalin is a selective herbicide which is absorbed by the roots and leaves. It acts as a microtubule disruptor on pre-emergent plants. It inhibits cell division and cell elongation to prevent seedling development and is active on the roots and coleoptile of susceptible weeds. Affected plants die shortly after germination or following emergence from the soil.

Aquatic animals

No data on the mode of action of pendimethalin in aquatic fauna was found.

2.6.6 Mesocosm and field studies

Freshwater mesocosm and field studies

Only limited information on the effects of pendimethalin on freshwater communities could be found in the open literature.

Poleksić *et al.* (1995) treated sections of an experimental irrigation canal near Belgrade, Serbia, with 30 and 60 µg l^{-1} pendimethalin. Changes in phytoplankton production rate, abundance and diversity of zooplankton and bottom fauna were investigated. In addition, cages containing first year carp were placed on the bottom of the canal. Measurements of functional enzyme activity were taken and pathomorphological changes to the gills, liver and kidney were also studied. No specific details of the results are available. However, the authors report that the applied concentrations induced adverse effects on biotic components of the ecosystem studied.

A study published by the Danish Environmental Protection Agency (2004) found that pendimethalin affected algal communities at relatively low concentrations. Green algae were particularly sensitive, and effects on their biomass intensified even after transfer into clean water following exposure to the lowest test concentration (10 µg l^{-1}). In comparison, mortality of the crustacean, *Gammarus pulex*, first occurred at concentrations in the mg l^{-1} range, and then only after continuous exposure for several days.

Ebke *et al*. (2001) investigated the acute and chronic effects of a pendimethalin formulation product (STOMP 400: 400 g l^{-1}) in outdoor mesocosms in Germany, focusing on effects on biological parameters under field conditions in naturalised pond water. The study was conducted in compliance with GLP. The protocol and test design were based on a series of documents (including the draft OECD guideline and SETAC workshop outputs). Pendimethalin was applied in a single application of formulated product STOMP 400 (400 g l^{-1} SC) to a series of naturalised ecosystems in stainless steel enclosures containing freshwater assemblages of pelagic and benthic organisms. Twenty large enclosures, containing 1600 litres of water with a sediment layer, were used to assess the impact on pelagic and benthic species, phytoplankton and periphyton. A further six smaller enclosures were used to assess the impact of pendimethalin in macrophyte dominated systems, three of them were untreated and used as controls and three of them were treated with 5 µg l^{-1} pendimethalin (nominal) in a single application.

The pond was filled and allowed to equilibrate for 21 weeks before application. The nominal ranges of test concentrations for the biological effects test were 0.05, 0.23, 1.1, 4.9, 22.4, 50 and 150 µg l^{-1} pendimethalin with two enclosures per concentration. Six untreated enclosures were used as controls. One replicate of each test concentration of 50, 4.9 and 1.1 µg l^{-1} pendimethalin and a control were used to sample water and sediment for post-application analysis. The test substance was applied as a single application directly into the water column. The test duration was 128 days.

Water and sediment samples from selected enclosures were analysed for pendimethalin using GC-NPD analysis. All enclosures were monitored for physical and

chemical parameters of the water, phytoplankton and macrophytes in weekly or bi-weekly intervals as appropriate. Biological effects were monitored at weekly intervals for periphyton, zooplankton, macrozoobenthos and emerging insects.

The measured concentrations at two hours after application ranged from 70% to 114% (overall average 87 ± 14%) of the nominal dose, confirming target exposure concentrations were achieved. In the dissipation segment of the study, single enclosures were treated with 1.1, 4.9 and 50 μg l^{-1} pendimethalin. Pendimethalin decreased rapidly from the water column regardless of exposure rate. The pendimethalin half-lives in the water column were 1.5, 1.9 and 1.2 days respectively. Sediment analysis demonstrated that pendimethalin remained in the top 5 cm. No pendimethalin was detected in the 5 to 15 cm sediment layer. Following the initial adsorption, pendimethalin residues declined with time.

The results of the study showed that for all zooplankton species, except Synchaetidae, there were no apparent treatment related effects. Transient decreases in population were seen with the Synchaetidae at day 28 and populations recovered rapidly. With regard to emerging insects, no statistically significant effects were observed in Diptera Ephemeroptera, Heteroptera or Coleoptera abundance at any concentration. Similarly, no statistically significant effects were observed for any macrozoobenthos (Arthropoda, Annelida, Gastropoda, Ostracoda or Baetidae) abundances at any concentration.

For chlorophyll a, a significant reduction was observed following applications at 1.1 μg l^{-1} pendimethalin and above. The decreases appeared to be concentration-dependent, however they were transient and measured concentrations recovered rapidly. No effects on chlorophyll a were observed at concentrations of 0.23 μg l^{-1} pendimethalin and below.

No statistically significant effects were observed in Bacillariophyceae, Chrysophyceae, Euglenophyceae or Cyanophyceae abundance at any concentration. Transient decreases in populations were observed with both Chlorophyceae and Cryptophyceae. Statistically significant effects were observed in Chlorophyceae abundance at concentrations of 1.1 μg l^{-1} and above and in Cryptophyceae abundance at concentrations of 4.9 μg l^{-1} and above. However, these reductions were transient and populations rapidly recovered in all concentrations.

With regard to periphyton, no statistically significant treatment related effects were observed in Bacillariophyceae, Chrysophyceae, Chlorophyceae, Cyanophyceae and Euglenophyceae abundance at any concentration.

For macrophytes, strong positive growth and development of all macrophytes species *Potamogeton, Myriophyllum, Chara, Ceratophyllum* and *Lemna* was clearly demonstrated in both control ponds and those treated at 5 μg l^{-1} pendimethalin. No treatment-related or significant observations on macrophytes were observed during the study.

The EU DAR considered the Predicted No Effect Concentration (PNEC) from the study to be 1.1 μg l^{-1}. This exposure concentration resulted in transient reductions in chlorophyll a, Cryptophyceae and Chlorophyceae, but recoveries were observed within 5 days to a few weeks. In addition, this concentration did not result in impact on any other trophic levels. These findings support the results of laboratory single species tests indicating that algal species are the most sensitive to pendimethalin.

The Ebke *et al.* study also investigated the potential bioconcentration of pendimethalin under simulated field conditions and the possible effects on fish of water-borne and oral exposure, including biomagnification.

The test was conducted in compliance with GLP (except the analysis of water and sediment). Pendimethalin was dosed into the water column in a single application at nominally 5 µg l^{-1} to naturalised experimental ponds containing an omnivorous fish species, golden orfe (*Leuciscus idus idus*). Three ponds were treated (two of them using pendimethalin technical, and the other using ^{14}C-labelled pendimethalin) and the other three ponds were used as controls.

The ponds were filled and allowed to equilibrate for more than 6 months before application. Eighty fish were introduced in each pond approximately nine days before the treatment. Samples of fish, Daphniidae, Baetidae, phytoplankton and periphyton were collected at regular intervals from the ^{14}C-labelled pendimethalin pond, combusted and counted by LSC to determine radioactivity.

Fish were observed for behaviour, growth and mortality in all ponds. Zooplankton and macroinvertebrates abundances were monitored to assess the availability of food for fish during the study. Samples of water and sediment were taken to determine the change in radioactivity over time.

Measured pendimethalin concentrations in the water column on day 0 were 84% and 96% of nominal in the technical pendimethalin treated ponds and 124% of nominal in the ^{14}C-labelled pendimethalin treated pond. These results indicated that the target exposure concentration of 5 µg l^{-1} was achieved. At subsequent sample dates, the measured ^{14}C equivalent concentrations were 88% and 59% on days 1 and 3 respectively, declining to below 50% within 7 days, and continuing to decline to c. 15% at the end of the exposure phase. The characterisation showed that 19% of the total radioactivity was parent pendimethalin at 7 days and by day 14 the radioactivity consisted of polar metabolites.

Radioactivity was measured in the sediment (0 to 5 cm) on twelve occasions. The measured ^{14}C equivalent concentrations ranged from 11 µg/kg to 36 µg/kg dry weight except on two occasions when the concentrations were higher (49 and 72 µg/kg dry weight on days 84 and 112 respectively). In general, the concentration in sediment reached a plateau of between 20 to 30 µg/kg dry weight from day 14 onwards. The majority of the radioactivity in the sediment remained unextractable.

Growth of fish was observed in all the ponds and the feeding behaviour demonstrated that zooplankton, macrozobenthos, emerging insects and periphyton were used as food source. Mortality of the fish was observed, especially between days 29 and 39; the mortality was observed in all the ponds, was not treatment related and was attributable to an infestation of ecto-parasites (mean mortality in control ponds, 32.9%; 31.7% for treated ponds).

Uptake of radioactivity was observed in all sampled biota. For all groups, with the exception of Baetidae and periphyton, there was rapid depuration and loss of radioactivity with time.

Measured ^{14}C equivalent concentrations in fish rapidly declined to below 25% of the initial uptake in 7 days and to 10% by day 14. The majority of the radioactivity (70%) remained unextractable. Analysis of the extractable radioactivity characterised four unknown metabolites, all lower than 10% of radioactivity. Parent pendimethalin and two metabolites, 2,6-dinitro-3,4-dimethyl-aniline (the pendimethalin-alcohol) were not detected in any fish sample.

The conclusions of the study were:

- No biological effects on fish behaviour, growth or mortality were observed when a single application of 5 µg l^{-1} of pendimethalin was applied. Fish were exposed to pendimethalin via the water column and on pendimethalin exposed food sources.

The rapid environmental dissipation and degradation of pendimethalin within the test system, was reflected in the rapid depuration of ^{14}C pendimethalin equivalent concentrations in fish and all other sampled biota. The low percentage of applied radioactivity remaining in the test system was considered to be as a result of biotransformation of the available carbon-14.

- There were no significant transfers of radioactivity through primary producers to grazing zooplankton or macrozoobenthos or to omnivorous fish. Indeed, with the rapid losses from all biota in the test system, there was no evidence of biomagnification of pendimethalin or its metabolites within the aquatic food chain.

This study was considered by the EU DAR (2003) to be suitable to modify the BCF in fish and invertebrates to around 1000 with a clearance time of 5.1 days (>95% in 5 days) and to indicate a low potential for food chain biomagnification (see Section 3.5.2).

Saltwater mesocosm and field studies

No data from mesocosm or field studies using saltwater organisms were found.

3 Calculation of PNECs as a basis for the derivation of quality standards

3.1 Derivation of PNECs by the TGD deterministic approach (AF method)

3.1.1 PNECs for freshwaters

PNEC accounting for the annual average concentration

For the freshwater environment, data are available for the 'base set' of toxicity tests (i.e., tests with algae, crustaceans and fish) as well as macrophytes, which are target organisms based on the mode of toxicity, and therefore the EU TGD assessment factor (AF) method can be applied (ECB 2003). Long-term (lt) data were available for four taxonomic groups (algae, crustaceans, fish and macrophytes) for pendimethalin. Based on the information available all taxa appear to be of similar sensitivity to pendimethalin. Table 2.8 summarises the most sensitive long-term freshwater toxicity data that were found.

All of the most sensitive toxicity results are reported in unpublished studies carried out in support of pesticide registration. It is not possible to assess the reliability of these studies as only summary information on the tests is provided in compendium reports. Further information was sought and relevant extracts from the Draft Assessment Report (EU DAR 2003) prepared by the pendimethalin Rapporteur Member State (RMS) (Spain) under the Directive 91/414/EEC Review process were made available via the Pesticide Safety Directorate. For the critical data points it has been possible to establish that the studies were carried out under GLP, with analytical monitoring of the exposure concentrations and were considered valid in the EU DAR.

In the absence of any other suitable studies it is proposed that the PNEC is based on the following data:

- The lowest long-term result for algae is a 5-day growth inhibition (using the growth rate endpoint) NOEC of 3.0 µg l^{-1} for *Pseudokirchneriella subcapitata* (EU DAR 2003: Table 2.8). This is supported by a 5-day growth inhibition NOEC of 3.2 µg l^{-1} for *Navicula pellicosa* (EU DAR 2003; Table 2.8).
- The one available long-term study with macrophytes is a 14-day growth inhibition NOEC of 5.6 µg l^{-1} for *Lemna gibba* (EU DAR 2003; Table 2.8).
- The one available long-term study with invertebrates is a 21-day reproduction NOEC of 14.5 µg l^{-1} for *Daphnia magna* (EU DAR 2003; Table 2.8).
- The one available long-term study with fish is a 288-day reproduction NOEC of 6.3 µg l^{-1} for *Pimephales promelas* (EU DAR 2003; Table 2.8).

A long-term freshwater PNEC for pendimethalin should therefore be based on the NOEC for effects on the green alga *Pseudokirchneriella subcapitata* (3.0 μg l^{-1}) and an assessment factor of 10 because of the availability of long-term data for three trophic levels. This results in:

$PNEC_{freshwater\_lt}$ = 3.0 μg l^{-1}/AF (10) = 0.3 μg l^{-1} pendimethalin

Since pendimethalin has been shown to partition rapidly to sediments it is unlikely that aquatic habitats would be exposed to pendimethalin for sufficiently long to result in chronic toxicity unless field application is repeated, allowing a continuous release of herbicide into the water body, or there is a continuous discharge from a point source. The PNEC calculated above is, therefore, likely to be conservative under most natural conditions.

PNEC accounting for a maximum allowable concentration

Freshwater short-term toxicity data are available for four taxonomic groups (algae, crustaceans, fish and macrophytes). Table 2.9 summarises the most sensitive short-term freshwater toxicity data found for pendimethalin.

Short-term toxicity of pendimethalin has the greatest effects on algae and macrophytes, which is consistent with the mode of action of the substance. As with the long-term data, all the most sensitive results come from unpublished studies which were conducted in support of product registration. The lowest short-term result is a 5-day growth inhibition (using the growth rate endpoint) EC50 value of 5.8 μg l^{-1} for the diatom *Navicula pelliculosa,* which is supported by a 5-day growth inhibition EC50 of 6.7 μg l^{-1} for the green algae *Pseudokirchneriella subcapitata*. These studies are cited in the EU DAR (2003) and are considered valid as they were carried out under GLP, with analytical monitoring of the exposure concentrations (Table 2.9). A long-term study with macrophytes reported a 14-day growth inhibition EC50 of 12.5 μg l^{-1} for *Lemna gibba* (EU DAR 2003; Table 2.9). Short term toxicity values reported for invertebrates and fish are higher than those for algae and macrophytes being in the range 50 to 1000 μg l^{-1} (Table 2.9).

Based on guidance in the TGD on effects assessment for intermittent releases [Section 3.3.2 of Part II of the TGD document (ECB 2003)] and the fact that algae and macrophytes are clearly the most sensitive taxa, an assessment factor of 10 rather than 100 should be applied to the lowest data for *N. pelliculosa*. This results in:

$PNEC_{freshwater\_st}$ = 5.8 μg l^{-1}/AF (10) = 0.58 μg l^{-1} pendimethalin

3.1.2 PNECs for saltwaters

The effects database for saltwater species is considerably smaller than that for freshwater organisms. Short-term toxicity data are available for four different taxonomic groups (algae, crustaceans, fish and molluscs) but only one long-term datum (for algae) is available. The limited saltwater toxicity data do not differ markedly from the range of values obtained for corresponding freshwater species, and for algae and invertebrates are very similar (Tables 2.8 and 2.9).

Overall, the saltwater database is too small to draw firm conclusions on possible differences between freshwater and saltwater organisms, since no data are available for marine organisms such as echinoderms. However, for the most sensitive of those taxonomic groups that have been assessed, algae, there are no apparent differences

in the sensitivity of freshwater or saltwater species. Therefore, the TGD approach of using a combined freshwater and saltwater dataset for the saltwater effects assessment can be used and the proposed freshwater PNECs should be considered in deriving corresponding values for saltwater bodies.

PNEC accounting for the annual average concentration

The most sensitive result is a 5-day growth inhibition (using the growth rate endpoint) NOEC for the diatom, *Skeletonema costatum*, of 2.7 µg l^{-1} (EU DAR 2003: Table 2.10). This value is consistent with the lowest valid freshwater NOEC value of 3.0 µg l^{-1} for effects on the freshwater alga *Pseudokirchneriella.* The combined dataset indicates that algae are the most sensitive taxonomic group in both freshwater and saltwater with comparable species showing similar sensitivities. Therefore, it is recommended that the lowest marine toxicity value is used along with an assessment factor of 10 (but without the application of an additional assessment factor to account for the absence of data for groups such as echinoderms). This results in:

$PNEC_{saltwater\_lt}$ = 2.7 µg l^{-1}/AF (10) = 0.27 µg l^{-1} pendimethalin

PNEC accounting for a maximum allowable concentration

The lowest valid short-term saltwater toxicity value is a 5-day growth inhibition (using the growth rate endpoint) EC50 for *Skeletonema costatum* of 5.2 µg l^{-1}. This value is consistent with the lowest short-term freshwater 5-day growth inhibition EC50 of 5.8 µg l^{-1} for the alga *Navicula pelliculosa.*

Since the combined fresh- and salt-water datasets clearly show that algae and macrophytes are the most sensitive taxa, it is recommended that the 5-day growth inhibition EC50 for *Skeletonema costatum* of 5.2 µg l^{-1} is used to establish a PNEC to protect saltwater taxa. Although data are not available for marine taxa such as echninoderms it is proposed that, given the mode of action of pendimethalin, an assessment factor of 10 is applied without an additional factor to account for specifically saltwater species such as echinoderms. This results in:

$PNEC_{saltwater\_st}$ = 5.2 µg l^{-1}/AF (10) = 0.52 µg l^{-1} pendimethalin

3.2 Derivation of PNECs by the TGD probabilistic approach (SSD method)

The minimum number of long-term toxicity data (at least 10 NOECs from eight taxonomic groups) required by the TGD are not available. Therefore, the species sensitivity distribution (SSD) approach cannot be used for PNEC derivation.

3.3 Derivation of existing EQSs

The freshwater annual average (AA) for pendimethalin in Ayscough *et al.* (1997) was derived by applying a safety factor of 2 to the same data used in Section 3.1 above, resulting in an EQS of 1.5 µg l^{-1}. Since the standard was derived from chronic data for the group of organisms found to be most sensitive to pendimethalin only a small assessment factor was applied to allow for the possibility that other algal species may

be more sensitive. A maximum allowable concentration (MAC) of 6 µg l^{-1} was derived based on the EC50s for growth of 5.8 and. 6.7 µg l^{-1} reported for the algae, *Navicula pelliculosa*, and *Selenastrum capricornutum*, respectively. No additional assessment factor was considered necessary as the standard was based on data reported for inhibition of growth rather than algicidal effects and algae rapidly recover when exposed to short episodic events.

Based on the few data available on the toxicity of pendimethalin to saltwater organisms, the view was taken by Ayscough *et al.* (1997) that saltwater and freshwater species have similar sensitivities. Therefore, the EQSs proposed for the protection of freshwater organisms were considered adequate for the protection of saltwater life and the same values were proposed as tentative EQSs.

3.4 Derivation of PNECs for sediment

The TGD trigger value of a log Koc or log Kow of ≥3 is met, as the reported log Kow is 5.2 (EC 2003) and the log Koc is 3.83 – 4.47 (EC 2003). The reported Koc values in Table 2.5 range from 6700 to 29400. There are no reliable experimental data on sediment toxicity for pendimethalin and, therefore, no $PNEC_{sediment}$ can be derived.

3.5 Derivation of PNECs for secondary poisoning of predators

3.5.1 Mammalian and avian toxicity data

Three reviews have been published on pendimethalin toxicity (ESR 1998, EC 2003, WHO 2003). All three reviews were consulted as the primary sources and assumed to contain the most sound and comprehensive mammalian data. Additional literature searches were performed from 2003 to the present day to locate any lower effects data since 2003, but none were found.

For birds, due to the lack of relevant data in the above mentioned reviews, a comprehensive literature search was performed to locate any relevant data. No data could be located for chronic effects on birds.

Table 3.1 summarise the most sensitive mammalian and bird oral toxicity data relevant for the assessment of secondary poisoning.

Table 3.1 Most sensitive mammalian and bird oral toxicity data relevant for the assessment of secondary poisoning

Type of study, reference and result	Details
Sub-chronic toxicity studies to mammals	
American Cyanamid Co., 1986 Cited in WHO (2003) **Sub-chronic NOAEL = 100 mg kg^{-1}diet (approximately 5 mg kg^{-1} bw d^{-1})**	Charles River CD rats received pendimethalin via the diet for 13 weeks at doses of 0, 100, 500 or 5000 mg kg^{-1} diet (approximately 0, 5, 25 and 250 mg kg^{-1} bw d^{-1}). At the top dose, food intake and body weight gain were decreased; absolute and relative kidney weights were increased; and unspecified signs of hepatotoxicity occurred. Absolute and relative uterus and ovary weights decreased in females at 500 mg kg^{-1} diet. The NOAEL was based on no treatment-related effects being observed.
Cited in EC (2003) **NOAEL = 500 mg kg^{-1}diet (stated to be 41.3 mg kg^{-1} bw d^{-1})**	Rats (strain unspecified) received pendimethalin orally (route of administration unspecified) for 90 days at unspecified doses. The NOAEL was based on unspecified effects on the liver and thyroid. No further details were available.
Chronic toxicity studies to mammals	
Biodynamics Inc., 1974a Cited in WHO (2003) **Chronic LOAEL = 100 mg kg^{-1} diet (approximately 5 mg kg^{-1} bw d^{-1})**	Male and female CD-1 mice (75/sex/group) received pendimethalin via the diet for 18 months at doses of 0, 100, 500 or 2500 mg kg^{-1} diet (approximately 0, 5, 25 and 125 mg kg^{-1} bw d^{-1}). At the highest dose level, hyperglycaemia and increased thyroid and adrenal gland weights occurred. Hyperglycaemia was also observed in the lowest treatment group. The LOAEL was based on hyperglycaemia being observed at the lowest dose.
Biodynamics Inc., 1974b Cited in WHO (2003) **Chronic LOAEL = 100 mg kg^{-1} diet (approximately 5 mg kg^{-1} bw d^{-1})**	Male and female Long-Evans rats (60/sex/group) received pendimethalin via the diet for 2 years at doses of 0, 100, 500 or 2500 mg kg^{-1} diet (approximately 0, 5, 25 and 125 mg kg^{-1} bw d^{-1}). Hepatomegaly, increased alkaline phosphatase levels and increased thyroid and kidney weights were observed at the top dose. Unspecified hepatotoxicity was observed in the lowest treatment group. The LOAEL was based on hepatotoxicity being observed at the lowest dose.
Reproductive and developmental toxicity in mammals	
American Cyanamid Co., 1972; 1979 Cited in WHO (2003) **Foetal LOAEL = 250 mg kg^{-1} bw d^{-1}**	Rats (strain unspecified) received pendimethalin orally via gavage on days 6 to 15 of gestation at unspecified doses. Unspecified minor anomalies and reduced foetal weight were observed at 1000 mg kg^{-1} bw d^{-1} and the LOAEL was based on reduced ossification of the extremities being present at 250 and 500 mg kg^{-1} bw d^{-1}. No teratogenicity was observed.
Biodynamics Inc. 1974c Cited in WHO (2003) **Reproductive NOAEL = 1000 mg kg^{-1} bw d^{-1}**	In a three-generation reproductive study, Long-Evans rats received pendimethalin via the diet at unspecified doses and for an unspecified exposure period. The NOAEL was based on no reproductive toxicity being observed. No further details were available.

American Cyanamid Co. 1986 Cited in ESR reports (1998); WHO (2003) **Reproductive NOAEL = 30 mg kg^{-1} bw d^{-1}** **Developmental NOAEL = 60 mg kg^{-1} bw d^{-1}**	Female New Zealand White rabbits (20/dose) received pendimethalin orally via gavage on gestation days 6 to 18 at doses of 0, 15, 30 or 60 mg kg^{-1} bw d^{-1}. The NOAEL for reproductive toxicity was based on increased incidences and frequencies of anorexia, adipsia and reduction in body weight gain. The developmental NOAEL was based on no adverse effects occurring on uterine or foetal parameters.
Carcinogenicity in mammals	
Cited in EC (2003) **Chronic NOAEL = 12.5 mg kg^{-1} bw d^{-1}**	Dogs (strain unspecified) received pendimethalin orally (unspecified) for 2 years at unspecified doses. The NOAEL was based on unspecified effects in the liver and adenomas in the thyroid. No further details were available.
Toxicity to birds	
OPP, 2000 Cited in ECOTOX (2007) **Sub-chronic LC50 = >4640 mg kg^{-1} diet**	Five-day to ten-day old Mallard ducks (*Anas platyrhnchos*) received pendimethalin via the diet at unspecified doses for 8 days. The LC50 represents the dose at which 50% mortality occurs. No further details were provided.
OPP, 2000 Cited in ECOTOX (2007) **Sub-chronic LD50 = 1421 mg kg^{-1} bw**	Fourteen-day old Mallard ducks (*Anas platyrhnchos*) received pendimethalin via the diet at unspecified doses for 8 days. The LD50 represents the dose at which 50% mortality occurs. No further details were provided.
OPP, 2000 Cited in ECOTOX (2007) **Sub-chronic LC50 = 4187 mg kg^{-1} diet**	Ten-day to fourteen-day old Northern Bobwhites (*Colinus virginianus*) received pendimethalin via the diet at unspecified doses for 8 days. The LC50 represents the dose at which 50% mortality occurs. No further details were provided.

3.5.2 PNECs for secondary poisoning of predators

Fish bioconcentration data (as BCF values) for pendimethalin range from 1000 to 5100, hence the trigger of a BCF >100 is met and derivation of PNECs for secondary poisoning of predators is required. The highest (worst case) value for the bioaccumulation potential of pendimethalin was determined for the bluegill sunfish, *Lepomis macrochirus* in a study by Forbis *et al.* (1986 cited in US EPA RED 1997). This study was also evaluated by the Rapporteur Member State under the Directive 91/414/EEC Review process (EU DAR 2003). The test followed US EPA recommendations for the study of bioaccumulation in fish using radiolabelled materials and was carried out under GLP. The test was considered valid. Lower BCFs for fish and invertebrates have been reported in a mesocosm study (see Section 2.6.6) which is also considered by the EU DAR to be valid but to represent a more realistic estimate of the BCF, given the fate and behaviour of pendimethalin in the environment.

The lowest relevant mammalian NOAEL for pendimethalin is <5 mg kg^{-1} bw d^{-1}. This NOAEL is based on a sub-chronic study (13-week) with rats in which a NOAEL of 5 mg kg^{-1} bw d^{-1} was identified (American Cyanamid 1986) and on two chronic studies with

rats in which a LOAEL of 5 mg kg^{-1} bw d^{-1} was identified (Biodynamics Inc 1974a and b). However, it has not been possible to review the raw data from these studies.

Since the LOAELs for the chronic studies were identical to the NOAEL of a sub-chronic study it is recommended that the NOAEL for pendimethalin is lower than the chronic LOAELs, i.e. < 5 mg kg^{-1} bw d^{-1}. It is proposed that a NOAEL value of 2.5 mg kg^{-1} bw d^{-1} is used which is derived by dividing the chronic LOAELs by a factor of 2.

The appropriate assessment factor to derive a PNEC based on a sub-chronic $NOEC_{food}$ of a mammalian study is 90 (Table 23 of TGD).

$PNEC_{secpois.biota}$ = $NOEC_{food}$ (50 mg kg^{-1}) / AF 90 = 0.55 mg kg^{-1} prey (wet wt)

Bioconcentration of pendimethalin in aquatic organisms is high with Bioconcentration Factors in fish ranging from 1000 (in a mesocosm study) to 5100 (in a laboratory study). The mesocosm study, which used a single dose and was carried out to GLP, was considered by the EU DAR (2003) to be valid and suitable to modify the BCF in fish and invertebrates to around 1000 with a clearance time of 5.1 days (>95% in 5 days) and to indicate a low potential for food chain biomagnification. However, there is a possibility that pendimethalin could be released on a more continuous basis and, as a precautionary measure, it would be more appropriate to use the BCF of 5100 in the calculation of the $PNEC_{secpois.water}$.

The corresponding safe concentration in water (preventing bioaccumulation in prey to levels > $PNEC_{secpois.biota}$) can therefore be calculated as follows:

$PNEC_{secpois.water}$ = $PNEC_{secpois.biota}$ (0.55 mg kg^{-1}) / BCF

If the BCF of 5100 reported in the laboratory study (which represents a precautionary value) is used for the calculation, this would result in a corresponding water concentration of:

$PNEC_{secpois.water}$ = 0.55 mg kg^{-1} prey / BCF (5100) = 0.1 µg pendimethalin l^{-1}

This concentration is lower than the proposed long-term PNEC for the protection of freshwater and saltwater organisms (i.e. 0.27 µg l^{-1}). Therefore, if quality standards are set on the basis of the proposed long-term water column it is probable that predators would not be protected from secondary poisoning.

4 Analysis and monitoring

Limited information is available on analytical methods. WHO (1996) report the limit of detection (LoD) as 0.01 µg l^{-1} by gas chromatography-mass spectrometry (GC/MS).

More recently, Bruzzoniti *et al.* (2006) describe a solid phase extraction (SPE) method that has been optimized for the GC/MS simultaneous determination of herbicides, including pendimethalin. This optimized method was successfully checked for the identification and quantification of selected herbicides in raw and drinking water samples, with quantification limits of 0.01 µg l^{-1}.

The Environment Agency (2000) gives details for the determination of organochlorine pesticides in waters and complex matrices. A footnote in this report suggests that the method described therein (SPE with ethyl acetate) can be used for analysis of pendimethalin with 'similar performance' as for those chemicals described in detail. The LoDs ranged from 0.001 to 0.009 µg l^{-1}.

For water, the lowest proposed PNEC derived for pendimethalin is 0.1 µg l^{-1}. The data quality requirements are that, at a third of the EQS, total error of measurement should not exceed 50 per cent. Using this criterion, it is evident that current analytical methodologies should offer adequate performance to analyse for pendimethalin.

5 Conclusions

5.1 Availability of data

Very limited long- and short-term laboratory data are available for four different freshwater taxonomic groups including algae, crustaceans, fish and macrophytes. Based on the limited information available these taxa would appear to be similarly sensitive to chronic exposure to pendimethalin at very low concentrations. However algae and macrophytes appear to be the most sensitive taxa in acute exposures. This is consistent with the use of the substance as an herbicide. For saltwater organisms, single species short-term toxicity data are available for four different taxonomic groups (algae, crustaceans, fish and molluscs). However, long-term toxicity data are not available for the minimum of three saltwater taxa (algae, crustaceans and fish) required under Annex V of the Water Framework Directive. The results from freshwater mesocosm studies confirm effects on algal species at very low concentrations.

Pendimethalin has been shown to cause fluctuations in thyroid hormones in humans and in rats. However, chronic toxicity studies in three different animal species did not induce any oestrogenic or treatment-related effects on any other component of the endocrine system. *In vitro* assays using MCF-7 cells suggest that pendimethalin is a partial oestrogen receptor agonist.

5.2 Derivation of PNECs

The proposed PNECS are described below and summarised in Table 5.1.

5.2.1 Long-term PNEC for freshwaters

The lowest valid long-term toxicity value is for freshwater algae where a 5-day NOEC of 3.0 $\mu g\ l^{-1}$ for effects on growth inhibition (using the growth rate endpoint) of the green alga *Pseudokirchneriella subcapitata* has been reported in the EU DAR (2003). Reliable long-term NOECs are available for algae, invertebrates and fish, and therefore an assessment factor of 10 can be applied resulting in a $PNEC_{freshwater\_lt}$ of 0.3 $\mu g\ l^{-1}$.

5.2.2 Short-term PNEC for freshwaters

Reliable short-term data are available for algal, invertebrate and fish species. The lowest valid short-term toxicity values is a 5-day growth inhibition (using the growth rate endpoint) EC50 values of 5.8 $\mu g\ l^{-1}$ for the diatom *Navicula pelliculosa* reported in the EU DAR (2003). The short-term toxicity database for freshwater organisms is not extensive, but does adequately indicate that algae and macrophytes are the most sensitive taxa to short-term exposure to pendimethalin. An assessment factor of 10 was therefore applied, resulting in a $PNEC_{freshwater\_st}$ of 0.58 $\mu g\ l^{-1}$.

5.2.3 Long-term PNEC for saltwaters

Since long-term single species toxicity data are only available for algae, a combined freshwater and saltwater dataset for the saltwater effects assessment was used to derive the PNEC. The most sensitive result is a 5-day growth inhibition (using the growth rate endpoint) NOEC for the diatom, *Skeletonema costatum*, of 2.7 µg l^{-1} reported in the EU DAR (2003). The combined dataset indicate that algae are evidently the most sensitive taxonomic group in both freshwater and saltwater with comparable species showing similar sensitivities. Therefore, it is recommended that the lowest marine toxicity value is used along with an assessment factor of 10 (but without the application of an additional assessment factor to account for the absence of data for groups such as echinoderms). This results in a $PNEC_{saltwater\_lt}$ of 0.27 µg l^{-1} which is essentially the same as the freshwater PNEC.

5.2.4 Short-term PNEC for saltwaters

Reliable short-term saltwater toxicity data are available for algae, invertebrates and fish. The most sensitive result is a 5-day growth inhibition (using the growth rate endpoint) EC50 for the diatom, *Skeletonema costatum*, of 5.2 µg l^{-1} reported in the EU DAR (2003), which is consistent with toxicity values for freshwater species. The combined dataset indicate that algae are evidently the most sensitive taxonomic group in both freshwater and saltwater with comparable species showing similar sensitivities. Therefore, it is recommended that the lowest toxicity value for marine algae is adopted along with an assessment factor of 10 (but without the application of an additional assessment factor to account for the absence of data for groups such as echinoderms). This results in a $PNEC_{saltwater\_lt}$ of 0.52 µg l^{-1} which is consistent with the freshwater PNEC.

5.2.5 PNEC for sediments

The TGD trigger value of a log Koc or log Kow of ≥3 is met, as the reported log Kow is 5.2 and reported Koc values range from 6700 to 29400. There are no reliable experimental data on sediment toxicity for pendimethalin and therefore no $PNEC_{sediment}$ can be derived.

5.2.6 PNEC for secondary poisoning

Bioconcentration data (as BCF values) for pendimethalin for fish range from an 'environmentally realistic' value of 1000 (in a mesocosm study) to a 'worst case' value of 5100 (in a laboratory study). Hence the trigger of a BCF >100 is exceeded and derivation of PNECs for secondary poisoning of predators is required. The lowest relevant $NOEC_{food}$ is 50 mg kg^{-1} derived by extrapolation from a LOEC of 100 mg kg^{-1} from a 13-week study with rats.

Bioconcentration of pendimethalin in aquatic organisms is high with Bioconcentration Factors in fish ranging from 1000 (in a mesocosm study) to 5100 (in a laboratory study). The mesocosm study, which used a single dose and was carried out to GLP, was considered by the EU DAR (2003) to be valid and suitable to modify the BCF in fish and invertebrates to around 1000 with a clearance time of 5.1 days (>95% in 5 days) and to indicate a low potential for food chain biomagnification. However, there is a possibility that pendimethalin could be released on a more continuous basis and, as a precautionary measure, it would be more appropriate to use the BCF of 5100 in the calculation of the $PNEC_{secpois.water}$.

Using the reported BCF of 5100 from the laboratory study in the US EPA RED (1997) for the calculation results in a corresponding water concentration of $PNEC_{secpois.water}$ = 0.55 mg kg^{-1} prey / BCF (5100) = 0.1 µg pendimethalin l^{-1}.

This concentration is lower than the proposed long-term PNEC for the protection of freshwater and saltwater organisms (i.e. 0.27 µg l^{-1}). Therefore, if quality standards are set on the basis of the proposed long-term water column it is probable that predators would not be protected from secondary poisoning.

Table 5.1 Summary of proposed PNECs

Receiving medium/exposure scenario	Proposed PNEC (µg l^{-1})	Existing EQS (µg l^{-1})
Freshwater/long-term	0.3	1.5
Freshwater/short-term	0.58	6.0
Saltwater/long-term	0.27	1.5
Saltwater/short-term	0.52	6.0
Sediments	Insufficient data	-
Secondary poisoning	0.1	-

5.3 Analysis

For water, the lowest proposed PNEC derived for pendimethalin is 0.1 µg l^{-1}. The data quality requirements are that, at a third of the EQS, total error of measurement should not exceed 50%. Using this criterion, it is evident that current analytical methodologies using solid phase extraction followed by gas chromatography-mass spectrometry, should offer adequate performance to analyse for pendimethalin.

5.4 Implementation issues

Based on consideration of the information collated within the report and the proposed PNECs the following comments are made re: implementation:-

- Current analytical methods should be adequate for compliance assessment.
- The PNECs derived are not subject to excessive uncertainty with assessment factors of 10 being applied for their derivation.
- Pendimethalin adsorbs rapidly to sediment and therefore its potential impact on sediment dwelling organisms will need to be considered as more data becomes available.
-

References & Bibliography

American Cyanamid Co. (1972). Report of December 12. Wayne, NJ, USA. Cited in ESR (1998) and WHO (1996, 2003).

American Cyanamid Co. (1979). Teratology study in rats. Wayne, NJ, USA. Cited in ESR (1998) and WHO (1996, 2003).

American Cyanamid Co. (1986). Toxicology report AX86-1 experiment L-2190. AC 92, 553: A 13 week rat feeding study. Wayne, NJ, USA. Cited in ESR (1998) and WHO (1996, 2003).

Ayscough NJ, Hedgecott S, Mascarenhas R and Sutton AJ (1997) *Proposed environmental quality standards for pendimethalin in water.* R&D Technical Report No DoE 4244/1 Swindon:WRc plc.

Barker (2000) Toxicity of pendimethalin (AC 92553) to the alga *Selenastrum capricornutum* following a post-exposure recovery period. Report ETX-99-211. Cited in EU DAR (2003).

Biodynamics Inc. (1974a). Report on project no. 72R-747. Wayne, NJ, USA. Cited in ESR (1998) and WHO (1996, 2003).

Biodynamics Inc. (1974b). Report on project no. 72R-746. Wayne, NJ, USA. Cited in ESR (1998) and WHO (1996, 2003).

Biodynamics Inc. (1974c). 3 Generation reproduction study in AC 92, 553 in rats. Report on project no. 72R-748. Wayne, NJ, USA. Cited in ESR (1998) and WHO (1996, 2003).

Bohmler G and Borowski U (2004) Detection of estrogenic activity by means of a biological test system- part 2: Investigation of food relevant substances. *Deutsche Lebensmittel-Rundschau*, **100**, 133-139.

Bruzzoniti MC, Sarzanini C, Costantino G, Fungi M (2006) Determination of herbicides by solid phase extraction gas chromatography-mass spectrometry in drinking waters. *Analytica Chimica Acta*, **578**, 241-249.

Cedergreen N and Streibig JC (2005) The toxicity to non-target aquatic plants and algae: assessment of predictive factors and hazard. *Pest Management Science*, **61**, 1152-1160.

Chemfinder (2005) *ChemFinder database.* Cambridge, MA: CambridgeSoft Corp. Available from: http://chemfinder.cambridgesoft.com/ [Accessed 9 May 2007]

Cornell University (1999) *Pendimethalin (Prowl) Pesticide Petition Filing 8/99*. Available from: http://pmep.cce.cornell.edu/profiles/herb-growthreg/naa-rimsulfuron/pendimethalin/pendimethalin_pet_899.html [accessed 14 May 2007]

Danish Environmental Protection Agency (2004) Effects of Pesticides on Flora and Fauna in Danish Streams. Report No 82 2004. In Danish with English summary.

Available from: http://www.2.mst.dk/Udgiv/publikationer/2004/87-7614-110-1/html/sum.htm [accessed 14 May 2007]

Ebke, P., Rosenwald, J., Satter, P., Greener, N., Mitchell, G.C. and Horton, W. (2001a) Evaluation of direct and indirect effects of a 400 g/L SC formulation (Stomp 400) of pendimethalin (AC 92553) on aquatic organisms in outdoor pond mesocosms. Unpublished Report No ECO 00-101. 24 March 2001. Cited in EU DAR (2003).

ECOTOX (2007). US Environmental Protection Agency. ECOTOX Database.

EG&G, Bionomics (1975) Chronic toxicity of CL 92553 to the fathead minnow (*Pimephales promelas*). EG&G, Bionomics. Cited in EU DAR (2003).

Environment Agency (2000). *The determination of organochlorine pesticides and polychlorinated biphenyls in waters and complex matrices (2000): Methods for the Examination of Waters and Associated Materials*. Standing Committee of Analysts, Environment Agency, Lancaster, England.

ESR Report (1998). Pendimethalin. ECBI/54/98 – Add.1. Available from http:// www.ecb.jrc.it {Accessed May 07]

European Chemicals Bureau (ECB) (2003) *Technical Guidance Document in Support of Commission Directive 93/67/EEC on risk assessment for new and notified substances: Commission Directive (EC) No. 1488/94 on Risk Assessment for Existing Substances and Directive 98/8/EC of the European Parliament and of the Council Concerning the Placing of Biocidal Products on the Market*. Parts I–IV. Luxembourg: Office for Official Publications of the European Communities. Available from: http://ecb.jrc.it/tgdoc [Accessed 9 May 2007].

European Chemicals Bureau (ECB) (2005) European Substances Information System (ESIS). Available from: http://ecb.jrc.it/esis/ [Accessed 9 May 2007]

European Commission (2011). Common Implementation Strategy for the Water Framework Directive (2000/60/EC); Guidance document No. 27, Technical Guidance for Deriving Environmental Quality Standards; Technical Report 2001-055. Available at http://circa.europa.eu/Public/irc/env/wfd/library?l=/framework_directive/guidance_documents/tgd-eqs_cis-wfd/_EN_1.0_&a=d

EU DAR (2003) *Draft Assessment Report for the active substance pendimethalin. Finalised in the Standing Committee on Plant Health at its meeting on 13 November 2002 in view of the inclusion of pendimethalin in Annex I of Directive 91/414/EEC*. 7477/VI/98-Final. Brussels: European Commission Directorate-General Health & Consumer Protection. Available from: http://europa.eu.int/comm/food/plant/protection/evaluation/exist_subs_rep_en.htm [Accessed 29 March 2007].

Forbis, A, Georgie, L and Burgess, D (1985) Acute toxicity of AC 92553 to Daphnia magna: Static Acute Toxicity. Report #33409. Analytical Bio-Chemistry Laboratories Inc. Cited in EU DAR (2003).

Graney, RL (1981) Chronic (21-day) toxicity of AC92553 to Daphnia magna. Report 5179. Biospherics. Cited in EU DAR (2003).

Hazardous Substances Data Bank (HSDB) (2006) *Toxicology Data Network (TOXNET®): Hazardous Substances Data Bank (HSDB®)* [online]. Bethesda, MD:

Division of Specialized Institute for Environment and Health (IEH) (2005) *Chemicals purported to be endocrine disruptors. A compilation of published lists.* (Web Report W20). Leicester: IEH. Available from: http://www.silsoe.cranfield.ac.uk/ieh/publications/endocrine.html [Accessed 9 May 2007].

Helweg C, Mogensen BB, Sørensen PB, Madsen T, Bossi R, Rasmussen D and Petersen S (2003) *Fate of pesticides in surface waters, laboratory and field experiments*. Danish Environmental Protection Agency. Denmark. Pesticides Research No 68.

Hughes, JS, Alexander, MM and Wisk JD (1991) Effect of AC 92553 on growth of duckweed *Lemna gibba.* Report B400-31-4. Malcom Pirnie. Cited in EU DAR (2003).

Hughes, JS, Alexander, MM and Wisk JD (1992a) Effect of AC 92553 on growth of the freshwater diatom *N.pelliculosa*. Report B400-32-3. Malcom Pirnie. Cited in EU DAR (2003).

Hughes, JS, Alexander, MM and Wisk JD (1992b) Effect of AC 92553 on growth of the green alga, *S.capricornutum*. Report B400-32-1. Malcom Pirnie. Cited in EU DAR (2003).

Hughes, JS, Alexander, MM and Wisk JD (1992c) Effect of AC 92553 on growth of the marine diatom *S.costatum.* Report B400-32-4. Malcom Pirnie. Cited in EU DAR (2003).

Isensee AR and Dubey PS (1983) Distribution of pendimethalin in an aquatic microecosystem. *Bulletin of Environmental Contamination and Toxicology*, **30**, 239-244.

Mäenpää KA, Sormunrn AJ, Kukkonen JVK (2003) Bioaccumulation and toxicity of sediment associated herbicides (ioxynil, pendimethalin, and bentazone) in *Lumbriculus variegates* (Oligochaeta) and *Chironomus riparius* (Insecta). *Ecotoxicology and Environmental Safety*, **56**, 398-410.

Thompson, CM, Griffen, J and McAllister, WA (1980) Acute toxicity of AC 92553 to the freshwater crayfish (*Procambarus simulans*). Static Acute Bioassay. Report #25725. Analytical Bio-Chemistry Laboratories Inc. Cited in EU DAR (2003).

Meylan W M and Howard P H (1993) Computer estimation of the atmospheric gas-phase reaction rate of organic compounds with hydroxyl radicals and ozone. *Chemosphere*, **26**, 2293-2299.
OPP Pesticide Ecotoxicity Database (OPP) (2007) available at http://www.ipmcenters.org/Ecotox/index.cfm [accessed 10 May 2007].

OPP (2000). Pesticide Ecotoxicity Database (Formerly: Environmental Effects Database (EEDB)) Environmental Fate and Effects Division, U.S. EPA, Washington, D.C. Office of Pesticide Programs (OPP).

OPP Pesticide Ecotoxicity Database (OPP) (2007) available at http://www.ipmcenters.org/Ecotox/index.cfm [Accessed 9 May 2007].

Poleksić V, Karan V, Vidmanić L, Elezović I, Tutundžić V (1995) Field study of the indirect effects of a herbicide pendimethalin on freshwater communities. *Toxicology Letters, suppl* **1/78**, 67. (Abstract only).

Rasmussen D, Kukkonen JVK, Källqvist T, Helweg C, Madsen T, Mäenpää K, Sormunrn A and Efraimsen H (2002) *Distribution, degradation and toxicity of pesticides at environmentally realistic temperatures*. Nordtest Report TR496. Finland.

Sleight, B (1973) Acute toxicity of AC 92553 to bluegill sunfish (*Lepomis macrochirus*), rainbow trout (*Salmo gairdneri*) and channel catfish (*Ictaluras punctatus*). Report 5G1567. Bionomics Inc. Cited in EU DAR (2003)

Syracuse Research Corporation (SCR) (2005) PhysProp physical properties database [online]. Syracuse, NY: SRC. Available from: http://www.syrres.com/esc/physdemo.htm [Accessed 9 May 2007].

US Environmental Protection Agency (USEPA) (1997) Reregistration Eligibility Decision (RED) pendimethalin. EPA 738-R-97-007. Washington, DC: Office of Prevention, Pesticides and Toxic Substances, US EPA. Available from: http://www.epa.gov/oppsrrd1/REDs/0187red.pdf [Accessed 10 May 2006].

World Health Organization (WHO) (2003) Pendimethalin in Drinking Water, Background document for the development of WHO Guidelines for Drinking Water Quality. Available from: http://www.who.int/water_sanitation_health/dwq/chemicals/pendimethalin.pdf. [Accessed May 07]

(WHO) (1996) *Guidelines for drinking-water quality,* 2nd Edition Vol.2. Health Criteria and other supporting information. Geneva.

List of abbreviations

AA	annual average
AF	assessment factor
a.i.	active ingredient
BCF	bioconcentration factor
bw	body weight
CAS	Chemical Abstracts Service
CT	Elimination half-life
d	day
DO	dissolved oxygen
EC50	Concentration effective against 50 per cent of the organisms or animals tested
EQS	Environmental Quality Standard
FIFRA	Federal Insecticide, Fungicide and Rodenticide Act [US]
GC	gas chromatography
GLP	Good Laboratory Practice (OECD)
h	hour
LC50	Concentration lethal to 50 per cent of the organisms or animals tested
LOAEL	lowest observed adverse effect level
LOEC	lowest observed effect concentration
lt	long-term
MAC	maximum allowable concentration
NOAEL	no observed adverse effect level
NOEC	no observed effect concentration
OECD	Organization for Economic Co-operation and Development
PNEC	predicted no-effect concentration
RED	Reregistration Eligibility Document
SPE	Solid Phase Extraction
SSD	species sensitivity distribution
st	short-term
TGD	Technical Guidance Document
UKTAG	UK Technical Advisory Group
US EPA	US Environmental Protection Agency
WFD	Water Framework Directive

ANNEX I Data quality assessment sheets

Identified and ordered by alphabetical order of references.

Data relevant for PNEC derivation were quality assessed in accordance with the so-called Klimisch Criteria (Table A1).

Table A1 Klimisch Criteria*

Code	Category	Description
1	Reliable without restrictions	Refers to studies/data carried out or generated according to internationally accepted testing-guidelines (preferably GLP**) or in which the test parameters documented are based on a specific (national) testing guideline (preferably GLP), or in which all parameters described are closely related/comparable to a guideline method.
2	Reliable with restrictions	Studies or data (mostly not performed according to GLP) in which the test parameters documented do not comply totally with the specific testing guideline, but are sufficient to accept the data or in which investigations are described that cannot be subsumed under a testing guideline, but which are nevertheless well-documented and scientifically acceptable.
3	Not reliable	Studies/data in which there are interferences between the measuring system and the test substance, or in which organisms/test systems were used that are not relevant in relation to exposure, or which were carried out or generated according to a method which is not acceptable, the documentation of which is not sufficient for an assessment and which is not convincing for an expert assessment.
4	Not assignable	Studies or data which do not give sufficient experimental details and which are only listed in short abstracts or secondary literature.

* Klimisch H.-J, Andreae M and Tillmann U, 1997 A systematic approach for evaluating the quality of experimental toxicological and ecotoxicological data. *Regulatory Toxicology and Pharmacology*, **25**, 1–5.
** OECD Principles of Good Laboratory Practice (GLP). See: http://www.oecd.org/department/0,2688,en_2649_34381_1_1_1_1_1,00.html

Reference	Barker *et al.* (2000)

Information on the test species	
Test species used	*Pseudokirchneriella subcapitata (Selenastrum capricornutum)*
Source of the test organisms	-
Holding conditions prior to test	-
Life stage of the test species used	-

Information on the test design	
Methodology used	-
Form of the test substance	Not stated
Source of the test substance	-
Type and source of the exposure medium	-
Test concentrations used	1.6, 3.2, 6.3, 12.5, 25 and 50 µg l^{-1} + solvent control (acetone 100 µl l^{-1}) + control
Number of replicates per concentration	3 (6 for solvent control group)
Number of organisms per replicate	-
Nature of test system (Static, Semi-static or, Flow- through, duration, feeding)	Static
Measurement of exposure concentrations	Yes at beginning and end of 72 h exposure period. Measured concentrations between 72 and 99% of nominal at start and between 48 and 62.5% at end
Measurement of water quality parameters	Not stated
Test validity criteria satisfied	Yes
Water quality criteria satisfied	Not stated
Study conducted to GLP	Yes
Comments	Results of post-exposure recovery showed that after 5 days there was a difference in growth rate and biomass between highest concentration and remaining treatments and control. By day 7 average specific growth rate were the same for all treatments and control.

Reliability of study	**Reliable**
Relevance of study	**Relevant**
Klimisch Code	**1**

Reference	Cedergreen and Streibig 2005

Information on the test species	
Test species used	*(1) Lemna minor* *(2) Pseudokirchneriella subcapitata*
Source of the test organisms	(1) field collected from local pond, Copenhagen, Denmark (2) culture originated from Insitut for Vanforskning (NIVA) Norway
Holding conditions prior to test	(1) Sterilized and held in 'K'-medium, pH5 at 24°C. (clone tested for sensitivity towards standard ref. compounds) (2) stock culture held in dark at 5°C; transferred to 22°C and continuous light.
Life stage of the test species used	Not stated

Information on the test design	
Methodology used	(1) Non-standard but well described (2) Cited references comparable to ISO 1989
Form of the test substance	Technical grade 98% purity pendimethalin and Stomp (400 g l^{-1} a.i.)
Source of the test substance	BASF
Type and source of the exposure medium	Culture medium
Test concentrations used	8 dilutions (factor of 2)
Number of replicates per concentration	(1) 3 + 6 controls (2) 2 + 6 controls
Number of organisms per replicate	(1) 1 (2) not stated
Nature of test system (Static, Semi-static or, Flow- through, duration, feeding)	static
Measurement of exposure concentrations	No
Measurement of water quality parameters	Temp and pH monitored
Test validity criteria satisfied	Yes
Water quality criteria satisfied	Yes
Study conducted to GLP	Not stated
Comments	<table><tr><td></td><td colspan="2">Technical µg l^{-1}</td><td colspan="2">Formulated µg l^{-1}</td></tr><tr><td></td><td>EC10</td><td>EC50</td><td>EC10</td><td>EC50</td></tr><tr><td>*L. minor*</td><td>90</td><td>634</td><td>41</td><td>280</td></tr><tr><td>Alga</td><td>-</td><td>-</td><td>21</td><td>368</td></tr><tr><td></td><td colspan="2">7-day</td><td colspan="2">48-hour</td></tr></table>

Reliability of study	**Reliable with restrictions**
Relevance of study	**Relevant**
Klimisch Code	**2**

Reference	EG & G Bionomics (1975) cited in EU DAR (2003)

Information on the test species	
Test species used	*Pimephales promelas*
Source of the test organisms	Not stated in EU DAR, 2003
Holding conditions prior to test	Not stated in EU DAR, 2003
Life stage of the test species used	Not stated in EU DAR, 2003

Information on the test design	
Methodology used	US EPA series 75-2 Life cycle study
Form of the test substance	98.3% pure
Source of the test substance	Not stated in EU DAR, 2003
Type and source of the exposure medium	Not stated in EU DAR, 2003
Test concentrations used	Not stated in EU DAR, 2003
Number of replicates per concentration	Not stated in EU DAR, 2003
Number of organisms per replicate	Not stated in EU DAR, 2003
Nature of test system (Static, semi-static or flow- through, duration, feeding)	Flow-through, 288 days, feeding
Measurement of exposure concentrations	Yes
Measurement of water quality parameters	Yes
Test validity criteria satisfied	Yes
Water quality criteria satisfied	Not stated in EU DAR, 2003
Study conducted to GLP	Yes
Overall comment on quality	The study is of good quality having been conducted to a standardised methodology under GLP.

Reliability of study	**Reliable**
Relevance of study	**Relevant**
Klimisch Code	**1**

Reference	Graney (1981) cited in EU DAR (2003)

Information on the test species	
Test species used	*Daphnia magna*
Source of the test organisms	Not stated in EU DAR, 2003
Holding conditions prior to test	Not stated in EU DAR, 2003
Life stage of the test species used	Not stated in EU DAR, 2003

Information on the test design	
Methodology used	US EPA series 72-4b
Form of the test substance	92.2% pure
Source of the test substance	Not stated in EU DAR, 2003
Type and source of the exposure medium	Not stated in EU DAR, 2003
Test concentrations used	Not stated in EU DAR, 2003
Number of replicates per concentration	Not stated in EU DAR, 2003
Number of organisms per replicate	Not stated in EU DAR, 2003
Nature of test system (Static, semi-static or flow- through, duration, feeding)	Semi-static, 21 days, feeding
Measurement of exposure concentrations	Yes
Measurement of water quality parameters	Yes
Test validity criteria satisfied	Yes
Water quality criteria satisfied	Not stated in EU DAR, 2003
Study conducted to GLP	Yes
Overall comment on quality	The study is of good quality having been conducted to a standardised methodology under GLP.

Reliability of study	**Reliable**
Relevance of study	**Relevant**
Klimisch Code	**1**

Reference	Hughes *et al.* (1992a) cited in EU DAR (2003)

Information on the test species	
Test species used	*Navicula pelliculosa*
Source of the test organisms	Not stated in EU DAR, 2003
Holding conditions prior to test	Not stated in EU DAR, 2003
Life stage of the test species used	Growth phase

Information on the test design	
Methodology used	OECD guideline 201 and US EPA series 123-2
Form of the test substance	93% pure
Source of the test substance	Not stated in EU DAR, 2003
Type and source of the exposure medium	Not stated in EU DAR, 2003
Test concentrations used	Not stated in EU DAR, 2003
Number of replicates per concentration	Not stated in EU DAR, 2003
Number of organisms per replicate	Not stated in EU DAR, 2003
Nature of test system (Static, semi-static or flow- through, duration, feeding)	Static, 5 days, not applicable
Measurement of exposure concentrations	Yes
Measurement of water quality parameters	Yes
Test validity criteria satisfied	Yes
Water quality criteria satisfied	Not stated in EU DAR, 2003
Study conducted to GLP	Yes
Overall comment on quality	The study is of good quality having been conducted to a standardised methodology under GLP.

Reliability of study	**Reliable**
Relevance of study	**Relevant**
Klimisch Code	**1**

Reference	Hughes *et al.* (1992b) cited in EU DAR (2003)

Information on the test species	
Test species used	*Pseudokirchneriella subcapitata* (formerly) *Selenastrum capricornutum*
Source of the test organisms	Not stated in EU DAR, 2003
Holding conditions prior to test	Not stated in EU DAR, 2003
Life stage of the test species used	Growth phase

Information on the test design	
Methodology used	OECD guideline 201 and US EPA series 123-2
Form of the test substance	93% pure
Source of the test substance	Not stated in EU DAR, 2003
Type and source of the exposure medium	Not stated in EU DAR, 2003
Test concentrations used	Not stated in EU DAR, 2003
Number of replicates per concentration	Not stated in EU DAR, 2003
Number of organisms per replicate	Not stated in EU DAR, 2003
Nature of test system (Static, semi-static or flow- through, duration, feeding)	Static, 5 days, not applicable
Measurement of exposure concentrations	Yes
Measurement of water quality parameters	Yes
Test validity criteria satisfied	Yes
Water quality criteria satisfied	Not stated in EU DAR, 2003
Study conducted to GLP	Yes
Overall comment on quality	The study is of good quality having been conducted to a standardised methodology under GLP.

Reliability of study	**Reliable**
Relevance of study	**Relevant**
Klimisch Code	**1**

Reference	Hughes *et al.* (1992c) cited in EU DAR (2003)

Information on the test species	
Test species used	*Skeletonema costatum*
Source of the test organisms	Not stated in EU DAR, 2003
Holding conditions prior to test	Not stated in EU DAR, 2003
Life stage of the test species used	Growth phase

Information on the test design	
Methodology used	OECD guideline 201 and US EPA series 123-2
Form of the test substance	93% pure
Source of the test substance	Not stated in EU DAR, 2003
Type and source of the exposure medium	Not stated in EU DAR, 2003
Test concentrations used	Not stated in EU DAR, 2003
Number of replicates per concentration	Not stated in EU DAR, 2003
Number of organisms per replicate	Not stated in EU DAR, 2003
Nature of test system (Static, semi-static or flow- through, duration, feeding)	Static, 5 days, not applicable
Measurement of exposure concentrations	Yes
Measurement of water quality parameters	Yes
Test validity criteria satisfied	Yes
Water quality criteria satisfied	Not stated in EU DAR, 2003
Study conducted to GLP	Yes
Overall comment on quality	The study is of good quality having been conducted to a standardised methodology under GLP.

Reliability of study	**Reliable**
Relevance of study	**Relevant**
Klimisch Code	**1**

ANNEX II Summary data from OPP 2007

This annex contains information taken from:

- Ecotoxicity Database – an online US EPA database held by the Office of Pesticide Programs that summarises ecotoxicological data used by the EPA for ecotoxicological assessments. Consists primarily of the endpoint data submitted in support of registration and re-registration of pesticide products (OPP 2007).

Data in the summary tables below are classified by the US EPA as 'core' if all essential information was reported and the study was performed according to recommended US EPA or American Society for Testing Materials (ASTM) methodology. Minor inconsistencies with standard recommended procedures may be apparent, but the deviations do not detract from the study's soundness or intent. Studies within this category fulfil the basic requirements of current FIFRA guidelines and are acceptable for use in a risk assessment (equivalent Klimisch code 1). Data not meeting this requirement are classified as either supplemental (Klimisch code 2) or invalid (Klimisch code 3). Supplemental studies are considered scientifically sound, however they were performed under conditions that deviated substantially from recommended protocols. Examples of the conditions that may place a study in this category include: non-native species, tested organisms were older/younger than guideline recommendations, deviations from recommended water quality characteristics (this list is not exhaustive). Where this data has been reported in Tables 2.9 to 2.12 the following notation has been used to identify the US EPA classification: c = core and s = supplemental.

The key data from the EU DAR (2003) were also reported in the US EPA OPP Pesticide Ecotoxicity Database although the toxicity values for certain species were slightly different (see values in bold in Tables A1 and A2). These differences were probably the result of re-interpretation of the data and/or re-calculation of toxicity values.

Table A1 Summary of most sensitive chronic core data taken from OPP 2007

Chemical	Species	Test	Age	Duration	Additional information	LOEC (µg a.i.l^{-1})	NOEC (µg a.i. l^{-1})	Reference (as cited in OPP2007)
Freshwater – algae (Table 2.8)								
92.98% technical grade	*Pseudokirchneriella subcapitata*	[123-2] static GLP (EU DAR 2003)		120 hours		-	3.0	Ref No 42372204 Hughes *et al.* (1992b)
92.98% technical grade	*Navicula pelliculosa*	[123-2] static GLP (EU DAR 2003)		120 hours		-	3.2	Ref No 42372206 Hughes *et al.* (1992a)
Freshwater – higher plants (Table 2.8)								
92.98% technical grade	*Lemna gibba*	[123-2] static GLP (EU DAR 2003)		14 days		-	5.6	Ref No 42137101 Hughes *et al.* (1991)
Freshwater – invertebrates (Table 2.8)								
92.2% technical grade	*Daphnia magna*	[72-4b] GLP (EU DAR 2003)	lifecycle	21 days	Endpoint: mean brood size (USEPA RED 1997)	35.8	14.5	Ref No 247299 Graney (1981)
Freshwater – fish (Table 2.8)								
98.3% technical grade	*Pimephales promelas*	[72-5] flow-through	lifecycle	288 days	Egg production reduced at 9.8 µg l^{-1}; reduced hatch at 22 and 43 µg l^{-1} (USEPA RED 1997)	9.8	6.3	Ref No 00096342 EG & G Bionomics (1975) EPA identification No 00037940 EG G Corp (1977)
Saltwater – algae (Table 2.10)								
92.98% technical grade	*Skeletonema costatum*	[123-2] static		120 hours		-	**0.7**	Ref No 42372205 Hughes *et al.* (1992c)

[72-4b] freshwater invertebrate life cycle chronic toxicity using TGAI or TEP (FIFRA 158.490)
[72-5] Full Fish Life Cycle using TGAI (FIFRA 158.490)
[122-2] Tier I Aquatic Plant Growth – single dose (FIFRA 158.540)
CI = confidence interval
TEP = typical end use product
TGAI = technical grade of the active ingredient

Table A2 Summary of most sensitive acute core data taken from OPP (2007)

Chemical a.i.%	Species	Test	Organism age/ size	Duration	LC/EC50 (µg a.i. l^{-1}) (95% CI)	NOEC (µg a.i. l^{-1})	Curve slope	Reference (as cited in OPP 2007)
Freshwater – algae (Table 2.9)								
92.98% technical grade	*Navicula pelliculosa*	[123-2] static GLP (EU DAR 2003)		120 hours	**6.7 (6 – 7.36)**	3.2	3.5	Ref No 42372206 Hughes *et al.* (1992a)
92.98% technical grade	*Pseudokirchneriella subcapitata*	[123-2] static GLP (EU DAR 2003)		120 hours	**5.4 (2.4 – 10.6)**	3.0	3.3	Ref No 42372204 Hughes *et al.* (1992b)
92.98% technical grade	*Anabaena flos-aquae*	[123-2] static GLP (EU DAR 2003)		120 hours	>174	98	NR	Ref No 42372207 Hughes *et al.* (1992)
Freshwater – higher plants (Table 2.9)								
92.98% technical grade	*Lemna gibba*	[123-2] static (GLP (EU DAR 2003)		14 days	12.5 (10 – 15.7)	5.6	NR	Ref No 42137101 Hughes *et al.* (1991)
Freshwater – invertebrate (Table 2.9)								
93.2% technical grade	*Daphnia magna*	[72-2a] static Immobilization	1st instar	48 hours	280 (230 -330)	160	NR	EG&G Bionomics (1976)
45.6% formulation	*Daphnia magna*	immobilization		48 hours	2,325	-	-	Ref No 260404 Forbis (1985)
94.2% technical grade	*Procambarus simulans*			96 hours	1,000			Ref No 00099889 ABC Inc (1980) (supplemental)
Freshwater – fish (Table 2.9)								
93.2% technical grade	*Oncorhynchus mykiss*	[72-1] static	1.5 g	96 hours	138 (113- 169)	75	7.5	US EPA RED (1997) Ref No 106764 1973
93.2% technical grade	*Lepomis macrochirus*	[72-1] static	1.2 g	96 hours	199 (162 – 244)	100	7.39	US EPA RED (1997) EPA identification: 00046291 Bionomics, Inc (1973)
93.2% technical grade	*Ictalurus punctatus*	[72-1] static	1.5 g	96 hours	418 (310 – 564)	320	NR	US EPA RED (1997) EPA Identification No 00046291 Bionomics, Inc (1973) supplementary

Chemical a.i.%	Species	Test	Organism age/ size	Duration	LC/EC50 (µg a.i. l^{-1}) (95% CI)	NOEC (µg a.i. l^{-1})	Curve slope	Reference (as cited in OPP 2007)
45% a.i. formulation	*Oncorhynchus mykiss*	[72-1] static	1.3 g	96 hours	234 (175 – 310)	94.5	4.3	Ref No 00037927 (1974)
45% a.i. formulation	*Lepomis macrochirus*	[72-1] static	0.9 g	96 hours	414 (320 – 540)	189	5.15	Ref No 00037927 (1974) Bionomics, Inc
45% a.i. formulation	*Ictalurus punctatus*	[72-1] static	2.19 g	96 hours	855 (585 – 1,170)	< 351	NR	Ref No 000251601 (00131773) bionomics, Inc (1983) supplementary
Saltwater – algae (Table 2.11)								
92.98% technical grade	*Skeletonema costatum*	[123-2] static		120 hours	5.2 (3.6 – 4.7)	**0.7**	2.3	Ref No 42372205 Hughes *et al.* (1992)
Saltwater – invertebrates (Table 2.11)								
92.2% technical grade	*Crassostrea virginica*	[72-3b] static	Embryo-larvae	48 hours	210 (160 – 340)	60	NR	Ref No 251601 Ward (1983)
45% formulation	*Crassostrea virginica*	[72-3e] static	Embryo-larvae	48 hours	202 (149 – 320)	112	NR	Ref No 251601 Ward (1983) supplemental
92.2% technical grade	*Penaeus duorarum*	[72-3c] static	1.4 g	96 hours	1,600 (1,200 – 2,200)	< 1000	NR	Ref No 251601 Ward (1983)
45% formulation	*Penaeus duorarum*	[72-3f] static	1.4 g	96 hours	4,950 (4,005 – 7,200)	< 2,790	NR	Ref No 251601 Ward (1983) (supplementary)
Saltwater – fish (Table 2.11)								
92.2% technical grade	*Cyprinodon variegatus*	[72-3a] static GLP (EC 2003)	0.3 g	96 hours	707 (550 – 910)	200	NR	Ref No 251601 Ward (1983) (core)
45% formulation	*Cyprinodon variegatus*	[72-3d] static GLP (EC 2003)	0.3 g	96 hours	765 (554 – 1,602)	225	NR	Ref No 251601 Ward (1983) (core)

[72-1] Freshwater fish acute-warm and coldwater species with TGAI or TEP (FIFRA 158.490)
[72-2] Freshwater invertebrate acute TGAI or TEP
[72-3] Estuarine/marine fish, shellfish, shrimp acute using TGAI or TEP
[122-2] Tier I Aquatic Plant Growth – single dose (FIFRA 158.540)
CI = confidence interval
TEP = typical end use product
TGAI = technical grade of the active ingredient