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

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

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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 members and partners.

This technical document has been developed through a collaborative project, managed and facilitated by the Scotland & Northern Ireland Forum for Environmental Research (SNIFFER), the Environment Agency and the Scottish Environment Protection Agency (SEPA) and has involved the members and partners of UKTAG. It provides background information to support the ongoing development of the standards and classification methods.

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

Note:

This report is an update of report Number SCHO0407BLWF-E-E 'Proposed EQSs for Water Directive Annex VIII substances: Permethrin' produced in 2007 as part of a programme of work commissioned by the UK Technical Advisory Group (UKTAG) to derive Environmental Quality Standards (EQSs) for substances falling under Annex VIII of the Water Framework Directive (WFD). The original report proposed PNECs derived according to the Annex V methodology but because of a lack of certain data, large assessment factors were used in their derivation. This led to the UKTAG concluding that the values were unsuitable for use as EQSs since they were subject to excessive uncertainty, but that this uncertainty may be reduced by appropriate additional ecotoxicity testing [61]. Consequently an ecotoxicity study on the alga *Pseudokirchneriella subcapitata* (Environment Agency 2008) was commissioned with the aim of reducing the data gap, assessment factors and ultimately the uncertainty in the PNEC values [62]. This report incorporates the results of this study and PNECs are re-visited using the more complete dataset. It should be noted that no additional review of any other data/literature that may have been published since the original 2007 report has been made.

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 permethrin using the methodology described in Annex V of the Directive. There are existing EQSs for permethrin, but they were derived using a method not considered to comply with the requirements of Annex V and so cannot be used to derive Annex VIII EQSs.

The PNECs described in this report are based on a technical assessment of the available ecotoxicity data for permethrin, along with any data that relate impacts under field conditions to exposure concentrations. The data have been subjected to rigorous quality assessment such 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. This report is an update of report Number SCHO0407BLWF-E-E 'Proposed EQSs for Water Directive Annex VIII substances: Permethrin' produced in 2007 as part of a programme of work commissioned by the UK Technical Advisory Group (UKTAG) to derive Environmental Quality Standards (EQSs) for substances falling under Annex VIII of the Water Framework Directive (WFD). The original report proposed PNECs derived according to the Annex V methodology but because of a lack of certain data, large assessment factors were used in their derivation. This led to the UKTAG concluding that the values were unsuitable for use as EQSs since they were subject to excessive uncertainty, but that this uncertainty may be reduced by appropriate additional ecotoxicity testing [61]. Consequently an ecotoxicity study on the alga *Pseudokirchneriella subcapitata* (Environment Agency 2008) was commissioned with the aim of reducing the data gap, assessment factors and ultimately the uncertainty in the PNEC values [62]. This report incorporates the results of this study and PNECs are re-visited using the more complete dataset. It should be noted that no additional review of any other data/literature that may have been published since the original 2007 report has been made.

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
Permethrin is a synthetic pyrethroid insecticide with a wide range of applications. It has four isomers (its *cis*- and *trans*-isomers both have two optical isomers) and is a potent neurotoxin. Permethrin is relatively nontoxic to mammals but very toxic to certain forms of aquatic life.

In water, permethrin is hydrolytically stable but readily biodegradable. It also undergoes photolysis. In general, the degradative processes are more rapid with the *trans*-isomer and both isomers degrade to less toxic products. Permethrin is lipophilic (log Kow 3.48–6.5) and has been found to sorb strongly to sediment, where it is persistent.

Availability of data
Acute toxicity data are available for six different freshwater taxonomic groups (algae, crustaceans, fish, amphibians, insects and molluscs); chronic data are available for algae, crustaceans, fish, insects and molluscs. Laboratory data are supplemented by pond and stream mesocosm studies.

By comparison, the toxicity data available for marine organisms represent just four taxonomic groups (algae, crustaceans, fish and molluscs), with only one chronic test found for fish.

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

Two publications on permethrin toxicity in sediment were found.

Derivation of PNECs

Long-term PNEC for freshwaters
As expected from the mode of action of permethrin, crustaceans and insects appear to be the most sensitive taxonomic groups.

Based on the available data, the lowest good quality long-term NOEC is a value of 0.029 μg l^{-1} for the stonefly *Pteronarcys dorsata*. In the study with *P. dorsata*, however, a very steep concentration response was observed with no effect at 0.029 μg l^{-1}, but 100 per cent immobilisation at 0.042 μg l^{-1} after 28 days [12]. However, In the same study, the caddisfly *Brachycentrus americanus* suffered 55 per cent mortality at 0.03 μg l^{-1} (the lowest concentration tested) and no NOEC value could be determined. Since the effects level is greater than 20% the TGD approach cannot be used to derive a NOEC from the LOEC. Therefore, it is proposed that the data for *B.americanus* is used in a supporting role.

The lowest reliable NOEC is the value of 0.029 μg l^{-1} for the stonefly *Pteronarcys dorsata*. As good quality long-term NOECs are available for a range of taxa (crustaceans, insects and fish) and, given the mode of action of permethrin, the most sensitive organisms are represented, an assessment factor of 10 could be used to derive the PNEC:

$PNEC_{freshwater\_lt}$ = (0.03 µg l^{-1} permethrin)/AF (10) = 0.003 µg l^{-1} permethrin

The TGD also proposes the derivation of the PNEC from acute data with an AF of 100 if acute effect data are available that are lower than the lowest long-term NOEC. The short-term database contains two 50 per cent effect concentrations at low concentrations of permethrin (*Oncorhynchus mykiss* LC50 of 0.014 µg l^{-1} and a *Daphnia magna* 96-hour LC50 of 0.039 µg l^{-1}).

Both values are likely to be outliers but, if the process were followed through, using the lowest reliable E(L)C50 (*Hexagenia bilineata* 96-hour LC50 of 0.1 µg l^{-1}) and applying an AF of 100 would generate a PNEC of 0.001 µg l^{-1} permethrin. These PNEC values are supported by the data from the freshwater mesocosm studies described in Section 2.6.6 which show that effects in complex natural systems may be observed at very low permethrin concentrations, which are close (with a factor <5) to the PNEC based on single species tests.

Based on the review of the available data it is proposed that the PNEC of 0.001 µg l^{-1} derived using short-term data is applied as the long-term value. This value provides a margin of safety with respect to the significant effects of permethrin on the survival of the caddisfly *Brachycentrus americanus* at 0.03 µg l^{-1}.

This is 10 times lower than the existing EQS of 0.01 µg l^{-1} total permethrin expressed as a 95th percentile. This was based on field and laboratory data that suggested levels <0.01 µg l^{-1} would be unlikely to affect aquatic invertebrates or dependent fisheries.

Short-term PNEC for freshwaters

The acute data show crustaceans and insects, followed by salmonid fish, to be the most sensitive taxonomic groups.

It is recommended that the short-term PNEC is derived on the basis of a 96-hour LC50 of 0.1 µg l^{-1} for the mayfly *Hexagenia bilineata* and guidance given in the EU Technical Guidance Document (TGD) on effects assessment for intermittent releases. Given that permethrin is a neurotoxin with a specific mode of action and that insects belong to the most sensitive organisms, a reduced assessment factor of 10 (instead of 100) is recommended in order to extrapolate from the 50 per cent acute effect level to the short-term no-effect level. This results in a $PNEC_{freshwater\_st}$ of 0.01 µg l^{-1}.

The available field studies support this suggested value. There is no existing short-term EQS for permethrin.

Long-term PNEC for saltwaters

The data suggest that there are no obvious differences between freshwater and saltwater species from the same taxonomic groups. Because of this and the lack of marine data, the freshwater and saltwater datasets were combined.

Therefore, the long-term PNEC for saltwater was derived on the same basis as the freshwater PNEC i.e. using the using the lowest reliable E(L)C50 (*Hexagenia bilineata* 96-hour LC50 of 0.1 µg l^{-1} and applying an AF of 100 to generate a PNEC of 0.001 µg l^{-1} permethrin . The TGD suggests a total assessment factor of 1000 if three long-term tests are available for three taxonomic groups, with a factor of 10 applied to account for the absence of data for marine species. However, short-term tests with additional marine species are available and a reduced assessment factor of 500 is recommended. These acute marine data indicate that molluscs are one of the least sensitive groups and would be protected by the proposed $PNEC_{saltwater\_lt}$ of 0.0002 µg l^{-1}.

This proposed PNEC is considerably lower than the existing EQS of 0.01 µg l^{-1}, which was 'read across' from the long-term freshwater EQS.

Short-term PNEC for saltwaters

Crustaceans appear to be the most sensitive taxonomic group.

The lowest acute value was the geometric mean 96-hour LC50 of 0.052 µg l^{-1} for the shrimp, *Americamysis bahia*, calculated from empirical LC50 values from a number of good quality studies. As with the freshwater PNEC, it is recommended that the PNEC be derived on the basis of general guidance given in the TGD on effects assessment for intermittent releases. Because permethrin acts specifically on the nervous system and crustaceans belong to the most sensitive organisms, a reduced assessment factor of 50 (instead of 100) is recommended in order to extrapolate from the 50 per cent acute effect level to the short-term no-effect level. This results in a $PNEC_{saltwater\_st}$ of 0.001 µg l^{-1}.

There is no existing short-term EQS for permethrin.

PNEC for secondary poisoning

For both freshwater and saltwater, PNECs based on the risks of secondary poisoning to mammals and birds (1.75 µg l^{-1}) are higher than those derived for the protection of aquatic life and so do not influence the development of EQSs for permethrin.

PNEC for sediments

Because the log Kow is >3, the derivation of a PNEC for the protection of benthic communities is required.

Two sediment studies are available and both the 10-day LC50 of 2.11 mg permethrin/kg sediment and the >20-day NOEC of 0.4 mg permethrin/kg sediment are suitable for PNEC derivation. Using the chronic toxicity data and the appropriate assessment factors of 100 (chronic) for freshwater and 1,000 (chronic) for saltwater results in a $PNEC_{sediment\_freshwater}$ of 4.0 µg permethrin/kg sediment dry weight (dw), and a $PNEC_{sediment\_saltwater}$ of 0.4 µg permethrin/kg sediment dry weight (dw), respectively.

Summary of proposed PNECs

Receiving medium/exposure scenario	Proposed PNEC (µg l^{-1}permethrin)	Existing EQS (µg l^{-1})
Freshwater/long-term	0.001	0.01
Freshwater/short-term	0.01	–
Saltwater/long-term	0.0002	0.01
Saltwater/short-term	0.001	–
Freshwater sediment/long-term	4.0 µg/kg dw	No standard
Saltwater sediment/long-term	0.4 µg/kg dw	No standard
Freshwater secondary poisoning	1.75	No standard
Saltwater secondary	1.75	No standard

Analysis

The lowest proposed PNECs derived for permethrin are 0.3 ng l^{-1} for waters and 0.4 µg/kg for sediments. The data quality requirements are that, at a third of the EQS, total error of measurement should not exceed 50 per cent. From the literature, it can be seen that analytical methodologies are capable of achieving detection limits in the low µg l^{-1} order in most media, suggesting that current analytical methods would not be adequate to analyse permethrin for compliance purposes.

Implementation issues

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

- Current analytical methods may not be sensitive enough to assess compliance with proposed PNECs in receiving waters. This will require further consideration.

- Additional marine toxicity data would be required to reduce the size of the assessment factor applied in the derivation of the saltwater PNECs.

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 permethrin using the methodology described in Annex V of the Directive. There are existing EQSs for permethrin but they were derived using a method not considered to comply with the requirements of Annex V and so cannot be used to derive Annex VIII EQSs.

The PNECs described in this report are based on a technical assessment of the available ecotoxicity data for permethrin, along with any data that relate impacts under field conditions to exposure concentrations. The data have been subjected to rigorous quality assessment such 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. This report is an update of report Number SCHO0407BLWF-E-E 'Proposed EQSs for Water Directive Annex VIII substances: Permethrin' produced in 2007 as part of a programme of work commissioned by the UK Technical Advisory Group (UKTAG) to derive Environmental Quality Standards (EQSs) for substances falling under Annex VIII of the Water Framework Directive (WFD). The original report proposed PNECs derived according to the Annex V methodology but because of a lack of certain data, large assessment factors were used in their derivation. This led to the UKTAG concluding that the values were unsuitable for use as EQSs since they were subject to excessive uncertainty, but that this uncertainty may be reduced by appropriate additional ecotoxicity testing [61]. Consequently an ecotoxicity study on the alga *Pseudokirchneriella subcapitata* (Environment Agency 2008) was commissioned with the aim of reducing the data gap, assessment factors and ultimately the uncertainty in the PNEC values [62]. This report incorporates the results of this study and PNECs are re-visited using the more complete dataset. It should be noted that no additional review of any other data/literature that may have been published since the original 2007 report has been made.

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] *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 1.

1.1 Properties and fate in water

Permethrin is a synthetic pyrethroid insecticide with a wide range of applications. It has four isomers (its *cis*- and *trans*-isomers both have two optical isomers) and is a potent neurotoxin. Permethrin is relatively nontoxic to mammals but very toxic to certain forms of aquatic life.

In water, permethrin is hydrolytically stable but readily biodegradable. It also undergoes photolysis. In general, the degradative processes are more rapid with the *trans*-isomer and both isomers degrade to less toxic products. Permethrin is lipophilic (log Kow 3.48–6.5) and has been found to sorb strongly to sediment, where it is persistent.

2. Results and observations

2.1 Identity of substance

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

Table 2.1 Substance covered by this report

Name	CAS Number
Permethrin	52645-53-1

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 [45], and existing EQSs obtained from the literature [53].

Section 2.6 summarises the effects data identified from the literature for permethrin. 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
Freshwater short-term	0.01 µg l^{-1} (Section 3.1.1)	-	-
Freshwater long-term	0.001 µg l^{-1} (Section 3.1.1)	Insufficient data	0.01 µg l^{-1} (95th percentile)
Saltwater short-term	0.001 µg l^{-1} (Section 3.1.2)	-	-
Saltwater long-term	0.0002 µg l^{-1} (Section 3.1.2)	Insufficient data	0.01 µg l^{-1} (95th percentile)
Freshwater sediment long-term	4.0 µg/kg dw (Section 3.4)	Insufficient data	-
Saltwater sediment long-term	0.4 µg/kg dw (Section 3.4)	Insufficient data	-
Freshwater secondary poisoning	1.75 µg l^{-1} (Section 3.5)	-	-
Saltwater secondary poisoning	1.75 µg l^{-1} (Section 3.5)	-	-

AF = assessment factor
SSD = species sensitivity distribution

2.3 Hazard classification

Table 2.3 gives the R-phrases (Risk-phrases) and labelling for the substance of interest.

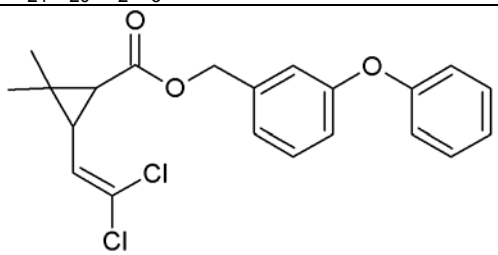
Table 2.3 Hazard classification

R-phrases and labelling	Reference
Xn; R20/22 R43 N; R50-53	[1]

2.4 Physical and chemical properties

Table 2.4 summarises the physical and chemical properties of the substance of interest.

Table 2.4 Physical and chemical properties of permethrin

Property	Value	Reference
Molecular formula	$C_{21}H_{20}Cl_2O_3$	[46]
Molecular structure		
Vapour pressure	1.3 µPa at 20°C [technical grade; pure: 2.5 µPa (*cis*), 1.5 µPa (*trans*)] 45 µPa at 25°C 4.5×10^{-7} mbar at 25°C 2.18×10^{-8} mmHg at 25°C	[2] [5] [6] [7]
Henry's Law constant	1.9×10^{-6} atm-m^3/mol	[7]
Solubility in water	0.2 mg l^{-1} at 30°C 0.2 mg l^{-1} at 20°C 0.04 mg l^{-1} at room temperature	[2] [5] [6]
Relative molecular weight	391.31	[2]

2.5 Environmental fate and partitioning

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

Table 2.5 Environmental fate and partitioning of permethrin

Property	Value	Reference
Hydrolytic stability (DT50)	At pH 5 and 7, permethrin is stable towards abiotic hydrolysis; at pH 9, the abiotic hydrolysis rate constant is 0.0139 per day at 25°C, which corresponds to a half-life of 50 days.	[7]

Property	Value	Reference
Photostability (DT50) (aqueous, sunlight, state pH)	In water and on soil surfaces, permethrin is photodegraded by sunlight. Ester cleavage and *cis–trans* interconversion are the major reactions. Permethrin deposited on plants degrades with a half-life of approximately 10 days. In water, the photolysis rate constant is 0.021 per day; this corresponds to a photodegradation half-life of 33 days. Photolysis half-lives of 27.1 and 19.6 hours were determined for respective *cis*- and *trans*-isomers in 800 ml pond water exposed to sunlight. The photolysis half-life of permethrin in seawater exposed to outdoor light was determined to be 14 days.	[2] [2] [7]
Readily biodegradable (yes/no)	Yes (in waters)	[53]
Degradation in water/sediment DT50 in water DT50 whole system Mineralisation Bound residue	 60 days (*trans*), 67 days (*cis*) The biodegradation half-life of permethrin in a sediment–seawater solution was less than 2.5 days; under sterile conditions there was no significant change in permethrin concentration. – –	 [4] [7]
Distribution in water/sediment systems (active substance)	Permethrin disappears rapidly from the environment, in 6–24 hours from ponds and streams and 7 days from pond sediment. The persistence of permethrin in water and sediment contained in open trenches (3 m × 1 m × 30 cm) lined with Alkathene sheet was investigated by spraying insecticide emulsion on the surface of the water at the normal recommended dosage and at twice this value. The dissipation of the insecticide from the water was rapid, with about 87–90% of the pesticide being lost within 24 hours at both rates of application. However, residues were found to be absorbed by the sediment and these persisted beyond 30 days.	[2] [50]
Distribution in water and sediment systems (metabolites)	–	

Property	Value	Reference
Residues relevant to the aquatic environment	In general, the degradative processes which occur in the environment lead to less toxic products.	[2]
Degradation in soil	DT50 ≤28 days in laboratory studies. The *trans*-isomer degraded more rapidly than the *cis*-isomer, ester cleavage being the major initial degradative reaction. The compounds generated by ester cleavage were then further oxidised, eventually yielding carbon dioxide as the major terminal product. Studies to investigate the leaching potential of permethrin and its degradates showed that very little downward movement occurs in soil.	[2]
	Low mobility in soil, DT50 <38 days	[5]
	Under anaerobic conditions in flooded silt loam soils, degradation half-lives were 32–34 days for *trans*-permethrin and greater than 64 days for ^{14}C-labeled *cis*-permethrin.	[7]
	Field dissipation half-lives for permethrin range from 6 to 106 days.	[7]
Partition coefficients log Kow	 6.5 6.1 at 20°C 3.48	 [2, 7] [4, 5] [6]
Koc	10,471–86,000 4.39 (log Koc) (24,550)	[7] [15]
Ksed	652 l/kg 389 l/kg	[4] [2]
Bioconcentration factor (BCF) General	Absorbed permethrin is rapidly lost on transfer to clean water.	[2]
	560	
<u>Fish</u> *Oncorhynchus mykiss* *Oncorhynchus mykiss* *Cyprinodon variegatus* *Cyprinodon variegatus* *Salmo salar* *Cyprinus carpio*	30 (blood), 30 (muscle), 300 (liver), 400 (fat) 480 290–620 55 330–750 1,900 (28-day steady state BCF)	 [7] [2] [7] [2] [2] [2]
<u>Molluscs</u> *Crassostrea virginica*		 [38]

Property	Value	Reference
	18	
Other	30	
Blackfly	7	[7]
Caddisfly	4	[7]
Damselfly	24	[7]
Water scavenger	43–570	[7]
Mayfly		[7]
Stonefly (*Pteronarcys dorsata*)		[2]

DT50 = time taken to degrade by 50%

Permethrin is a widely used contact insecticide. It is relatively nontoxic to mammals but very toxic to aquatic life. Monitoring data from the Environment Agency in the period 1995 to 2004 showed that the percentage of samples over 0.1 µg l^{-1} or the limit of detection ranged from 0.0% (in 2003 and 2004) to 2.7% (in 1998). The data indicate that in the monitoring period the percentage of samples over 0.1 µg l^{-1} or the limit of detection had declined from 2001 and was not above 0.2% in the period from 2001 to 2004.

In a laboratory adsorption–desorption study, more than 95 per cent of permethrin in aqueous solutions (6–42 µg l^{-1}) was rapidly adsorbed onto lake sediment and the adsorbed insecticide was not readily desorbed from the sediment by several water rinses. A high distribution coefficient of 389 l/kg was obtained from the adsorption isotherm. Permethrin in aqueous solution applied to the surface of a sediment column did not penetrate through more than 2 cm of the sediment [2].

During field tests in Canada where permethrin was sprayed in forests at 17.5 g active ingredient per hectare (a.i./ha), residues in water persisted for less than 96 hours. Accumulation of the pesticide in the bottom sediment of ponds was negligible and it persisted for less than 7 days. No (or only minimal) permethrin residues were found in stream sediments. The sprayed permethrin formulation had a density (0.88 g ml^{-1}) less than that of water and was practically insoluble in water. It therefore formed a surface film when brought into contact with stagnant or slowly moving water. This significantly reduced the likelihood of the insecticide reaching the bottom sediment or exposing fish in the treated ponds and streams [2].

However, a study on the occurrence and mobility of permethrin in rivers of the Southern Humber catchment in the UK [3] where discharges from the textile finishing industry and sewage treatment plants are major sources of permethrin demonstrated the permanent presence of permethrin in ‘whole waters’ (water plus suspended sediment) and (bed) sediments, particularly in the rivers Aire and Calder, and at concentrations in the sediment likely to cause ecotoxicological effects. Retention times of permethrin were estimated as 4–26 days in suspended sediments and 103–125 days in surface bed sediments. In another study in experimental fluvarium channels [4], half-lives of *cis*- and *trans*-permethrin in river water were found to be 67 and 60 days, respectively. The presence of natural sediment enhanced the removal of permethrin from the overlying water with penetration to 20 mm depth in 43 days. Overall, the sediment was a sink for permethrin with 97 per cent of the total permethrin in the sediment bed.

Investigation of a large accidental contamination of two Swiss rivers with permethrin involved field surveys to study the recovery of the rivers [5]. These surveys revealed that permethrin persisted for an extended period of time (5 months) in sediments and the food web.

Studies indicate that permethrin and its degradation products (certain carboxylic acid metabolites formed in the soil) may be taken up by plants from soil [2]. However, under field conditions, no residues of permethrin or its metabolites were detected in crops sown 60 days or more after soil treatment. Very little translocation of permethrin or its metabolites was observed following either topical application or stem injection of permethrin to plants [2]. Photochemical reactions played an important role in the fate of permethrin applied to the surface of plants. A major degradation pathway in plants was ester cleavage; this was followed by rapid conjugation with sugars of the two cleavage products, i.e. 3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropanecarboxyclic acid (Cl_2CA) and 3-phenoxybenzyl alcohol (Pbalc) [2].

2.6 Effects data

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

Data collation followed a tiered approach. For freshwater and saltwater data, critical data from the existing UK EQS document [53] were collated. Further data published after derivation of the current UK EQS were then retrieved from the US Environmental Protection Agency (US EPA) ECOTOX database.[3]

As no information on sediment-dwelling organisms, mammalian or avian chronic oral toxicity was available in ECOTOX, further databases were searched via the STN portal. Further data sources used were:

- Hazardous Substances Data Bank (HSDB®) database of the US National Library of Medicine;[4]
- US EPA Integrated Risk Information System (IRIS) database;[5]
- World Health Organization (WHO) *Environmental Health Criteria 94: Permethrin* [2].

The PNECs were derived using data from studies using technical grade permethrin rather than the microencapsulated substance or formulations and were based on concentrations of the active ingredient.

Only two publications on permethrin toxicity in sediment (mg permethrin/kg sediment) could be identified.

2.6.1 Toxicity to freshwater organisms

Single species test results for acute and chronic toxicity data are available for six different taxonomic groups: acute data for algae, crustaceans, fish, amphibians, insects

[3] http://www.epa.gov/ecotox/
[4] http://toxnet.nlm.nih.gov/cgi-bin/sis/htmlgen?HSDB
[5] http://www.epa.gov/iris/index.html

and molluscs; and chronic data for algae, crustaceans, fish, insects and molluscs. Table 2.6 summarises the long-term data and Table 2.7 the short-term data. Aquatic insect larvae and crustaceans, followed by salmonid fish, appear to be the groups most sensitive to permethrin. Molluscs, amphibians and algae are less sensitive.

Diagrammatic representations of the available freshwater data for permethrin (cumulative distribution functions) are presented in Figures 2.1 and 2.2. These diagrams include all data regardless of quality and provide an overview of the spread of the available data. These diagrams are not species sensitivity distributions and have not been used to set the permethrin PNECs. The lowest critical freshwater data for permethrin are presented in Tables 2.6 and 2.7. As well as the single species tests, there are some publications on field tests with permethrin in lentic and lotic waters. Test designs and results are summarised in Table 2.8.

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

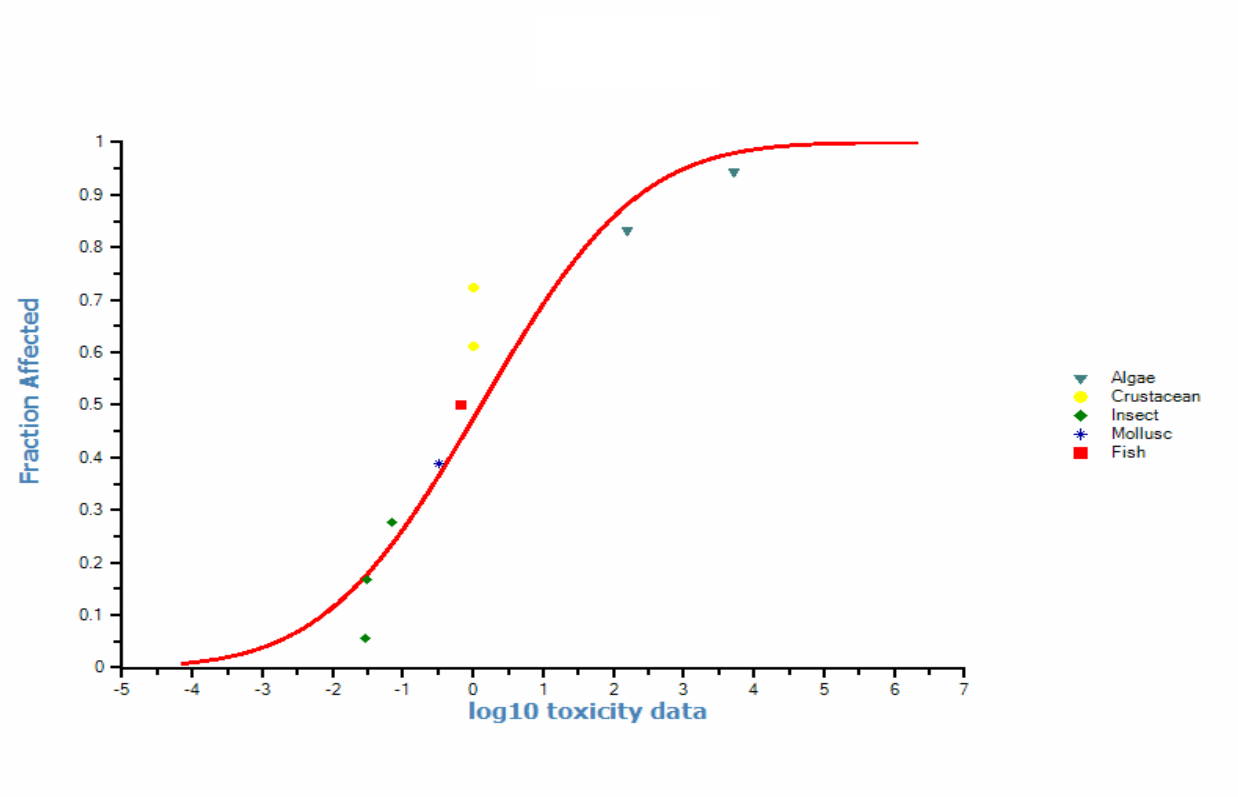


Figure 2.2 Cumulative distribution function of freshwater short-term data (µg a.i. l^{-1}) for permethrin

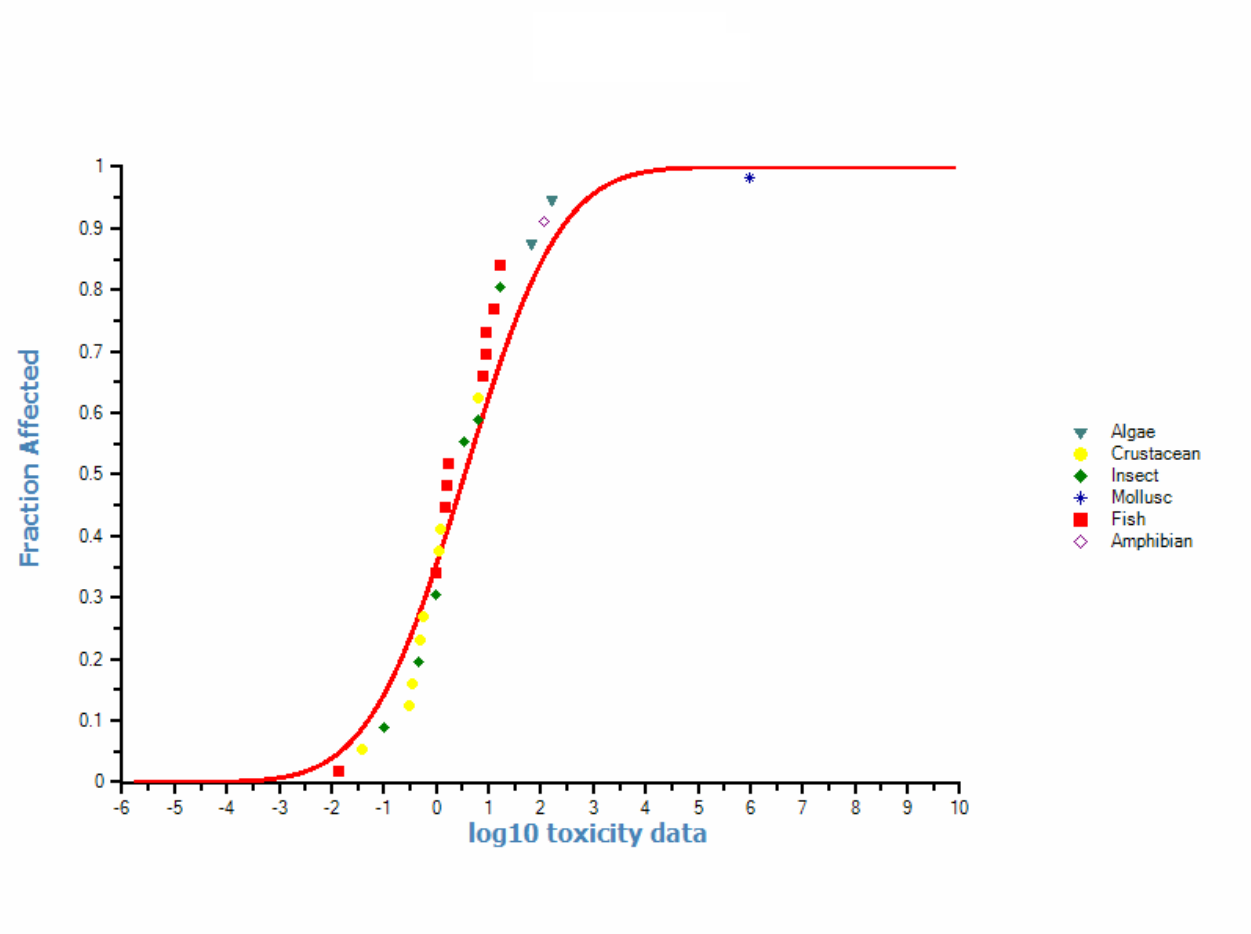


Table 2.6 Long-term aquatic toxicity data for freshwater organisms exposed to permethrin

Scientific name	Common name	Taxonomic group	End-point	Effect	Test duration	Conc. (µg a. i. l^{-1})[1]	Exposure[2]	Toxicant analysis[3]	Comments	Reliability index[4]	Ref.
Chlamydomonas reinhardtii	Green algae	ALG	EC10	GPOP	3 days	5100	s	n	EC0 is 4,700 µg l^{-1}, a 72-hour EC10 of ~5,100 µg l^{-1} can be deduced from Figure 1 of the publication	3	[18]
Pseudokirchneriella subcapitata	Green algae	ALG	NOEC	Growth (growth rate and biomass)	3 days	**160**	s	y	22±2°C	2	[62]
Daphnia magna	Water flea	CRU	NOEC (LOEC)	MOR	40 days	1 (5)	s	n	Microencapsulated permethrin formulation; LOEC already caused >50% mortality	2	[39]
Daphnia pulex	Water flea	CRU	NOEC (LOEC)	MOR	32 days	**<1** (1)	s	n	Microencapsulated permethrin formulation; NOEC survival <1 µg l^{-1} (the lowest concentration tested); LOEC 1 µg l^{-1} but already caused >90% mortality	2	[39]
Brachycentrus americanus	Caddisfly	INS	NOEC LC50	MOR	28 days 21 days	**<0.03** 0.17	f	m	LOEC 0.03 µg l^{-1} but, at this concentration, more than 55% of the exposed individuals were dead after 28 days	2	[12]
Hexagenia rigida	Mayfly	INS	21% effect	MOR	8 weeks	0.15	s	m	Treatment related mortality during 8 weeks observation in clean water after 6 hours exposure to 0.15 µg l^{-1}. NOEC estimate according to TGD provisions is approximately **0.07 µg l^{-1}**	2	[17]
Pteronarcys dorsata	Stonefly	INS	EC100	IMBL	21 days	0.042	f	m	100% immobilisation after 21 days	2	[12]
Pteronarcys dorsata	Stonefly	INS	NOEC	MOR, IMBL	28 days	**0.029**	f	m	-	2	[12]
Helisoma trivolvis	Snail	MOL	NOEC	MOR	28 days	>0.33	f	m	Unbounded NOEC	2	[40]
Pimephales promelas	Fathead minnow	FIS	NOEC	MOR	32 days	**0.66**	f	m		2	[40]

[1] The lowest no observed effect concentrations (NOECs) per taxonomic group are highlighted in bold. [2] Exposure: s = static; f = flow-through. [3] Toxicant analysis: m = measured; n = nominal. [4] The reliability index (RI) is assigned according to the Klimisch Criteria, defined in Annex 1. For data relevant for PNEC derivation, Data Quality Assessment Sheets are available in Annex 1 and Data Proformas in Annex 2.
ALG = algae; CRU = crustaceans; FIS = fish; INS = insects; MOL = molluscs
IMBL = immobilisation; MOR = mortality; GPOP = population growth
NOEC = no observed effect concentration; LOEC = lowest observed effect concentration
ECx = concentration effective against X% of the organisms tested; LC50 = concentration lethal to 50% of the organisms tested

Table 2.7 Short-term aquatic toxicity data for freshwater organisms exposed to permethrin

Scientific name	Common name	Taxonomic group	End-point	Effect	Test duration (hours)	Conc. (µg a.i. l^{-1})[1]	Exposure[2]	Toxicant analysis[3]	Comments	Reliability index[4]	Ref.
Diatomeae		ALG	EC50	-	96	**68**	-	-	-	4	[5]
Pseudokirchneriella subcapitata	Green algae	ALG	EC50	GRO (growth rate and biomass)	72	>160	s	y	22±2°C	2	[62]
Asellus aquaticus	Water hog louse	CRU	(EC50)*	NR	NR	**0.3**	-	-	*'Mean threshold for acute effects'	-	[10]
Ceriodaphnia dubia	Water flea	CRU	LC50	MOR	48	**0.55**	s	m	-	2	[32]
Daphnia magna	Water flea	CRU	EC50	ITX/ IMBL	48	0.112 0.32 0.6 7.2	-	-	GM 0.63 µg l^{-1}	2	[34]
Daphnia magna	Water flea	CRU	LC50	MOR	48	1.25	s	m	-	2	[32]
Daphnia magna	Water flea	CRU	EC50	ITX/ IMBL	96	**0.039**	-	-	Concentration may not be valid	2	[34]
Daphnia pulex	Water flea	CRU	LC50	MOR	48	2.75 7.45 13.1	-	-	GM 6.45 µg l^{-1}	-	[39]
Gammarus pulex	Shrimp	CRU	LC50 (NOEC)	MOR	96	**0.34** (0.03)	-	-	-	-	[30]
Procambarus clarkii	Red swamp crayfish	CRU	LC50	MOR	96	0.282	s	m	**GM 0.48 µg l^{-1}** – size class 8–12 mm	2	[24]
						0.39	s	n	Size class 8–12 mm	2	[24]
						0.499	s	m	Size class 8–12 mm	2	[24]
						0.532	s	m	Size class 8–12 mm (GM 25–35 mm 0.84 µg l^{-1}; GM 45–55 mm 1.3 µg l^{-1}; GM 65–75 mm 0.8 µg l^{-1})	2	[24]
Aedes aegypti	Yellow fever mosquito	INS	LC50	MOR	24	**0.45**	s	m	Technical permethrin	2	[35]
Aedes albopictus	Mosquito	INS	LC50	MOR	24	0.95	-	-	-	-	[11]

Scientific name	Common name	Taxonomic group	End-point	Effect	Test duration (hours)	Conc. (µg a.i. l^{-1})[1]	Exposure[2]	Toxicant analysis[3]	Comments	Reliability index[4]	Ref.
Aedes atropalpus	Mosquito	INS	LC50	MOR	24	6.168	-	-	-	-	[14]
Aedes hendersoni	Mosquito	INS	LC50	MOR	24	3.507	-	-	-	-	[14]
Aedes triseriatus	Mosquito	INS	LC50	MOR	24	4.46 6.23 6.39 7.38 7.68 8.39	-	-	**GM 6.62 µg l^{-1}**	-	[14]
Chironomus riparius	Midge	INS	LC50	MOR	24	34.4	-	-	Time dependency of toxicity	2	[15]
					48	9.27					
					72	4.62					
					96	2.89					
Chironomus thummi	Midge	INS	LC50	MOR	24	16.6	-	-	-	-	[23]
Hexagenia bilineata	Mayfly	INS	LC50	MOR	96	**0.1**	-	-	-	2	[34]
Lymnaea stagnalis	Great pond snail	MOL	LC50	MOR	48	100000	-	-	-	2	[34]
Catostomus commersoni	White sucker	FIS	LC50	MOR	2	**1** 10	s	m	20 days old, unfed 20 days old, fed (mortality observed after 2-hour pulse exposure plus 94-hour observation time)	2	[22]
Gambusia affinis	Western mosquitofish	FIS	LC50	MOR	96	6.3 12	- -	- -	GM 8.7 µg l^{-1}	- -	[13] [33]
Lepomis macrochirus	Bluegill	FIS	LC50	MOR	96	0.79 2.52 6.1 6.8 9 13.3 13.5 33.4 5.1	- - 	- - 	GM 6.54 µg l^{-1}	2 - 	[34] [13]
Oncorhynchus clarki	Lahontan	FIS	LC50	MOR	96	**1.6**	s	m	-	2	[37]

Scientific name	Common name	Taxonomic group	End-point	Effect	Test duration (hours)	Conc. (µg a.i. l^{-1})[1]	Exposure[2]	Toxicant analysis[3]	Comments	Reliability index[4]	Ref.
henshawi	cut-throat trout										
Oncorhynchus clarkii stomias	Greenback cut-throat trout	FIS	LC50	MOR	96	**>1**	s	m	-	2	[37]
Oncorhynchus gilae apache	Apache trout	FIS	LC50	MOR	96	**1.7**	s	m	-	2	[37]
Oncorhynchus kisutch	Coho salmon, silver salmon	FIS	LC50	MOR	96	**17**	-	-	-	2	[34]
Oncorhynchus mykiss (≈ Salmo gairdneri)	Rainbow trout	FIS	LC50	MOR	144	**0.014**	-	-	Result is given as µg l^{-1} but may be µmol l^{-1}, which equals 5.5 µg l^{-1}, which would then agree with other *O. mykiss* acute mortality data	4	[9]
Oncorhynchus mykiss	Rainbow Trout	FIS	LC50	MOR	96	5.5	-	-	**GM 5.88 µg l^{-1}**	-	[13]
						2.1	-	-		-	[34]
						5.3					
						9.8					
						20.9					
						3.3	-	-		-	[37]
Pimephales promelas	Fathead minnow	FIS	LC50	MOR	96	3	-	-	GM 12.96 µg l^{-1}	2	[34]
						9.4	-	-		-	[37]
						16	-	-		-	[19]
						62.6	-	-		-	[13]
Salmo salar	Atlantic salmon	FIS	LC50	MOR	96	**1.5**	-	-	-	2	[34]
Rana catesbeiana	Bullfrog	AMP	LC50	MOR	96	115	-	-	-	-	[13]

[1] The lowest L(E)C50s per taxonomic group are highlighted in bold. If more than one test per species with the same endpoint and test duration was available, geometric means (GMs) of these results were calculated. The GMs are presented in the 'Comments' column.
[2] Exposure: s = static.
[3] Toxicant analysis: m = measured; n = nominal.

[4] The reliability index (RI) is assigned according to the Klimisch Criteria, defined in Annex 1. For data relevant for PNEC derivation, Data Quality Assessment Sheets are available in Annex 1 and Data Proformas in Annex 2.
ALG = algae; AMP = amphibians; CRU = crustaceans; FIS = fish; INS = insects; MOL = molluscs
ITX/IMBL = intoxication/immobilisation; MOR = mortality; NR = not reported
LC50 = concentration lethal to 50% of the organisms tested; EC50 = concentration effective against 50% of the organisms tested
NOEC = no observed effect concentration

Table 2.8 Permethrin toxicity observed in field tests

Laboratory and field response of *Chironomus riparius* to a pyrethroid insecticide	**Reference: [15]**	**Reliability index: 2[1]**
Study NOEC: 1 µg l^{-1} (initial nominal concentration). Post-application observation period 52 days		
Exposure: Static, artificial ponds, one single application of permethrin to achieve 0 (control), 1, 10, 50 and 100 µg l^{-1}	Analysis: No analysis of permethrin in water. Study NOEC refers to initial nominal concentration. Permethrin residues in sediment were analysed.	
Description of test site/test facility: Five ponds, 5 × 5 m surface area, sloping to 4 × 4 m at the bottom, lined with butyl rubber pond liner at the premises of WRc plc, Medmenham, UK. The ponds contained 5–10 cm sediment layer from the CS Lewis Nature Reserve, Oxford (a known clean site) and a 60 cm depth of uncontaminated water from the River Thames. Plants and invertebrates were present in the ponds through natural colonisation, although a dense growth of pond weed (mostly *Elodea canadensis*) was removed by raking 27 days before dosing. A regression design was used for the experiments and the ponds were dosed with the commercially available formulation 'Picket' (Zeneca Agrochemicals, UK) at the beginning of July to achieve initial nominal concentrations of 0 (control), 1, 10, 50 and 100 µg l^{-1} permethrin.	Observations/effects: The pH in all ponds was high, ranging between 9 and 10.5. Dissolved oxygen remained high [90–120% by anodic stripping voltammetry (ASV)], often supersaturated throughout the study. Temperatures at the pond surface were 20–30°C, whereas those at the bottom were 10–15°C. Effects of permethrin application on water quality were not apparent. *Elodea canadensis* rapidly recolonised the ponds and no differences in weed density could be observed at the end of the study. Knockdown of aquatic invertebrates, particularly hemipterans, was observed immediately after spraying of the ponds dosed with the highest concentrations (100, 50 and 10 µg l^{-1}). On day 2 post dosing, dead chironomid larvae were found in sediment grab samples from ponds dosed at 50 and 100 µg l^{-1}. No emergence of chironomid adults was seen in ponds dosed with 50 and 100 µg l^{-1} until days 24 and 31, respectively. At 10 µg l^{-1}, insects were collected at all sampling dates but numbers were significantly reduced relative to the control until day 24 post treatment. Chironomid emergence at 1 µg l^{-1} was similar to the control. Regression analysis revealed that dose had a significant effect on abundance of the chironomids. The highest measured concentration in the sediment of the highest pond treatment (100 µg l^{-1}) was 217 µg/kg, which is an order of magnitude lower than the 10-day LC50 of 2,110 µg/kg for *C. riparius* exposed to permethrin-spiked sediment in the laboratory. Based on the laboratory sediment toxicity test alone, acute lethal effects would not have been expected in the pond systems. However, effects observed in the field might be due to concentrations of the test substance in the water column immediately after dosing. For organisms such as *C. riparius*, which live in close proximity to both the sediment and the overlying water, acute exposure during pollution events such as spray drifts is likely to be via the overlying water.	

Effects of permethrin on phytoplankton and zooplankton in an enclosure ecosystem in a pond	**Reference: [43]**	**Reliability index: 2**
Study NOEC: <0.75 µg l^{-1} (initial nominal concentration). NOEC based on effects observed on *Chaoborus flavicans* and *Daphnia rosea* after first application.		
Exposure: Static, three enclosures in a shallow, eutrophic pond; two applications of permethrin, the second 18 days after the first one. Target concentrations 0 (control); 0.75 and 1.5 µg l^{-1} at first application; at second application 0, 10 and 1.5 µg l^{-1}	Analysis: Analysis of permethrin in water post-application. Study NOEC refers to initial nominal concentration.	
Description of test site/test facility: Three enclosures (stainless steel frame covered with polyethylene film; 1 m diameter, 3.8 m deep) were placed into a shallow eutrophic pond. The enclosures were driven into the bottom of the pond to isolate the water column and the sediment. No aeration of the enclosures occurred during the experiment. In July, two enclosures were dosed with emulsifiable concentrate (EC) formulation of permethrin (*cis*/*trans*isomeric mixture) to yield initial nominal concentrations of 0.75 and 1.5 µg l^{-1}, respectively. The third enclosure served as control. Residue analysis at day 2 post-application resulted in water column concentrations of 0.04 and 0.28 µg l^{-1}; no permethrin could be detected at day 5. A second treatment with permethrin was applied 18 days after the first one. Target concentrations were 10 µg l^{-1} in the enclosure originally dosed to 0.75 µg l^{-1} and again 1.5 µg l^{-1} in the enclosure dosed to 1.5 µg l^{-1}.	Observations/effects: No effects on photosynthetic activity were observed at permethrin concentrations up to 10 µg l^{-1}. However, *Ceratium hirundinella*, a large dinoflagellate, was affected by permethrin. In the treated enclosures, its density declined from >1,000 to <500 per litre. The pelagic insect larva *Chaoborus flavicans* was the sole zooplankton predator. The first application of permethrin severely affected this organism. Floating dead larvae were collected for one week post-application and summed up to 5,729 and 12,044 m^{-2} in the 0.75 and 1.5 µg l^{-1} treatments, respectively (density in control and pond approx. 12,700 $\pm$ 700 m^{-2}). Thus, at 1.5 µg l^{-1}, the total *Chaoborus* population was almost extinct, whereas at 0.75 µg l^{-1} a large proportion survived treatment. The second treatment seemed to have killed almost all *Chaoborus* larvae and only some early instar larvae appeared in the treated enclosures at the end of the observation period (ca. 10 days post second application). *Daphnia rosea*, a dominant species in the pond, was virtually eliminated upon the first treatment with 1.5 µg l^{-1}, while the 0.75 µg l^{-1} treatment reduced its population, but it recovered within 5 days, possibly because of less pressure from *Chaoborus*. The second application hit the populations more seriously; dosing to a target concentration of 10 µg l^{-1} eliminated the *Daphnia* whereas at 1.5 µg l^{-1} some individuals reappeared at the end of the observation period.	

Evaluation of a field bioassay technique to predict the impact of aerial applications of forestry insecticides on stream invertebrates		**Reference: [36]**	**Reliability index: 2**
Study NOEC: 0.5 µg l^{-1} (initial nominal concentration). NOEC based on stream invertebrate drift. Lowest LC50 observed after pulse-exposure for 1 hour and a 47 hour observation period 2 µg l^{-1}.			
Exposure: Flow-through, continuous flow bioassay apparatus.	Analysis: No analysis of permethrin in water. Study NOEC and LC50s refer to nominal concentrations.		
Description of test site/test facility: Field bioassay experiments were conducted on the east tributary of Icewater Creek, 20 km north of Searchmount, Ontario. The continuous flow bioassay apparatus consisted of 2-m long vinyl troughs, supplied individually with water from a head tank (water diverted from Icewater Creek). Test organisms were collected in Icewater Creek and the Goulais River. Test organisms were black flies (*Simulium* sp.), mayflies (Isonychia), caddisflies (Pycnopsyche), stoneflies (Acroneuria), dragonflies (*Ophiogomphus*) and crayfish (*Orconectes*); 8–20 individuals of each species were placed in each trough immediately after collection and allowed to acclimate for 4 hours prior to treatment.	Observations/effects: Duplicate groups of organisms were exposed to a particular concentration for 1 hour and then observed for mortality for 47 hours after exposure. Each concentration was replicated 2–4 times. Drift was induced at concentrations >0.5 µg l^{-1} permethrin (i.e. $NOEC_{drift}$ 0.5 µg l^{-1}). LC50 observed at the described exposure conditions ranged from 2.0 µg l^{-1} (Acroneuria) to 7.1 µg l^{-1} (*Ophiogomphus*).		
Response of a brook trout (*Salvelinus fontinalis*) population to a reduction in stream benthos following an insecticide treatment Invertebrate drift in a headwater stream treated with permethrin		**Reference: [27, 28]**	**Reliability index: Not in Annex 1**
Study NOEC: <1.5 µg l^{-1} (catastrophic drift of stream invertebrates).			
Exposure: Flow-through experiment in a natural stream.	Analysis: Analysis of permethrin in water. Study NOEC refers to analysed concentration.		
Description of test site/test facility: Icewater Creek is a third order cold-water stream in the Lake Superior watershed approximately 50 km north of Sault Ste, Marie, Ontario. In the upper reaches of this	Observations/effects: Following the insecticide injection, massive increases in the number of drifting invertebrates occurred at each site as the contaminated water reached the sampling stations. Drift increases were most pronounced in the East Branch and		

creek, four discrete study sections were established: Crossover, West Branch, East Branch and West Trib. A fifth section (East Trib) was used as an untreated control area for the measurement of trout growth. Permethrin was injected into the stream at 9:30 on 3 June 1987. The delivery rate was calculated to produce a permethrin concentration of approximately 16 µg l^{-1} immediately below the injection site and to maintain a concentration of about 1 µg l^{-1} for 1 hour in the lower section of the East Branch. Four composite samples were collected from all sites at or near the time of drift sampling. In the East Branch, 1.53 µg l^{-1} was measured at 9:40; the peak concentration of 8.64 µg l^{-1} was reached at 10:00, followed by 0.287 µg l^{-1} at 10:30. At 14:30, the concentration at this monitoring point had declined to 0.036 µg l^{-1}. The second highest concentration profile was found for the upper West Trib (1.39 µg l^{-1} at 10:00, 0.231 µg l^{-1} at 11:00; decrease to 0.041 µg l^{-1} by 13:30). At the other sampling points, the peak concentrations never exceeded 0.175 µg l^{-1}. Treatment effects on most parameters were determined by comparison to temporal controls. The experimental design included 3-year pre-treatment data and 1-year post-treatment. Differences in parameters over the four years were tested statistically; thus, the pre-treatment years functioned as temporal controls. In addition, the section East Trib was used as spatial control for fish growth. A site 300 m upstream from the injection point on East Branch was used as a control site for the benthos study.	West Trib sections, where total invertebrate drift densities increased by 5,600 times the pre-treatment levels. Drift levels declined sharply within several hours of the application, but remained at elevated levels (up to 10-fold) for >15 hours. By 36 hours post-treatment, drift rates had resumed to normal levels. The invertebrate drift consisted mainly of insect larvae belonging to the orders Plecoptera, Ephemeroptera and Trichoptera. Drift rates in the control stream remained near zero during the treatment day and exhibited normal diurnal increases at dusk. The abundance of Ephemeroptera, Plecopera and Trichoptera in benthos samples was significantly reduced in East Branch and West Trib following the insecticide treatment. By 50 days post-treatment, numbers of some taxa were still depressed, but with the exception of Ephemeroptera in artificial substrates, there were no significant differences between the treated and the control sites. Recovery of benthos appeared to be largely accomplished by recruitment from egg hatching and oviposition (samples from 50 days post treatment and later contained substantial numbers of early instar larvae). Although there were no post-treatment changes in the density, population age structure, movement patterns, or condition of the fish, the growth of young brook trout was significantly reduced following permethrin injection. This resulted in significant reductions in the size of post-treatment 0+ and 1+ fish compared with pre-treatment years. This reduced growth is especially critical to the productivity of the trout community because of the high percentage composition of these age classes in the population. Annual production of trout in the treatment year was reduced but was not significantly lower than in pre-treatment years. However, although the growth rates and size of 0+ and 1+ trout in the treated area were significantly reduced, significant reductions in the size of young trout in the control stream during the same period indicated that temperature stress was at least partially, if not entirely, responsible for growth reductions of the treated fish. Significant growth reductions of fish in the untreated control stream strongly imply that the insecticide impact did not produce adverse effects on the trout population in the treated stream over and above natural environmental stresses – in this case high temperatures and low water levels.

Effects of permethrin on aquatic organisms in a freshwater stream in South-Central Alaska	**Reference: [42]**	**Reliability index: 2**
Study NOEC: Significant increase of stream invertebrate drift at concentrations ≤0.05 µg l^{-1} (NOEC drift < 0.05 µg l^{-1}). No mortality observed up to concentration peaks of 0.14 µg l^{-1} for 3 h (NOEC mortality >0.14 µg l^{-1}).		
Exposure: Flow through experiment in a natural stream.	Analysis: Analysis of permethrin in water. Study NOEC refers to analysed concentration.	
Description of test site/test facility: A field test of permethrin was conducted along an approximately 2 m wide and 0.3 m deep stream in Chugach National Forest in South-Central Alaska. Permethrin was applied with a hydraulic sprayer to the bark surface of individual trees to a height of 12 m until the bark was thoroughly wet. Treatment plots were located within 5 m of the stream. The effects of permethrin on water chemistry, population levels of aquatic and terrestrial organisms, and residue levels were monitored at three locations in the stream: an untreated control area 800 m above the treatment area; within the treatment area (an area fronting the stream for 100 m); and 500 m below the treatment area. At the monitoring sites, 'biomonitors' (plexi glass tubes of 7.6 cm diameter and 20.3 cm length covered with 273 mesh net at the ends) with either stream invertebrates (Plecoptera and Ephemeroptera species) or salmonid fish fry (*Salvelinus malma* of either 2 or 5 cm length) were placed into the stream and examined hourly for a 24-hour period immediately after treatment.	Observations/effects: Permethrin residues monitored within the treatment site reached 0.05 ± 0.01 µg l^{-1} 5 hours after treatment, 0.09 ± 0.02 µg l^{-1} 6 hours after treatment, 0.14 ± 0.03 µg l^{-1} 8–11 hours after treatment, and 0.02 ± 0.01 µg l^{-1} 14 hours after treatment. No permethrin (<0.01 µg l^{-1}) was found at the control sites above and below the treatment site. Periphyton was not affected by permethrin and its residues in periphyton were all <20 µg/kg at the three monitoring sites, indicating that permethrin residues at the sites within and below the treatment area were not present at sufficient concentrations for sufficiently long to be adsorbed onto or absorbed into the periphyton. Aquatic invertebrate drift, however, increased significantly (fourfold) 3 hours after permethrin use at the treatment site but, within 9 hours, declined to levels observed before spray application. Dipteran (Chironomidae) larvae, and trichopteran (Limnophilidae) larvae accounted for the increase in drift. Aquatic invertebrates and fish fry caged in the biomonitors did not exhibit increased mortality because of the permethrin treatment. Only one Plecopteran died within the treatment site within 24 hours of treatment and no mortality was observed at sites above or below the treatment area.	

Response of procambarid crayfish populations to permethrin applications in earthen ponds	**Reference: [25]**	**Reliability index: 2**
Study NOEC: NOEC crayfish population density <1 µg l^{-1}. Crayfish population exposed to a single permethrin application of nominally 1 µg l^{-1} was reduced 54% 7 days post-application.		
Exposure: Static, in earthen ponds used for rice cultivation.	Analysis: No analysis of permethrin concentration in water. Effect data refer to initial nominal concentrations.	
Description of test site/test facility: Six earthen ponds of 0.044 ha surface used for rice production according to standard practices contained reproducing populations of the two crayfish species *Procambarus zonangulus* and *Procambarus clarkii*. To ensure adequate population size, *P. clarkii* broodstock adults (1:1 male:female ratio) were stocked into ponds at a rate of 171 kg/ha. Population densities were determined immediately prior to and 7 days after pesticide application. Permethrin was administered to the ponds to yield concentrations between 1 and 3 µg l^{-1}. Nine adjacent ponds which had not been treated with permethrin served as controls.	Observations/effects: Seven days post-application, total crayfish population densities had decreased by 54.4% in the pond nominally treated with 1 µg l^{-1} permethrin, 70.0, 80.4 and 83.1% in the three ponds dosed with 2 µg l^{-1}, 78.6% in the pond that received 2.5 µg l^{-1}, and 79.8% in the pond with the highest dose (3.0 µg l^{-1}). *Procambarus zonangulus* comprised 22–25% of the crayfish >40 mm length in two ponds before permethrin exposure. Permethrin caused 100% mortality among this species. Mortality trends within the *P. clarkii* population indicate that permethrin toxicity to this species can be influenced by size, sex and maturity.	

[1] The reliability index (RI) is assigned according to the Klimisch Criteria, defined in Annex 1. For data relevant for PNEC derivation, Data Quality Assessment Sheets are available in Annex 1 and Data Proformas in Annex 2.

2.6.2 Toxicity to saltwater organisms

Single species test toxicity data for marine organisms are only available for four different taxonomic groups, i.e. algae, crustaceans, fish and molluscs (bivalves). Only one chronic toxicity test could be found with the estuarine fish species *Cyprinodon variegatus*. Based on the acute data crustaceans appear to be by far the most sensitive group (see Table 2.9).

A diagrammatic representation of the available short-term saltwater data for permethrin (cumulative distribution functions) is presented in Figure 2.3. The 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 the permethrin PNECs. The lowest critical saltwater data for permethrin are presented in Table 2.9.

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

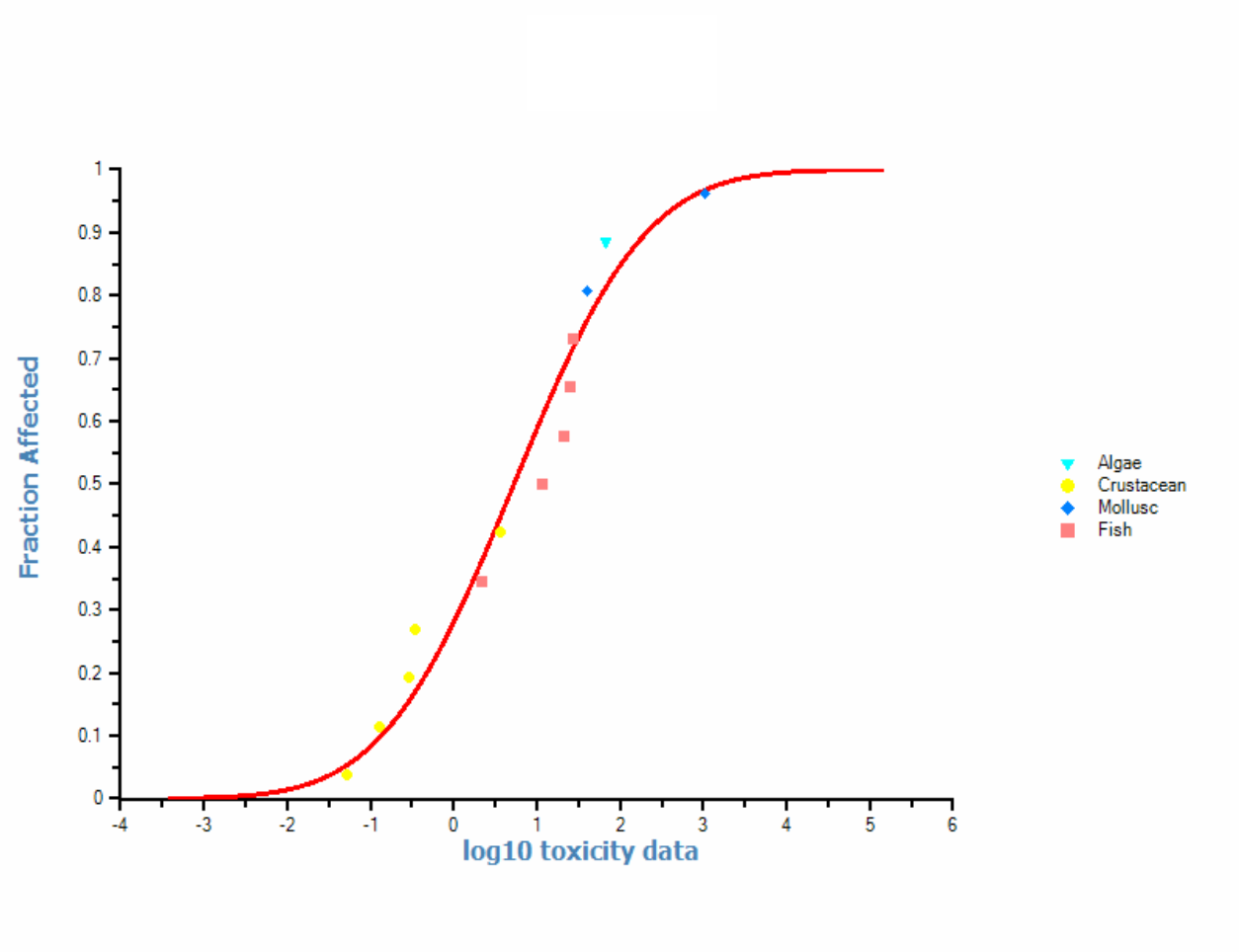


Table 2.9 Short-term and long-term toxicity data for saltwater organisms exposed to permethrin

Scientific name	Common name	Taxonomic group	End-point	Effect	Test duration	Conc. (µg a.i. l^{-1})[1]	Expo-sure[2]	Toxicant analysis[3]	Comments	Reliability index[4]	Refer-ence
Short-term tests											
Skeletonema costatum	Diatom	ALG	EC50	GRO	96 hours	68	s	n		2	[63]
Americamysis bahia	Opossum shrimp	CRU	LC50	MOR	96 hours	0.095	s	m	**GM 0.052 µg l^{-1}**	2	[16]
						0.075	-	-		2	[34]
						0.02	f	m		2	[38]
Crangon septemspinosa	Shrimp	CRU	LC50	MOR	96 hours	0.13	-	-	-	-	[31]
Penaeus aztecus	Brown shrimp	CRU	LC50	MOR	96 hours	0.34	-	-	-	2	[34]
Penaeus duorarum	Pink shrimp	CRU	LC50	MOR	96 hours	0.17	s	m	GM 0.29 µg l^{-1}	2	[16]
						0.22	f	m		2	[38]
						0.35	-	-		2	[34]
						0.51	-	-		2	[34]
Uca pugilator	Fiddler crab	CRU	LC50	MOR	96 hours	2.39	-	-	**GM 3.64 µg l^{-1}**	2	[34]
						2.65					
						7.6					
Crassostrea gigas	Pacific oyster	MOL	EC50	PHY	48 hours	1050	-	-	Embryo-larvae	2	[34]
Crassostrea virginica	American oyster	MOL	EC50	PHY	96 hours	**40.7**	-	-	Spat	2	[34]
Atherinops affinis	Topsmelt	FIS	LC50	MOR	96 hours	25.3	-	-	-	-	[21]
Cyprinodon variegatus	Sheepshead minnow	FIS	LC50	MOR	96 hours	7.8	f	m	**GM 11.5 µg l^{-1}**	2	[38]
						17	s	m		2	[37]
Cyprinodon bovinus	Leon Springs pupfish	FIS	LC50	MOR	96 hours	21	s	m	-	2	[37]
Menidia menidia	Atlantic silverside	FIS	LC50	MOR	96 hours	**2.2**	f	m	-	2	[38]
Menidia beryllina	Inland silverside	FIS	LC50	MOR	96 hours	27.5	-	-	-	-	[21]
Long-term tests											
Cyprinodon variegatus	Sheepshead minnow	FIS	NOEC	MOR (fry)	28 days	**10**	f	m	Early life stage test	2	[44]

[1] The lowest L(E)C50s per taxonomic group are highlighted in bold. If more than one test per species with the same endpoint and test duration was available, geometric means (GMs) of these results were calculated. The GMs are presented in the 'Comments' column.
[2] Exposure: s = static; f = flow-through. [3] Toxicant analysis: m = measured. [4] The reliability index (RI) is assigned according to the Klimisch Criteria, defined in Annex 1. For data relevant for PNEC derivation, Data Quality Assessment Sheets are available in Annex 1 and Data Proformas in Annex 2.
CRU = crustaceans; FIS = fish; MOL = molluscs; PHY = shell deposition; MOR = mortality
LC50 = concentration lethal to 50% of the organisms tested; EC50 = concentration effective against 50% of the organisms tested
NOEC = no observed effect concentration

2.6.3 Toxicity to sediment-dwelling organisms

Only two publications on permethrin toxicity in sediment (mg permethrin/kg sediment basis) could be found and the data from the one acute study [15] and one chronic study [47] (both of which used *Chironomus riparius*) are shown in Table 2.10.

In the chronic study the natural sediment had an organic carbon content of 1.23% [47] whilst the acute study used a natural sediment with an organic carbon content of 9.64% [15].

Table 2.10 Permethrin sediment toxicity data

Scientific name	Taxo-nomic group	Endpoint/ Effect	Test duration (days)	Conc. (mg/kg)	Expo-sure[1]	Toxicant analysis[2]	Comments	Reliability index[3]	Refer-ence
Chironomus riparius	INS	LC50	10	2.11	s	n	Spiked natural sediment	2	[15]
Chironomus riparius	INS	Emergence of adults	>20	0.8 0.4	s	n	Spiked natural sediment, at 0.8 mg/kg, 63% reduction of emergence compared with controls. No significant effect	2	[47]

[1] Exposure: s = static.
[2] Toxicant analysis: n = nominal.
[3] The reliability index (RI) is assigned according to the Klimisch Criteria, defined in Annex 1. For data relevant for PNEC derivation, Data Quality Assessment Sheets are available in Annex 1 and Data Proformas in Annex 2.
INS = insects

In a field study conducted alongside the short-term *C.riparius* lethality test (see Table 2.8, Reference 15) the highest measured concentration in the sediment of the highest pond treatment (100 µg l^{-1}) was 0.22 mg/kg. A sediment permethrin concentration of 0.004 ug/kg resulted in emergence patterns similar to those in the control pond. In the 0.014 ug/kg treatment insects were collected in all sampling dates, but were much reduced relative to the control, especially on days 6, 8, 10 and 15. At the highest sediment exposure concentration of 0.22 mg/kg adult emergence was delayed until day 31. This is an order of magnitude lower than the 10-day LC50 of 2.11 mg/kg for *C. riparius* exposed to permethrin-spiked sediment in the laboratory. Based on the laboratory sediment toxicity test alone, acute lethal effects would not have been expected in the pond systems. However, effects observed in the field at these lower measured sediment concentrations might have been due to concentrations of the test substance in the water column immediately after dosing. For organisms such as *C. riparius*, which live in close proximity to both the sediment and the overlying water, acute exposure during pollution events such as spray drifts is likely to be via the overlying water.

2.6.4 Endocrine-disrupting effects

Various studies have investigated the effects of permethrin on the endocrine function of mammals. However, the results of these studies are often contradictory and no weight-of-

evidence conclusions can currently be drawn on the possible endocrine-disrupting effects of permethrin.

Kunimatsu *et al.* [58] investigated the effects of permethrin on oestrogen-receptor-mediated (uterotrophic assay) and androgen-receptor-mediated (Hershberger assay) mechanisms in rats. No effects were seen in either assay up to the highest dose tested (150 mg/kg per day in the uterotrophic assay and 75 mg/kg per day in the Hershberger assay). However, the use of the same two assays by Kim *et al.* [54] suggested that permethrin had an oestrogen-like effect on female rats and an anti-androgen-like effect on male rats. In 3-day studies, the effects of subcutaneous treatments of permethrin on the uterus of 18-day-old rats resulted in a significant increase in uterine weight and oestradiol (E2)-induced uterine weights at 200 mg/kg and 800 mg/kg, respectively. These concentrations are higher than those used by Kunimatsu *et al.*, who reported that the highest dose used in their tests (150 mg/kg per day) was 'the maximum level that could be used without causing excessive systemic toxicity'. This factor may put into question the results obtained by Kim *et al*. However, in the Hershberger assay, the latter reported significant reductions in androgen-dependent sex tissue weights at all doses tested (10, 50 and 100 mg/kg). According to Kunimatsu *et al.*, these levels (up to 75 mg/kg) are low enough as to not be excessively cytotoxic.

Kakko *et al.* [56] studied the proliferation of the breast cancer cell line, MCF7, after a 7-day exposure to the combined effects of oestradiol (0.10 nM) and permethrin (0.1–100 μM). Proliferation and cell toxicity were studied by measuring the adenosine triphosphate (ATP) content with a luminescence method. In the ATP test, low concentrations (0.1–1 μM) of permethrin in co-exposure with oestradiol caused a significant increase in the proliferation of MCF7 cells. Similar results were found by Go *et al*. [59], where a concentration of 100 μM had a noticeable effect on cell proliferation of MCF7. In contradiction to these result, Kim *et al.* [57] found no dose-dependent cell proliferation in MCF7 BUS cells. However, they did report that permethrin significantly inhibited 17β-oestradiol-induced MCF7 BUS cell proliferation at 10^{-6} M, i.e. an anti-oestrogenic effect.

Eil and Nisula [60] tested permethrin for its ability to interact with androgen-binding sites in dispersed, intact human genital skin fibroblasts and in human plasma to sex hormone-binding globulin (SHBG). Permethrin inhibited fibroblast binding of [^{3}H]methyltrienolone (R1881) at 22°C by 50 per cent at a concentration of 44×10^{-5} M. The authors concluded that the data indicate that permethrin can interact competitively with human androgen receptors and SHBG.

Studies so far have indicated both oestrogenic and anti-oestrogenic effects in mammals, and it is unclear whether there is oestrogen-receptor binding. The assays used so far are undergoing validation by the Organisation for Economic Co-operation and Development (OECD) and the significance of these results for human health effects are at present unclear. Evidence so far suggests that permethrin may potentially have endocrine-disrupting effects.

Only one study could be located that has investigated the endocrine effects of permethrin on a non-mammalian system. Zou and Bonvillain [55] investigated the effects of permethrin on epidermal chitinase activity in the fiddler crab (*Uca pugilator*). A 7-day exposure to 5 μg l^{-1} had no effect on the chitinase activity of the crabs. This was the only

concentration tested and so a NOEC could not be calculated. However, the data is unexpected given that the geometric mean 96-hour LC50 for this species is lower than the exposure concentration.

2.6.5 Mode of action of permethrin and occurrence of relevant metabolites in the aquatic environment

Synthetic pyrethroids are neuropoisons acting on the axons in the peripheral and central nervous systems by interacting with sodium channels in mammals and/or insects. At near-lethal dose levels, synthetic pyrethroids cause transient changes in the nervous system such as axonal swelling and/or breaks and myelin degeneration in sciatic nerves. They are not considered to cause delayed neurotoxicity of the kind induced by some organophosphorus compounds. Electrophysiological recording from dosed cockroaches reveal trains of cercal sensory spikes and, sometimes, spike trains from the cercal motor nerves and the central nervous system.

The signs of poisoning caused by permethrin in mammals are restlessness, incoordination, hyperactivity, prostration, and paralysis [2]. Poisoning closely resembles that produced by DDT and involves a progressive development of fine whole-body tremor, exaggerated startle response, uncoordinated twitching of the dorsal muscles, hyperexcitability and death. The tremor is associated with a large increase in metabolic rate and leads to hyperthermia which, with metabolic exhaustion, is the usual cause of death. Respiration and blood pressure are well-sustained but plasma noradrenaline, lactate and, to a lesser extent, adrenaline are greatly increased.

Permethrin administered to mammals is metabolised rapidly and almost completely eliminated from the body within a short period of time. Major routes of metabolism for both *trans*- and *cis*-isomers are ester cleavage and oxidation of the 4'-position of the terminal aromatic ring. A less important reaction in mammals is hydroxylation of the geminal dimethyl group of the cyclopropane ring. Major metabolites thus formed are:

- Cl_2CA in free and glucuronide form;
- the sulfate conjugate of 4'-hydroxy-3-phenoxybenzoic acid (Pbacid) in free and conjugate form;
- hydroxymethyl-Cl_2CA as a glucuronide conjugate.

2.6.6 Mesocosm and field studies

Freshwater mesocosm and field studies

The six available field tests (for description of study details see Table 2.8) support the suggested PNEC of 0.01 µg l^{-1} for transient concentration peaks.

In five of the six studies, the observations of effects are based on permethrin initial concentrations (i.e. a transient concentration peak since permethrin, due to its physico-chemical properties, is removed rapidly from the water column). Severe effects on insect and crustacean populations with reductions of >40 and >50 per cent are reported at concentrations of 0.75 [43] and 1 µg l^{-1} [25], respectively. Significantly increased drift of stream invertebrates is reported at concentrations <0.05 µg l^{-1}. Whatever the long-term

ecological consequences of such an increase in drift rate, the event shows that effects in complex natural systems may be observed at very low permethrin concentrations, which are close (with a factor <5) to the PNEC based on single species tests [20, 29, 41].

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
As would be expected from the mode of action of this insecticide, crustaceans and insects are the most sensitive species among the taxonomic groups for which long-term single species toxicity tests are available (algae, crustaceans, fish, insects and molluscs). Hence, a base set of toxicity data (i.e. tests with algae, crustaceans and fish) is available and the assessment factor method can be applied.

In the original review (carried out in 2004) the lowest available data point for algae was a 72-hour EC10 of 5,100 µg l^{-1} in *Chlamydomonas reinhardii* [18].[6] The corresponding EC0 was approximately 4,700 µg l^{-1}. These data were based on nominal concentrations of permethrin in a static exposure system. Consequently, they are not suitable for PNEC derivation. No other long-term freshwater data for algae could be located. As a result there was uncertainty over the potential toxicity of permethrin to algae. Although, the available data and the mode of action indicate that algae should not be the most sensitive taxa to permethrin it was considered important to reduce the uncertainty in the dataset (and increase confidence in the assigned assessment factor) by generating valid algal data. Therefore, the Environment Agency commissioned a study of the effects of pemethrin on *Pseudokirchneriella subcapitata* [61]. The 72-hour study was carried out to OECD Guideline 201 and involved analytical confirmation of the exposure concentrations. The study reported 72-hour NOEC values of 160 µg l^{-1} for effects on growth as measured using both growth rate and biomass endpoints.

The lowest high quality long-term NOEC available for arthropods is for the stonefly species *Pteronarcys dorsata* (28-day NOEC 0.029 µg l^{-1}) [12]. This value is based on measured data in a flow-through exposure system. However, the same paper reported more than 55 per cent mortality of the caddisfly *Brachycentrus americanus* after a 28-day exposure to the lowest tested exposure concentration of 0.03 µg l^{-1} permethrin, but no NOEC value could be determined. These are the lowest good quality long-term single species test values available.

[6] This value was taken from a graph within the published document.

Other arthropods are of lower sensitivity with long-term NOECs of 1 µg l^{-1} for *Daphnia magna* and <1 µg l^{-1} for *Daphnia pulex* [39]. The USEPA OPP database reports a 21-day NOEC of 0.039 µg l^{-1} based on a.i. which has been extrapolated from the Brixham study data [39]. However, all these values were derived using microencapsulated permethrin, and are not appropriate for the generation of the PNEC.

The lowest good quality NOEC reported for fish was a value of 0.66 µg l^{-1} in fathead minnow after a 32-day exposure [40]. This value was generated in a flow-through system with measured exposure concentrations.

The TGD [45] proposes the derivation of the PNEC from either the lowest NOEC with an assessment factor of 10 (provided three NOECs from different trophic levels are available) or, if acute effect data are available that are lower than the lowest long-term NOEC, based on the lowest E(L)C50 with an assessment factor of 100. Based on the available data, the lowest good quality long-term NOEC is a value of 0.029 µg l^{-1} for the stonefly *Pteronarcys dorsata*.In the study with *P. dorsata*, however, a very steep concentration response was observed with no effect at 0.029 µg l^{-1}, but 100 per cent immobilisation at 0.042 µg l^{-1} after 28 days [12]. , In the same study, the caddisfly *Brachycentrus americanus* suffered 55 per cent mortality at 0.03 µg l^{-1} (the lowest concentration tested) and no NOEC value could be determined. Since the effects level in the *B.americanus* is greater than 20% the TGD approach cannot be used to derive a NOEC from the LOEC. . Therefore, the data for *B.americanus* is used in a supporting role.

The lowest reliable NOEC is the value of 0.029 µg l^{-1} for the stonefly *Pteronarcys dorsata*. As good quality long-term NOECs are available for a range of taxa (crustaceans, insects and fish) and, given the mode of action of permethrin, the most sensitive organisms are represented, an assessment factor of 10 could be used to derive the PNEC:

$PNEC_{freshwater\_lt}$ = (0.03 µg l^{-1} permethrin)/AF (10) = 0.003 µg l^{-1} permethrin

The TGD also proposes the derivation of the PNEC from acute data with an AF of 100 if acute effect data are available that are lower than the lowest long-term NOEC. The short-term database contains two 50 per cent effect concentrations at low concentrations of permethrin (*Oncorhynchus mykiss* LC50 of 0.014 µg l^{-1} and a *Daphnia magna* 96-hour LC50 of 0.039 µg l^{-1}).

Both values are likely to be outliers as discussed in the section below but, if the process were followed through, using the lowest reliable E(L)C50 (*Hexagenia bilineata* 96-hour LC50 of 0.1 µg l^{-1}; see below) and applying an AF of 100 would generate a **PNEC** of 0.001 µg l^{-1} permethrin. This value is lower than the PNEC derived using long-term data. These PNEC values are supported by the data from the freshwater mesocosm studies described in Section 2.6.6 which show that effects in complex natural systems may be observed at very low permethrin concentrations, which are close (with a factor <5) to the PNEC based on single species tests.

Based on the review of the available data it is proposed that the PNEC of 0.001 µg l^{-1} derived using short-term data is applied as the long-term value. This value provides a

margin of safety with respect to the significant effects of permethrin on the survival of the caddisfly *Brachycentrus americanus* at 0.03 µg l^{-1}.

PNEC accounting for transient concentration peaks
Short-term toxicity data are available for six different taxonomic groups, i.e. algae, crustaceans, fish, amphibians, molluscs and insects. Crustaceans and insects are the most sensitive organisms, followed by salmonid fish.

Algae are less sensitive to permethrin than crustaceans or fish. The lowest available EC50 for algae was a value of 68 µg l^{-1} for diatoms [5]. In addition, the UK EQS document [53] reports 50 per cent effect concentrations for algae exposed to permethrin all above 1,000 µg l^{-1}, confirming the comparatively low sensitivity of these organisms. However, full details of these studies were not available and it has not been possible to assess the quality of the data. As a result in the original review in 2004, there was uncertainty over the potential toxicity of permethrin to algae. Although, the available data and the mode of action indicate that algae should not be the most sensitive taxa to permethrin it was considered important to reduce the uncertainty in the dataset (and increase confidence in the assigned assessment factor) by generating valid algal data. Therefore, the Environment Agency commissioned a study of the long-term effects of permethrin on *Pseudokirchneriella subcapitata* [61] and this also provides a relevant EC50 value. The 72-hour study was carried out to OECD Guideline 201 and involved analytical confirmation of the exposure concentrations. The study reported 72-hour EC50 value of >160 µg l^{-1} for effects on growth as measured using both growth rate and biomass endpoints.

The lowest available crustacean value was 96-hour LC50 of 0.039 µg l^{-1} in *Daphnia magna* [34]. There were only limited data with which to assess this study, but it was found acceptable by the US EPA Office of Pesticide Programs for registration of permethrin and so has been classified as reliable with restriction in this report. However, the study was conducted over 96 hours, twice as long as in the standard 48-hour procedure. Comparison of this value with those generated in *Daphnia* tests adhering to the standard 48-hour exposure scheme (48-hour LC50 range of 0.112–7.2 µg l^{-1} with a geometric mean (GM) of 0.63 µg l^{-1}) [see Table 2.7] indicate that this value may be an outlier. The 96-hour *Daphnia magna* LC50 has, therefore, been used only as supporting data. Indeed the GM for *D. magna* would be greater than the *H. bilineata* LC50 even if the value of 0.039 were included.

The lowest good quality acute value for arthropods is a 96-hour LC50 of 0.1 µg l^{-1} for the insect species *Hexagenia bilineata* [34]. This value was found acceptable by the US EPA Office of Pesticide Programs for registration of permethrin and so was classified as reliable with restriction in this report. This value is supported by several tests on crustacean and insect species with short-term effect concentrations within the range 0.3–0.6 µg l^{-1} (Table 2.7).

The lowest available fish value was a 144-hour LC50 of 0.014 µg l^{-1} in rainbow trout *Oncorhynchus mykiss* [9]. However, this value is likely to be an outlier as it is more than a factor of 500 lower than the geometric mean (5.88 µg l^{-1}) of six acute tests with *O. mykiss*. The result is given as µg l^{-1} but may be µmol l^{-1}, which is equivalent to 5.5

µg l^{-1}; which would then agree with other *O. mykiss* acute mortality data. In addition, there were few details available with which to assess the quality of this study.

The lowest good quality data for fish indicate lower sensitivity than insects with 96-hour LC50 values of 1–2 µg l^{-1} reported in various salmonid species (Atlantic salmon [34] Lahontan cut-throat trout, greenback cutthroat trout and Apache trout [37]).

It is recommended that the short-term PNEC is derived on the basis of the *Hexagenia bilineata* 96-hour LC50 of 0.1 µg l^{-1} and guidance given in the TGD on effects assessment for intermittent releases (Section 3.3.2 of Part II of the TGD [45]). As permethrin is a neurotoxin with a specific mode of action and insects belong to the most sensitive organisms, it is also recommended that a reduced assessment factor of 10 (instead of 100) is used to extrapolate from the 50 per cent acute effect level to the short-term no effect level.

$PNEC_{freshwater\_st}$ = 0.1 µg l^{-1}permethrin/AF (10) = 0.01 µg l^{-1} permethrin

This value is lower than the lowest available crustacean value (96-hour LC50 of 0.039 µg l^{-1} in *Daphnia magna*) which is only considered appropriate for use in a supporting role.

Data on mesocosm and field studies have shown that effects in complex natural systems may be observed at very low permethrin concentrations, which are close (within a factor <5) to the PNEC based on single species tests [20, 29, 41](see Section 2.6.6).

3.1.2 PNECs for saltwaters

The effects dataset for marine species is very small, with chronic data for fish and acute toxicity tests for algae, crustaceans, fish and molluscs (Table 2.9). The toxicity data for marine taxa do not differ obviously from the range of values obtained for similar freshwater taxa (see Tables 2.6 and 2.7). However, the marine database is too small to draw firm conclusions about any possible differences.

As there are no obvious differences in the sensitivity of freshwater or saltwater species from the same taxonomic groups, the freshwater and saltwater data were combined for derivation of the PNECs for marine water bodies.

PNEC accounting for the annual average concentration
Only one long-term saltwater data point could be located. A 28-day NOEC of 10 µg l^{-1} was reported for sheepshead minnow [44]. This was a well-documented study with measured exposure concentrations and is suitable for PNEC derivation.

Due to the lack of additional saltwater data, the freshwater and saltwater datasets have been combined. The lowest long-term data available in the combined saltwater and freshwater database are those used for derivation of the annual average $PNEC_{freshwater}$.

Therefore, the long-term PNEC for saltwater was derived on the same basis as the freshwater PNEC, i.e. using the using the lowest reliable E(L)C50 (*Hexagenia bilineata* 96-hour LC50 of 0.1 µg l^{-1} and applying an AF of 100 to generate a PNEC of 0.001 µg l^{-1} permethrin . The TGD suggests a total assessment factor of 1000 if three long-term tests

are available for three taxonomic groups, with a factor of 10 applied to account for the absence of data for marine species. However, short-term tests with additional marine species are available and a reduced assessment factor of 500 is recommended. These acute marine data indicate that molluscs belong to the least sensitive groups and would be protected by the proposed $PNEC_{saltwater\_lt}$ of 0.0002 µg l^{-1}.

$PNEC_{saltwater\_lt}$ = (0.1 µg l^{-1} permethrin)/AF (500) = 0.0002 µg l^{-1} permethrin

PNEC accounting for transient concentration peaks
Acute toxicity data are available for four different marine taxonomic groups (algae, crustaceans, fish and molluscs), with crustaceans appearing to be the most sensitive group.

The lowest data for marine algae is a 96-hour EC50 of 68 µg l^{-1} for the diatom *Skeletonema costatum*, in a well described study without the measurement of exposure concentrations [63].

The lowest acute crustacean value is the geometric mean 96-hour LC50 of 0.052 µg l^{-1} for the shrimp *Americamysis bahia* [16, 34, 38]. This value was calculated from LC50 values from a number of good quality studies with measured exposure concentrations reported in the majority of cases.

Saltwater fish appear to be of lower sensitivity than crustaceans. The lowest reliable data point was a 96-hour LC50 in the Atlantic silverside (*Menidia menidia*) of 2.2 µg l^{-1} [38]. All other fish species were of lower sensitivity.

Saltwater data were also available for molluscs and indicate comparatively low sensitivity in these organisms. The lowest reported effect concentration was a 96-hour EC50 for shell deposition of 40.7 µg l^{-1} in the American oyster [34]. This value was found acceptable by the US EPA Office of Pesticide Programs for registration of permethrin and so was classified as reliable with restriction in this report.

The TGD does not provide specific guidance for the assessment of acute effects of intermittent releases to marine water bodies. Therefore, it is recommended that the short-term PNEC is derived on the basis of general guidance given in the TGD on effects assessment for intermittent releases (Section 3.3.2 of Part II of the TGD [45]). As permethrin acts specifically on the nervous system and crustaceans belong to the most sensitive organisms, it is also recommended that only a reduced assessment factor of 50 (instead of 100) be used with the *Americamysis bahia* 96-hour LC50 of 0.052 µg l^{-1} to extrapolate from the 50 per cent effect level to the short-term no effect level.

$PNEC_{saltwater\_st}$ = 0.052 µg l^{-1}permethrin/AF (50) = 0.001 µg l^{-1} permethrin

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

The minimum number of long-term toxicity studies (at least 10 NOECs from eight taxonomic groups) is not available. Therefore, the SSD approach cannot be used for PNEC derivations.

3.3 Derivation of existing EQSs

The 1988 report [53] proposing UK EQSs for mothproofing agents took into account both laboratory and field data to derive a standard for total permethrin. From the data, it was unlikely that levels of total permethrin below 0.01 µg l^{-1} would adversely affect either aquatic invertebrate populations or dependent fisheries. For the protection of aquatic life, therefore, an EQS of 0.01 µg l^{-1} total permethrin expressed as a 95th percentile was proposed.

The database for marine invertebrates was smaller than that for freshwater life and the reported toxicity data was generally comparable. Therefore, the EQS for the protection of saltwater life was 'read across' from the freshwater standard, i.e. 0.01 µg l^{-1} total permethrin expressed as a 95th percentile.

3.4 Derivation of PNECs for sediment

3.4.1 PNEC derivation by the TGD deterministic approach

Because the log Kow of permethrin is >3, the derivation of PNECs for the protection of benthic organisms is required.

Only two experimental toxicity tests on the effects of permethrin concentrations in sediment are available (Table 2.10). The 10-day LC50 of 2.11 mg permethrin/kg sediment [15] (in a sediment with an organic carbon content of 9.64%) may be used as a short-term acute value for PNEC derivation. According to the TGD, the appropriate assessment factors are 1,000 for freshwater and 10,000 for saltwater.

- $PNEC_{sediment\_freshwater}$ = 2,110 µg permethrin/kg dw/AF (1,000) = 2.1 µg permethrin/kg dw

- $PNEC_{sediment\_saltwater}$ = 2,110 µg permethrin/kg dw/AF (10,000) = 0.21 µg permethrin/kg dw

The chronic (emergence) NOEC of 0.4 mg/kg in natural sediment [47] (in a sediment with an organic carbon content of 1.23%) can be used as long-term chronic value for PNEC derivation. According to the TGD, the appropriate assessment factors are 100 for freshwater and 1,000 for saltwater.

- $PNEC_{sediment\_freshwater}$ = 400 µg permethrin/kg dw/AF (100) = 4.0 µg permethrin/kg dw

- $PNEC_{sediment\_saltwater}$ = 400 µg permethrin/kg dw/AF (1,000) = 0.4 µg permethrin/kg dw

In a field study conducted alongside the short-term *C.riparius* lethality test (see Table 2.8, Reference 15) no effects on chironomid emergence were evident at a sediment permethrin concentration of 0.004 ug/kg. At the next concentration of the 0.014 ug/kg the numbers of chironomids emerging were reduced whilst at the highest measured concentration of 0.22 mg/kg adult emergence was delayed until day 31.This is an order of magnitude lower than the 10-day LC50 of 2.11 mg/kg for *C. riparius* exposed to permethrin-spiked sediment in the laboratory. Based on the laboratory sediment toxicity test alone, acute lethal effects would not have been expected in the pond systems. However, effects observed in the field might at these lower measured sediment concentrations have been due to concentrations of the test substance in the water column immediately after dosing. For organisms such as *C. riparius*, which live in close proximity to both the sediment and the overlying water, acute exposure during pollution events such as spray drifts is likely to be via the overlying water.

It is proposed that the sediment PNEC is based on the chronic data because:

1. The preferred approach in the TGD is the use of chronic data and smaller factor over acute data and larger factor;
2. The OC content in the acute study (9.64%) is well outside the range of "preferred" OC (5% in "standard" sediment, 2%-5% recommended in the TGD, 2% ±0.5% in the draft OECD guideline for *Chironomus* test).
3. The sediment OC content of 1.23% in the chronic study is also outside the "preferred" ranges, but is only just outside the OECD draft guideline content of 2% ±0.5%.

And therefore the following PNECs are proposed:

$PNEC_{sediment\_freshwater}$ = 400 µg permethrin/kg dw/AF (100) = 4.0 µg permethrin/kg dw

$PNEC_{sediment\_saltwater}$ = 400 µg permethrin/kg dw/AF (1,000) = 0.4 µg permethrin/kg dw

3.4.2 PNEC derivation by the TGD probabilistic approach

As no suitable experimental sediment toxicity data are available, the SSD approach cannot be used for $PNEC_{sediment}$ derivation.

3.5 Derivation of PNECs for secondary poisoning of predators

3.5.1 Mammalian and avian toxicity data

The Joint FAO/WHO Meeting on Pesticide Residues (JMPR) has discussed and evaluated permethrin several times. In 1985, an acceptable daily intake (ADI) of 0–0.05 mg/kg body weight (bw) was established (for permethrin with *cis*/*trans* isomer ratios of 40:60 and 27:75) [2]. The Reference Dose for Chronic Oral Exposure (RfD) established by the US EPA is 0.05 mg/kg per day [8].

Permethrin has a low acute toxicity to mammals such as rats, mice, rabbits and guinea pigs. None of the metabolites of permethrin shows a higher acute (oral or intraperitoneal) toxicity than permethrin itself [2]. Permethrin administered to mammals is rapidly metabolised and almost completely excreted in urine and faeces within 12 days. The *trans*-isomer is eliminated faster than the *cis*-isomer (the former is much more susceptible to esterase attack than the latter).

The major metabolic reactions are ester cleavage and oxidation, particularly at the terminal aromatic ring of the phenoxybenzyl moiety and the geminal dimethyl group of the cyclopropane ring, followed by conjugation. Acute oral LD50s for mammals (e.g. rat, mouse, guinea pig, rabbit) are approximately 4000 mg/kg if the *cis*/*trans* isomer ratio is 40:60 as in the commercial product [5].

With long-term repeated dose oral exposure, an increase in liver weight was found in mice and rats [2]; this was considered to be associated with an induction of the liver microsomal enzyme system. The critical no observed effect level (NOEL) is 100 mg/kg diet, corresponding to 5.0 mg/kg bw, obtained in a 2-year rat study (see Table 3.1). In feeding experiments with dogs, the 90-day no-effect level was 200 parts per million (ppm).

With regard to carcinogenic and mutagenic effects of permethrin as well as adverse effects on reproduction, toxicological evidence from mutagenicity studies and from long-term mouse and rat studies suggests that permethrin's oncogenic potential is very low, limited to female mice, and probably nongenotoxic. Permethrin was not mutagenic in *in vivo* or *in vitro* studies. It is not teratogenic to rats, mice or rabbits at dose levels up to 225, 150, and 1,800 mg/kg bw, respectively. In a three-generation reproduction study with rats, permethrin did not induce adverse effects at levels up to 2,500 mg/kg diet (≈180 mg/kg bw diet).

Permethrin has low toxicity to birds when given orally or fed in the diet. The LD50 is ≥3,000 mg/kg bw for acute single oral dosage and ≥5,000 mg/kg diet for dietary exposure [2]. In hens, permethrin had no effect on reproduction at dose levels up to 40 mg/kg diet (unbounded NOEC, Table 3.1).

Table 3.1 Mammalian and avian oral toxicity data relevant for the assessment of secondary poisoning

Study and result	Details
Long-term toxicity to mammals	
Rat **NOEL 5 mg/kg bw/day** **≈ 100 mg/kg diet** **LOEL 25 mg/kg bw/day** **≈ 500 mg/kg diet** Unpublished reports submitted to WHO [2] by FMC Corporation, Environmental Pathology Services (Bio-Dynamics Inc. Project): Braun W G and Rinehart W E, 1977 *A twenty-four month oral toxicity/carcinogenicity study of FMC33297 in rats*. Billups L H, 1978a *Histopathologic examination of a twenty-four month toxicity/carcinogenicity study of compound FMC33297 in rats*. Billups L H, 1978b *Twenty-four month toxicity/carcinogenicity study of compound FMC33297 in rats*.	Long-Evans rats (60 males and 60 females per group) fed permethrin in the diet at dose levels of 0, 20, 100 or 500 mg/kg for 2 years did not show any mortality or adverse effects on growth, food consumption or behaviour attributable to the administration. Haematology, clinical chemistry and urinalysis measurements were performed at either 6 months or 1 year, and at the end of the study. There were no compound-related effects on a wide variety of parameters examined, and ophthalmological examination indicated no abnormalities. Blood glucose levels were higher in the highest-dose males at 24 months and in the highest-dose females at 18 months compared with the values of control animals. Two independent evaluations of the histopathological data concluded that there was no oncogenic potential for permethrin. The NOEL for general toxicity in this study was estimated to be 100 mg/kg.
Effects on reproduction of mammals	
Rat **NOAEL 180 mg/kg bw/day** Unpublished data submitted to WHO [2]: James J A, 1979 *A multigeneration reproduction study of 21Z73 (permethrin) in the rat.* Report No. BPAT 79-3. Beckenham, Kent: Wellcome Research Laboratories.	In a three-generation reproduction study, groups of 20 male and 20 female Wistar COBS rats received permethrin (25:75) in the diet at 0, 5, 30 and 180 mg/kg bw/day during growth, mating, gestation, parturition and lactation for three generations, each with two litters. Foetal toxicity and teratogenicity were assessed in the second pregnancy of the F2 generation. Treatment with permethrin had no effect on general behaviour or condition, food intake, body weight gain, or pregnancy rate of the dams, or on parturition, sex ratio, or pup weight. Examination of F3b foetuses showed no treatment-related effect on sex ratio, body weight, or the occurrence of visceral or skeletal abnormalities. This study indicated that permethrin (25:75) has no effect on the reproduction of rats at doses up to 180 mg/kg bw/day.

Effects on reproduction of birds	
Hen **NOAEL 40 mg/kg diet** (apparently unbounded NOEC, not suitable for PNEC derivation) Unpublished data submitted to WHO [2] by ICI Ltd: Ross D B, Prentice D E, Majeed S K, Gibson W A, Cameron D M, Cameron M M C D and Roberts N L, 1977 *The incorporation of permethrin in the diet of laying hens (part I)*. Report No. ICI 152/77387. Huntingdon: Huntingdon Research Centre.	The inclusion of permethrin at up to 40 mg/kg in the diet of laying hens for 28 days had no adverse effects on the health of parent birds or on egg production quality, hatchability or the viability of the chicks produced.

LOEL = lowest observed effect level
NOEL = no observed effect level
NOAEL = no observed adverse effect level

3.5.2 PNECs for secondary poisoning of predators

The BCF data for permethrin are 4–570 for insects, 55–750 for fish and 1,900 for the oyster species *Crassostrea virginica* (see Section 2.5). Hence, the trigger of BCF >100 is met and the derivation of PNECs for secondary poisoning (secpois) of predators is required.

The two lowest reported oral NOELs are 40 and 100 mg/kg diet for hens and rats, respectively (Table 3.1). The NOEL for hens is unbounded (i.e. the highest concentration tested) and, therefore, not suitable for the assessment of secondary poisoning. The rat NOEL, however, refers to a 2-year chronic study and is relevant for PNEC derivation.

The appropriate assessment factor to derive a PNEC based on a chronic $NOEC_{food}$ of a mammalian study is 30 (Table 23 of the TGD [45]).

$PNEC_{secpois\_biota}$ = $NOEC_{food}$ (100 mg/kg)/AF 30 = 3.33 mg/kg prey (wet weight)

Reported BCF values for insects, fish and molluscs range up to 570, 750 and 1,900, respectively. Information on biomagnification of permethrin is not available but, due to its rapid metabolism and elimination from the body within a short period of time, the occurrence of biomagnification is considered unlikely (see Section 2.5). Biomagnification is, therefore, not considered in the following calculations.

The corresponding safe concentration in water (preventing bioaccumulation in prey to levels >$PNEC_{secpois\_biota}$) is calculated as follows:

$PNEC_{secpois\_water}$ = $PNEC_{secpois\_biota}$/BCF

If the highest reported BCF of 1900 is used for the calculation, this results in a (lowest) corresponding water concentration of:

$PNEC_{secpois\_water}$ = 3.33/1,900 = 1.75 µg l^{-1} permethrin

This concentration is much higher than the proposed long-term PNECs for the protection of the pelagic communities in both inland and marine water bodies. Therefore, if quality standards are set on the basis of these PNECs, the protection of predators from secondary poisoning is included and the derivation of additional quality standards with particular reference to secondary poisoning is not considered necessary.

4. Analysis and monitoring

The most common methods of analysis for permethrin are:

- gas chromatography combined with mass spectrometry (GC-MS) and detection by electron capture detection (ECD), flame ionisation detection (FID) or flame photometric detection (FPD);
- high performance liquid chromatography (HPLC) coupled with ultraviolet (UV) detection.

Thermal conductivity detection, thermionic detection and nitrogen phosphorus detection (NPD) have also been used in conjunction with GC.

To extract permethrin from its matrix, samples are generally homogenised with an appropriate solvent (hexane, benzene or a binary solvent mixture, such as hexane/acetone, hexane/isopropanol or light petroleum/diethyl ether). This co-extracts a wide variety of other lipophilic substances as well as permethrin, which means that further liquid–liquid or column chromatography partitioning may be required to remove potential interferents.

Soil samples (mechanically ground) are extracted with acetone/hexane, methanol, acetone or acetonitrile.

Permethrin is extracted from water samples with hexane, dichloromethane (methylene chloride) or acetonitrile with subsequent drying with anhydrous sodium sulphate.

Chen and Wang [48] offer an extensive review of the chromatographic methods employed for the determination of permethrin and other pyrethrins and pyrethroids in foods, crops and environmental media. These compounds possess one or more halogenated atoms, which are sensitive to ECD; hence, GC-ECD is a popular method for determining permethrin at environmental concentrations. For more selective determination of permethrin, GC-MS may be used. For screening purposes, HPLC coupled with an UV detector may be used.

For all extraction methods, recovery of permethrin from the matrix is generally high and sensitivity is in the low μg l^{-1} range [49].

Its simplicity and rapid throughput of samples has made the solid-phase extraction (SPE) method described by Junting and Chichang [51] an increasingly popular method for the isolation and analysis of synthetic pyrethroids. A similar method that employs HPLC for analysis was used to quantify pyrethrins in plasma by Wintersteiger *et al.* [52]. This method eliminates time-consuming repeated extractions with organic solvents and centrifugations without losing the efficiency of recovery.

Proposed quality standards and PNECs derived for permethrin range from 0.3 ng l^{-1} to 0.01 µg l^{-1} for waters and 0.4–4.0 µg/kg for sediments. To provide adequate precision and accuracy, the data quality requirements are that, at a third of the EQS, total error of measurement should not exceed 50 per cent. From the literature, it can be seen that analytical methodologies are only capable of achieving detection limits in the low µg l^{-1} order in most media. This suggests that current analytical methods would not be adequate to analyse permethrin for compliance purposes.

5. Conclusions

5.1 Availability of data

Acute toxicity data are available for six different freshwater taxonomic groups (algae, crustaceans, fish, amphibians, insects and molluscs); chronic data are available for algae, crustaceans, fish, insects and molluscs. Laboratory data are supplemented by pond and stream mesocosm studies.

By comparison, the toxicity data available for marine organisms represent just four taxonomic groups (algae, crustaceans, fish and molluscs), with only one chronic test found for fish.

Two publications on permethrin toxicity in sediment were found.

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

As expected from the mode of action of permethrin, crustaceans and insects appear to be the most sensitive taxonomic groups.

Based on the available data, the lowest good quality long-term NOEC is a value of 0.029 µg l^{-1} for the stonefly *Pteronarcys dorsata*. In the study with *P. dorsata*, however, a very steep concentration response was observed with no effect at 0.029 µg l^{-1}, but 100 per cent immobilisation at 0.042 µg l^{-1} after 28 days [12]. However, In the same study, the caddisfly *Brachycentrus americanus* suffered 55 per cent mortality in the same study at 0.03 µg l^{-1} (the lowest concentration tested) and no NOEC value could be determined. Since the effects level is greater than 20% the TGD approach cannot be used to derive a NOEC from the LOEC. Therefore, it is proposed that the data for *B.americanus* is used in a supporting role.

The lowest reliable NOEC is the value of 0.029 µg l^{-1} for the stonefly *Pteronarcys dorsata*. As good quality long-term NOECs are available for a range of taxa (crustaceans, insects and fish) and, given the mode of action of permethrin, the most sensitive organisms are represented, an assessment factor of 10 could be used to derive the PNEC:

$PNEC_{freshwater\_lt}$ = (0.03 µg l^{-1} permethrin)/AF (10) = 0.003 µg l^{-1} permethrin

The TGD also proposes the derivation of the PNEC from acute data with an AF of 100 if acute effect data are available that are lower than the lowest long-term NOEC. The short-term database contains two 50 per cent effect concentrations at low concentrations

of permethrin (*Oncorhynchus mykiss* LC50 of 0.014 µg l^{-1} and a *Daphnia magna* 96-hour LC50 of 0.039 µg l^{-1}).

Both values are likely to be outliers but, if the process were followed through, using the lowest reliable E(L)C50 (*Hexagenia bilineata* 96-hour LC50 of 0.1 µg l^{-1}) and applying an AF of 100 would generate a PNEC of 0.001 µg l^{-1} permethrin. These PNEC values are supported by the data from the freshwater mesocosm studies described in Section 2.6.6 which show that effects in complex natural systems may be observed at very low permethrin concentrations, which are close (with a factor <5) to the PNEC based on single species tests.

Based on the review of the available data it is proposed that the PNEC of 0.001 µg l^{-1} derived using short-term data is applied as the long-term value. This value provides a margin of safety with respect to the significant effects of permethrin on the survival of the caddisfly *Brachycentrus americanus* at 0.03 µg l^{-1}.

This is 10 times lower than the existing EQS of 0.01 µg l^{-1} total permethrin expressed as a 95th percentile. This was based on field and laboratory data that suggested levels <0.01 µg l^{-1} would be unlikely to affect aquatic invertebrates or dependent fisheries.

5.2.2 Short-term PNEC for freshwaters

The acute data show crustaceans and insects, followed by salmonid fish, to be the most sensitive taxonomic groups.

It is recommended that the short-term PNEC is derived on the basis of a 96-hour LC50 of 0.1 µg l^{-1} for the mayfly *Hexagenia bilineata* and guidance given in the TGD on effects assessment for intermittent releases. Given that permethrin is a neurotoxin with a specific mode of action and that insects belong to the most sensitive organisms, a reduced assessment factor of 10 (instead of 100) is recommended in order to extrapolate from the 50 per cent acute effect level to the short-term no-effect level. This results in a $PNEC_{freshwater\_st}$ of 0.01 µg l^{-1}.

The available field studies support this suggested value. There is no existing short-term EQS for permethrin.

5.2.3 Long-term PNEC for saltwaters

The data suggest that there are no obvious differences between freshwater and saltwater species from the same taxonomic groups. Because of this and the lack of marine data, the freshwater and saltwater datasets were combined.

Therefore, the long-term PNEC for saltwater was derived on the same basis as the freshwater PNEC i.e. using the using the lowest reliable E(L)C50 (*Hexagenia bilineata* 96-hour LC50 of 0.1 µg l-1 and applying an AF of 100 to generate a PNEC of 0.001 µg l^{-1} permethrin .. The TGD suggests a total assessment factor of 1000 if three long-term tests are available for three taxonomic groups, with a factor of 10 applied to account for the absence of data for marine species,. However, short-term tests with additional marine species are available and a reduced assessment factor of 500 is recommended. These acute marine data indicate that molluscs belong to the least sensitive groups and would be protected by the proposed $PNEC_{saltwater\_lt}$ of 0.0002 µg l^{-1}.

This proposed PNEC is considerably lower than the existing EQS of 0.01 µg l^{-1}, which was 'read across' from the long-term freshwater EQS.

5.2.4 Short-term PNEC for saltwaters

Crustaceans appear to be the most sensitive taxonomic group.

The lowest acute value was the geometric mean 96-hour LC50 of 0.052 µg l^{-1} for the shrimp, *Americamysis bahia*, calculated from empirical LC50 values from a number of good quality studies. As with the freshwater PNEC, it is recommended that the PNEC be derived on the basis of general guidance given in the TGD on effects assessment for intermittent releases. Because permethrin acts specifically on the nervous system and crustaceans belong to the most sensitive organisms, a reduced assessment factor of 50 (instead of 100) is recommended in order to extrapolate from the 50 per cent acute effect level to the short-term no-effect level. This results in a $PNEC_{saltwater\_st}$ of 0.001 µg l^{-1}.

There is no existing short-term EQS for permethrin.

5.2.5 PNEC for secondary poisoning

For both freshwater and saltwater, PNECs based on the risks of secondary poisoning to mammals and birds (1.75 µg l^{-1}) are higher than those derived for the protection of aquatic life and so do not influence the development of EQSs for permethrin.

5.2.6 PNEC for sediments

Because the log Kow is >3, the derivation of a PNEC for the protection of benthic communities is required.

Two sediment studies are available and both the 10-day LC50 of 2.11 mg permethrin/kg sediment and the >20-day NOEC of 0.4 mg permethrin/kg sediment are suitable for PNEC derivation. Using the chronic toxicity data and the appropriate ssessment factors of 100 (chronic) for freshwater and 1,000 (chronic) for saltwater results in a $PNEC_{sediment\_freshwater}$ of 4.0 µg permethrin/kg sediment dry weight (dw), and a $PNEC_{sediment\_saltwater}$ of 0.4 µg permethrin/kg sediment dry weight (dw), respectively.

Table 5.1 Summary of proposed PNECs

Receiving medium/exposure scenario	Proposed PNEC (µg l^{-1}permethrin)	Existing EQS (µg l^{-1})
Freshwater/long-term	0.001	0.01
Freshwater/short-term	0.01	–
Saltwater/long-term	0.0002	0.01
Saltwater/short-term	0.001	–
Freshwater sediment/long-term	4.0 µg/kg dw	No standard
Saltwater sediment/long-term	0.4 µg/kg dw	No standard
Freshwater secondary poisoning	1.75	No standard
Saltwater secondary poisoning	1.75	No standard

5.3 Analysis

The lowest proposed PNECs derived for permethrin are 0.3 ng l^{-1} for waters and 0.4 µg/kg for sediments. The data quality requirements are that, at a third of the EQS, total error of measurement should not exceed 50 per cent. From the literature, it can be seen that analytical methodologies are capable of achieving detection limits in the low µg l^{-1} order in most media, suggesting that current analytical methods would not be adequate to analyse permethrin for compliance purposes.

5.4 Implementation issues

Before PNECs for permethrin can be adopted as EQSs, it will be necessary to address the following issues:

- The provision of additional data for marine species (such as echinoderms or molluscs) potentially through further testing, in order to reduce the uncertainty factor applied in the derivation of long-term and short-term.

- Current analytical methods may not be sensitive enough to assess compliance with proposed PNECs in receiving waters. This will require further consideration.

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List of abbreviations

AF	assessment factor
a.i.	active ingredient
ASTM	American Society for Testing and Materials
ATP	adenosine triphosphate
BCF	bioconcentration factor
bw	body weight
CAS	Chemical Abstracts Service
Cl_2CA	3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropanecarboxylic acid
EC	emulsifiable concentrate
EC50	concentration effective against 50% of the organisms tested
ECx	concentration effective against X% of the organisms tested
ECB	European Chemicals Bureau
ECD	electron capture detection
ELS	early life stage
EQS	Environmental Quality Standard
GC-MS	gas chromatography/mass spectrometry
GLC	gas liquid chromatography
GLP	Good Laboratory Practice (OECD)
GM	geometric mean
HPLC	high pressure liquid chromatography
HSDB	Hazardous Substances Data Bank
IRIS	Integrated Risk Information System
IUPAC	International Union of Pure and Applied Chemistry
LC50	concentration lethal to 50% of the organisms tested
LOAEL	lowest observed adverse effect level
LOEC	lowest observed effect concentration
lt	long term
NOAEL	no observed adverse effect level
NOEC	no observed effect concentration
NR	not reported
OECD	Organisation for the Economic Co-operation and Development
PNEC	predicted no-effect concentration
secpois	secondary poisoning

SEPA	Scottish Environment Protection Agency
SNIFFER	Scotland & Northern Ireland Forum for Environmental Research
SSD	species sensitivity distribution
st	short term
TGD	Technical Guidance Document
UKTAG	UK Technical Advisory Group
US EPA	US Environmental Protection Agency
UV	ultraviolet
WFD	Water Framework Directive
WHO	World Health Organization

ANNEX 1 Data quality assessment sheets

Identified and ordered by reference number (see References & Bibliography).

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 number	9

Information on the test species	
Test species used	*Oncorhynchus mykiss*
Source of the test organisms	Not stated
Holding conditions prior to test	Not stated
Life stage of the test species used	Juveniles

Information on the test design	
Methodology used	Not stated
Form of the test substance	Formulation 40:60 *cis*/*trans*
Source of the test substance	Not stated
Type and source of the exposure medium	Not stated
Test concentrations used	Not stated
Number of replicates per concentration	Not stated
Number of organisms per replicate	Not stated
Nature of test system (Static, Semi-static or, Flow- through, duration, feeding)	Flow-through
Measurement of exposure concentrations	Not stated
Measurement of water quality parameters	pH, hardness and temperature
Test validity criteria satisfied	Not stated
Water quality criteria satisfied	Not stated
Study conducted to GLP	Not stated
Overall comment on quality	Only limited data to assess study. Value is very low in comparison with other trout data so is likely to be an outlier.

Reliability of study	Reliable
Relevance of study	Relevant
Klimisch Code	4

Reference number	12

Information on the test species	
Test species used	*Brachycentrus americanus* (caddisfly) *Pteronarcys dorsata* (stonefly)
Source of the test organisms	Streams in the Duluth, MN, area
Holding conditions prior to test	Adaptation to test temperature at least for 1 week prior to test. *P. dorsata* was fed on a diet of birch and poplar leaves collected in autumn and soaked with water. *B. americana* was maintained on a diet of thawed adult brine shrimp. Unfiltered Lake Superior water was used for all rearing and testing. Dissolved oxygen, alkalinity, pH and hardness determined with methods outlined by the American Public Health Association (1975).* Dissolved oxygen was >95% saturation, pH 7.6–7.8, harness 46–48 mg l^{-1} as $CaCO_3$ and 15 $\pm$ 0.6°C. Photoperiod 14 hours light by fluorescent bulbs at ~194–312 lux).
Life stage of the test species used	larvae

Information on the test design	
Methodology used	Toxicity test with a flow-through exposure system
Form of the test substance	Permethrin lot no. 909, experimental purity
Source of the test substance	ICI Corp.
Type and source of the exposure medium	Dechlorinated tap water, hardness adjusted to 100 µg l^{-1} (as $CaCO_3$).
Test concentrations used	*B. americana*: 0.52, 0.22, 0.12, 0.064, 0.030 µg l^{-1} *P. dorsata*: 0.43, 0.21, 0.12, 0.042, 0.029 µg l^{-1} plus control
Number of replicates per concentration	2
Number of organisms per replicate	10
Nature of test system (static, semi-static or flow-through, duration, feeding)	Flow-through, feeding
Measurement of exposure concentrations	Yes, by GC. Accuracy of analytical procedure checked. Mean recovery of permethrin 85%. Reported concentrations are measured and corrected for recovery.
Measurement of water quality parameters	Yes (see above)
Test validity criteria satisfied	Not stated

Water quality criteria satisfied	Not stated
Study conducted to GLP	Flow-through toxicity tests following US EPA standard test procedures.
Overall comment on quality	Good

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

* American Public Health Association (APHA), 1975 *Standard methods for the examination of water and waste water*. 14th ed. Washington, DC: APHA.

Reference number	15

Information on the test species	
Test species used	*Chironomus riparius* (midge) *Elodea canadensis* (pond weed)
Source of the test organisms	See below.
Holding conditions prior to test	Five ponds, 5 × 5 m surface area sloping to 4 × 4 m at the bottom, lined with butyl rubber pond liner at the premises of WRc plc, Medmenham, UK. The ponds contained 5–10 cm sediment layer from the CS Lewis Nature Reserve, Oxford, a known clean site and a 60-cm depth of uncontaminated water from the River Thames. Plants and invertebrates were present in the ponds through natural colonisation, although a dense growth of pond weed (mostly *Elodea canadensis*) was removed by raking 27 days before dosing. A regression design was used for the experiments and the ponds were dosed with the commercially available formulation 'Picket' (Zeneca Agrochemicals, UK) at the beginning of July to achieve initial nominal concentrations of 0 (control), 1, 10, 50 and 100 µg l^{-1} permethrin
Life stage of the test species used	larvae (midge)

Information on the test design	
Methodology used	A regression design was used for the experiments.
Form of the test substance	Commercially available formulation 'Picket'
Source of the test substance	Zeneca Agrochemicals, UK
Type and source of the exposure medium	Uncontaminated water from the River Thames
Test concentrations used	Initial nominal concentrations of 0 (control), 1, 10, 50 and 100 µg l^{-1} permethrin
Number of replicates per concentration	1 mesocosm
Number of organisms per replicate	NA (freshwater pond community)
Nature of test system (static, semi-static or flow-through, duration, feeding)	Static mesocosm, naturally established trophic web, no extra feeding
Measurement of exposure concentrations	Yes, by gas liquid chromatography (GLC), but in sediment only.
Measurement of water quality parameters	Temperature, pH, dissolved oxygen, turbidity
Test validity criteria satisfied	Not stated
Water quality criteria satisfied	Not stated

Study conducted to GLP	-
Overall comment on quality	Good, however water concentrations not measured.

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

Reference number	15

Information on the test species	
Test species used	*Chironomus riparius* (midge)
Source of the test organisms	Laboratory stock
Holding conditions prior to test	Egg masses were maintained under constant temperature (20 ± 1°C) and light (16 hours light/8 hours dark) conditions. Larvae were reared in 8-litre aquaria with 2 cm substrate of fine acid-washed quartz-sand with 6 cm overlying culture water (Royal Holloway groundwater). Larvae were fed daily with finely ground TetraMin® fish food.
Life stage of the test species used	8–10-day-old larvae

Information on the test design	
Methodology used	Laboratory sediment test
Form of the test substance	Commercially available formulation 'Picket'
Source of the test substance	Zeneca Agrochemicals, UK
Type and source of the exposure medium	Natural sediment from an uncontaminated experimental pond, culture water (Royal Holloway groundwater.
Test concentrations used	Nominal concentrations (ng/g) of 0 (control), 0.43, 4.3, 22, 43, 220, 430 and 4,300.
Number of replicates per concentration	3
Number of organisms per replicate	15
Nature of test system (static, semi-static or flow-through, duration, feeding)	Static sediment toxicity test. No feeding of test animals during tests
Measurement of exposure concentrations	Yes, by GLC, but in sediment only.
Measurement of water quality parameters	Temperature, pH, dissolved oxygen, turbidity
Test validity criteria satisfied	Sensitivity of the test organism was checked according to ASTM 1995*
Water quality criteria satisfied	Not stated
Study conducted to GLP	Not stated
Overall comment on quality	Good, but concentrations in sediment and overlying water not measured.

Reliability of study	**Reliable (-)**
Relevance of study	**Relevant**
Klimisch Code	**2**

* American Society for Testing and Materials (ASTM), 1995 *Standard guide for conducting sediment toxicity tests with freshwater invertebrates*. ASTM 1995 Annual Book of Standards. Vol. 11.04, E1706-95. West Conshohocken, PA: ASTM.

Reference number	16

Information on the test species	
Test species used	*Americamysis bahia* (opossum shrimp) *Pennaeus duorarum* (pink shrimp)
Source of the test organisms	Laboratory cultures
Holding conditions prior to test	Static culture maintained at 25°C and 25‰ salinity.
Life stage of the test species used	*A. bahia*: ≤24-hour-old post-release juveniles *P. duorarum*: 3–5-day-old post-larvae

Information on the test design	
Methodology used	Static, 96-hour acute toxicity test according to ASTM 1988 guidelines.*
Form of the test substance	Not stated. Stock solutions were made by dissolving permethrin in 90% triethylene glycol and 10% acetone.
Source of the test substance	U.S. EPA repository
Type and source of the exposure medium	Sand and 1 µm fibre-filtered natural sea water adjusted to 25‰ salinity with deionised water
Test concentrations used	Five plus controls
Number of replicates per concentration	2
Number of organisms per replicate	10
Nature of test system (static, semi-static or flow-through, duration, feeding)	Static, according to ASTM 1988 guidelines. Feeding with *Artemia nauplii.*
Measurement of exposure concentrations	Stock solutions measured by GC prior to dosing
Measurement of water quality parameters	Yes (temperature, dissolved oxygen, pH, salinity)
Test validity criteria satisfied	Not stated
Water quality criteria satisfied	Not stated
Study conducted to GLP	According to ASTM 1988 guidelines
Overall comment on quality	Good

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

* American Society for Testing and Materials (ASTM), 1988 *Standard guide for conducting early life-stage toxicity tests with fishes*. E1241-88. pp. 26. West Conshohocken, PA: ASTM.

Reference number	17

Information on the test species	
Test species used	*Hexangenia rigide*
Source of the test organisms	Not stated
Holding conditions prior to test	Not stated
Life stage of the test species used	Not stated

Information on the test design	
Methodology used	Not stated
Form of the test substance	Not stated
Source of the test substance	Not stated
Type and source of the exposure medium	Not stated
Test concentrations used	Not stated
Number of replicates per concentration	Not stated
Number of organisms per replicate	Not stated
Nature of test system (static, semi-static or flow-through, duration, feeding)	Static
Measurement of exposure concentrations	Measured
Measurement of water quality parameters	Yes (temperature, dissolved oxygen, pH, salinity)
Test validity criteria satisfied	Not stated
Water quality criteria satisfied	Not stated
Study conducted to GLP	Not stated
Overall comment on quality	Reasonable

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

Reference number	18

Information on the test species	
Test species used	*Chlamydomonas reinhardtii*
Source of the test organisms	Cell culture of *Chlamydomonas reinhardtii* WT donated by Dr P E Brayant, Institute for Biology, Frankfurt, Germany
Holding conditions prior to test	Synthetic medium, continuous light from a band of fluorescent tubes (3,750 lux), 25°C
Life stage of the test species used	Vegetative growing cells

Information on the test design	
Methodology used	Algae test
Form of the test substance	Permethrin (93% purity); stock solution prepared with absolute ethanol. Sterilised by filtration through 0.45 µm filter, diluted to desired concentrations with presterilised growth medium.
Source of the test substance	The Alkali Chemical Corporation, Calcutta, India
Type and source of the exposure medium	Synthetic medium, not further specified but references regarding culture conditions cited
Test concentrations used	5 concentration levels (1.2×10^{-5} M – 1×10^{-3} M) plus control
Number of replicates per concentration	Not stated
Number of organisms per replicate	Inoculum 2.5×10^{6} cells (in a total volume of 20.5 ml)
Nature of test system (static, semi-static or flow-through, duration, feeding)	Static
Measurement of exposure concentrations	Nominal concentrations
Measurement of water quality parameters	Not stated
Test validity criteria satisfied	Not stated
Water quality criteria satisfied	Not stated
Study conducted to GLP	Not stated
Overall comment on quality	Moderate. Reference to nominal concentrations. Description of test conditions rather vague. However, references cited with regard to test conditions may be satisfactory.

Reliability of study	**Unreliable**
Relevance of study	**Relevant**
Klimisch Code	**3**

Reference number	22

Information on the test species	
Test species used	*Catostomus commersoni* (white sucker)
Source of the test organisms	White sucker larvae were hatched and reared in the laboratory from eggs obtained from five females that were dry-fertilised with milt from five males. Adults were seined from Oakville Creek, Oakville, and Ontario.
Holding conditions prior to test	The eggs were maintained in groups of approximately 500 in polyethylene strainers floating on the top of a 58-litre tank. The tank was aerated and supplied with 60 l h^{-1} temperature-controlled well water at 14.2°C. Eggs were treated daily (5-minute dip in 4 mg l^{-1} malachite green) to prevent fungal growth. At the first sign of hatch (10 days post-fertilisation), this procedure was discontinued. When the larvae were 1-day-old, the temperature in their tanks was raised to 20°C over a 12-hour period, after which the larvae were transferred to 23-litre aquaria (one aquarium for one batch of larvae obtained from a group of 500 eggs). Water temperature was kept at 20°C; water exchange rate in each aquarium was 120 l/day.
Life stage of the test species used	Larvae, 13, 20, 26 days old, either fed or unfed

Information on the test design	
Methodology used	
Form of the test substance	Permethrin 94.4% pure; Cat. No. PS 758, Lot No. 3-46; Chemical Service, West Chester, PA; vapour pressure $<10^{-6}$ mmHg, water solubility less than 1 mg l^{-1}. Added to test tanks in 95% ethanol carrier from a 100 mg l^{-1} stock 15 minutes before the start of exposure.
Source of the test substance	Chemical Service, West Chester, PA
Type and source of the exposure medium	Well water, 20.5°C, pH 8.09, dissolved oxygen 9 mg l^{-1}, total hardness 384 mg l^{-1} as $CaCO_3$.
Test concentrations used	0.1, 1, 10 and 100 µg l^{-1} plus a control and a carrier solvent control. After 2-hour pulse exposure, fish returned to clean exposure medium.
Number of replicates per concentration	6

Number of organisms per replicate	10
Nature of test system (static, semi-static or flow-through, duration, feeding)	Static. Food, when provided, consisted of two feedings per day with brine shrimp *Artemia nauplii*. All fish were fed twice daily until the start of the bioassay.
Measurement of exposure concentrations	Yes – 1 hour after beginning of exposure. GLC with ECD. Mean measured concentrations ranged from 97–106% of nominal.
Measurement of water quality parameters	Yes (dissolved oxygen, pH, temperature, total hardness, alkalinity)
Test validity criteria satisfied	Not stated
Water quality criteria satisfied	Not stated
Study conducted to GLP	Not stated
Overall comment on quality	Good

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

Reference number	24

Information on the test species	
Test species used	*Procambarus clarkii*
Source of the test organisms	Ponds of the Ben Hur Research Farm, Louisiana Agricultural Experiment Station, Baton Rouge, LA.
Holding conditions prior to test	Adaptation in 50-litre polyethylene tanks in the laboratory at 21–23°C for 10 days. For each tank, 10 litres of the pond water from which the animals were collected was used. 25% of this water was replaced daily by dechlorinated tap water adjusted to 100 mg l^{-1} hardness (as $CaCO_3$). Dissolved oxygen in the water was maintained at ≥60% saturation. Daily feeding with trout ration. Less than 15% mortality occurred during acclimation.
Life stage of the test species used	8–12; 25–35, 45–55, 65–75 mm body length

Information on the test design	
Methodology used	
Form of the test substance	Commercial permethrin formulation (GFU330). EC with 25.6% a.i. (w/v).
Source of the test substance	ICI America, Inc.
Type and source of the exposure medium	Dechlorinated tap water, hardness adjusted to 100 µg l^{-1} (as $CaCO_3$).
Test concentrations used	Six concentration levels plus control. Individual levels for each size class. Geometric series between 0 and 100% mortality of the respective size class as found in range-finding tests.
Number of replicates per concentration	3
Number of organisms per replicate	12–30 depending on size class
Nature of test system (static, semi-static or flow-through, duration, feeding)	Static, no feeding
Measurement of exposure concentrations	No
Measurement of water quality parameters	Yes (dissolved oxygen, pH, temperature, ammonia, total hardness, alkalinity and conductivity)
Test validity criteria satisfied	Not stated
Water quality criteria satisfied	Not stated
Study conducted to GLP	Static acute toxicity tests following standard test procedures (US EPA 1975* and APHA *et. al.* 1985).**

Overall comment on quality	Good – but exposure concentrations not analysed.

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

* US Environmental Protection Agency (US EPA), 1975 *Methods for the acute toxicity tests with fish, macroinvertebrates and amphibians*. The Committee on Methods for Toxicity Tests with Aquatic Organisms, Ecological Research Series EPA-660-75-009. Washington, DC: US EPA.

** American Public Health Association (APHA), 1985 *Standard methods for the examination of water and waste water* (15th edn.), 1268 pp. Washington, DC: APHA.

Reference number	25

Information on the test species	
Test species used	*Procambarus zonangulus* (White River crayfish) *Procambarus clarkii* (Red Swamp crayfish)
Source of the test organisms	Naturally reproducing population, in part indigenous to the experimental ponds, in part stocked from other ponds
Holding conditions prior to test	Ambient natural conditions
Life stage of the test species used	Entire populations of the crayfish species

Information on the test design	
Methodology used	Field test. One single application to each earthen experimental pond to yield nominal initial concentrations between 1 and 3 µg l^{-1} permethrin. Nine untreated ponds adjacent to the testing area served as controls.
Form of the test substance	Not stated
Source of the test substance	Not stated
Type and source of the exposure medium	Ambient surface water
Test concentrations used	4 plus controls
Number of replicates per concentration	1–3
Number of organisms per replicate	NA
Nature of test system (static, semi-static or flow-through, duration, feeding)	Static, feeding on the resources provided by the pond ecosystems. No complimentary feeding
Measurement of exposure concentrations	No
Measurement of water quality parameters	Yes (temperature, dissolved oxygen, pH, hardness, alkalinity, BOD5, total organic carbon, total solids)
Test validity criteria satisfied	Not stated
Water quality criteria satisfied	Not stated
Study conducted to GLP	Not stated
Overall comment on quality	Moderate

Reliability of study	**Reliable (-)**
Relevance of study	**Relevant**
Klimisch Code	**2**

BOD5 = biochemical oxygen demand (over 5 day period)

Reference number	32

Information on the test species	
Test species used	*Daphnia magna* *Ceriodaphnia dubia*
Source of the test organisms	*D. magna* and *C. dubia* were provided by the US EPA Environmental Research Laboratory in Duluth, MN.
Holding conditions prior to test	473 ml Manson jars, reconstituted moderately hard water (reference to standard method cited) at 25 ± 1°C under continuous low light (<10 µmol/s per m^2). Feeding with mixed algae culture.
Life stage of the test species used	Neonates

Information on the test design	
Methodology used	Sub-chronic daphnia test
Form of the test substance	Technical grade emulsified concentrate (EC) permethrin [25.6% a.i. (w/v)].
Source of the test substance	ICI Chemical Co.
Type and source of the exposure medium	Reconstituted medium hard water according to standard procedure (reference given in the paper)
Test concentrations used	Four concentration levels (0.5, 1.0, 3.0 and 9.0 µg l^{-1} permethrin) plus control
Number of replicates per concentration	10
Number of organisms per replicate	1
Nature of test system (static, semi-static or flow-through, duration, feeding)	Static, feeding
Measurement of exposure concentrations	Yes, by GC
Measurement of water quality parameters	Not stated, but use of reconstituted water
Test validity criteria satisfied	Not stated
Water quality criteria satisfied	Not stated
Study conducted to GLP	Not stated
Overall comment on quality	Good – acute test with measured exposure concentrations

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

Reference number	34

Information on the test species	
Test species used	*Daphnia magna* *Lepomis macrochirus* *Oncorhynchus kisutch* *Oncorhynchus mykiss* *Pimephales promelas* *Salmo salar* *Hexagenia bilineata* *Lymnaea stagnalis* *Penaeus duorarum* *Uca pugilator* *Crassostrea gigas, Crassostrea virginica*
Source of the test organisms	Not stated
Holding conditions prior to test	Not stated
Life stage of the test species used	Not stated

Information on the test design	
Methodology used	Not stated
Form of the test substance	Not stated
Source of the test substance	Not stated
Type and source of the exposure medium	Not stated
Test concentrations used	Not stated
Number of replicates per concentration	Not stated
Number of organisms per replicate	Not stated
Nature of test system (static, semi-static or flow-through, duration, feeding)	Not stated
Measurement of exposure concentrations	Not stated
Measurement of water quality parameters	Not stated
Test validity criteria satisfied	Not stated
Water quality criteria satisfied	Not stated
Study conducted to GLP	Not stated
Overall comment on quality	Data were quality assessed as part of the US pesticides programme and deemed suitable for registration purposes.
Reliability of study	**Reliable**
Relevance of study	**Relevant**
Klimisch Code	**2**

Reference number	35

Information on the test species	
Test species used	*Aedes aegypti* (yellow fever mosquito)
Source of the test organisms	Laboratory culture
Holding conditions prior to test	Adult mosquitoes were fed a 10% sucrose solution. Adult female mosquitoes were fed blood from a laboratory rat every 10–14 days. Eggs were collected on moist filter paper, air-dried and held in an environmental chamber (25°C) until needed. To obtain larvae for toxicity tests, a pan containing 10 mg of brewers yeast and 25 mg of liver powder were mixed in 500 ml tap water and placed in an environmental chamber at 25°C for 24 hours. Then a filter paper with eggs was placed in the pan. Unhatched eggs were removed after 6–8 hours to ensure a cohort of larvae of the same age.
Life stage of the test species used	Larvae used for testing generally were 3rd instars 72-hours post-hatch.

Information on the test design	
Methodology used	Static acute toxicity test
Form of the test substance	Technical grade permethrin (90.8% a.i.) and microencapsulated permethrin (20% a.i.)
Source of the test substance	Chipman, Inc., Stoney Creek, Ontario
Type and source of the exposure medium	City of Guelph tap water at 25°C, chlorine removed
Test concentrations used	6 plus control
Number of replicates per concentration	3–5
Number of organisms per replicate	20
Nature of test system (static, semi-static or flow-through, duration, feeding)	Static
Measurement of exposure concentrations	Technical grade permethrin: yes, by GC Microencapsulated permethrin: no, due to difficulties in separating permethrin in solution from that remaining in capsules.
Measurement of water quality parameters	Not stated (except temperature and pH)
Test validity criteria satisfied	Not stated
Water quality criteria satisfied	Not stated
Study conducted to GLP	Not stated
Overall comment on quality	Good

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

Reference number	36

Information on the test species	
Test species used	Blackflies (*Simulium* sp.) Mayflies (*Isonychia* sp.) Caddisflies (*Pycnopsyche* sp.) Stoneflies (*Acroneuria* sp.) Dragonflies (*Ophiogomphus* sp.) Crayfish (*Orconectes* sp.)
Source of the test organisms	Icewater Creek and Goulais River near Searchmount, Ontario.
Holding conditions prior to test	Test animals collected in Icewater Creek or Goulais River were placed in the bioassay system and allowed to adapt for 4 hours prior to testing.
Life stage of the test species used	Insect larvae, life stage of larvae and the crayfish species *Orconectes* not further specified

Information on the test design	
Methodology used	Continuous flow test system
Form of the test substance	Not mentioned but cited as the 'same as used by Poirier and Surgeoner 1987'
Source of the test substance	Not stated
Type and source of the exposure medium	Natural creek water from Icewater Creek
Test concentrations used	Not mentioned except control (0 µg l^{-1})
Number of replicates per concentration	2–4
Number of organisms per replicate	8–20 depending on test organism
Nature of test system (static, semi-static or flow-through, duration, feeding)	Flow-through, no extra feeding
Measurement of exposure concentrations	No
Measurement of water quality parameters	Yes, before each test-by-test kit (temperature, pH, hardness).
Test validity criteria satisfied	Not stated
Water quality criteria satisfied	Not stated
Study conducted to GLP	-
Overall comment on quality	Good, but water concentrations not measured

Reliability of study	Reliable
Relevance of study	Relevant
Klimisch Code	2

Reference number	37

Information on the test species	
Test species used	Freshwater fish *Oncorhynchus clarki henshawi* (lahontan cut-throat trout) *Oncorhynchus clarkii stomias* (greenback cut-throat trout) *Oncorhynchus gilae apache* (apache trout) *Oncorhynchus mykiss* (rainbow trout) Saltwater fish *Cyprinodon bovinus* (Leon Springs pupfish) *Cyprinodon variegatus* (sheepshead minnow)
Source of the test organisms	Various governmental and commercial sources not further specified but a reference describing the sources is cited.
Holding conditions prior to test	Freshwater fish: Flowing well water until testing Saltwater fish: sea water diluted to 2‰ salinity with deionised water
Life stage of the test species used	Juveniles; ca. 0.2–1 g body weight

Information on the test design	
Methodology used	Static acute toxicity test
Form of the test substance	99% a.i.
Source of the test substance	IC America, Richmond, CA
Type and source of the exposure medium	Freshwater fish: reconstituted hard water (hardness 160–180 mg l^{-1} as $CaCO_3$). Water quality alkalinity, hardness, pH measured on each batch. Saltwater fish: natural sea water diluted to 2‰ salinity with deionised water
Test concentrations used	6 plus control
Number of replicates per concentration	2 (saltwater), 3 (freshwater)
Number of organisms per replicate	10
Nature of test system (static, semi-static or flow-through, duration, feeding)	Static
Measurement of exposure concentrations	Yes, by GC
Measurement of water quality parameters	Yes. Temperature, dissolved oxygen and pH in all tests, alkalinity and hardness in freshwater only.
Test validity criteria satisfied	Not stated
Water quality criteria satisfied	Not stated

Study conducted to GLP	Tests in accordance with US EPA and ASTM (guidelines cited)
Overall comment on quality	Good

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

Reference number	38

Information on the test species	
Test species used	*Americamysis (Mysidopsis) bahia* (opossum shrimp) *Pennaeus duorarum* (pink shrimp) *Cyprinodon variegatus* (sheepshead minnow) *Menidia menidia* (Atlantic silverside)
Source of the test organisms	All test animals except Atlantic silversides were either collected from estuarine waters adjacent to the Environmental Research Laboratory (ERL), Gulf Breeze, FL, or cultured in laboratory from laboratory stock. Silversides were shipped as embryos to the laboratory from Charleston, SC, and reared at the Gulf Breeze ERL.
Holding conditions prior to test	Fishes were acclimated to laboratory conditions at least 14 days prior to testing.
Life stage of the test species used	*M. bahia*: ≤24-hour-old individuals Not stated for other test organisms

Information on the test design	
Methodology used	Flow-through, 96-hour acute toxicity test according to ASTM 1980 guidelines,* except that *Artemia nauplii* were fed to *M. bahia* and silversides to prevent starvation during the 96-hour test period.
Form of the test substance	Technical grade permethrin, 93% purity. Stock solutions were made by dissolving permethrin in triethylene glycol.
Source of the test substance	ICI Americas, Inc.
Type and source of the exposure medium	Sand and 1 µm fibre filtered natural sea water adjusted to 25‰ salinity with deionised water
Test concentrations used	Not stated, according to ASTM 1980 guidelines
Number of replicates per concentration	2
Number of organisms per replicate	20
Nature of test system (static, semi-static or flow-through, duration, feeding)	Flow-through, according to ASTM 1980 guidelines. Feeding with *Artemia nauplii.*
Measurement of exposure concentrations	Yes, by GC
Measurement of water quality parameters	According to ASTM 1980 guidelines
Test validity criteria satisfied	Not stated
Water quality criteria satisfied	Not stated
Study conducted to GLP	According to ASTM 1980 guidelines
Overall comment on quality	Good

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

* American Society for Testing and Materials (ASTM), 1980 *Standard practise for conducting static acute toxicity tests with fishes, macroinvertebrates and amphibians*. Standard E729-80. West Conshohocken, PA: ASTM.

Reference number	39

Information on the test species	
Test species used	*Daphnia magna* *Daphnia pulex* *Hydropsyche* spp. *Simulium vittatum* *Isonychia bicolor*
Source of the test organisms	*Hydropsyche* spp., *Simulium vittatum* and *Isonychia bicolor* were collected from streams around Guleph, Ontario, Canada. *Daphnia magna* and *Daphnia pulex* were obtained from laboratory cultures at the University of Guelph.
Holding conditions prior to test	Daphnia cultures were maintained in an environmental chamber at 18.5°C and a photoperiod of 16:8 light:dark. Laboratory grown *Chlamydomonas reinhardtii* were given as food. Other invertebrates were sorted in the laboratory and transferred to the test system (recirculating chambers) and allowed a 24-hour acclimation period. Temperature 14°C, 16:8 hour light:dark. Filtered (30 µm) river water, pH 7.8.
Life stage of the test species used	*Daphnia*: Neonates <48 hours Insects: Life stages as collected in the field

Information on the test design	
Methodology used	Acute toxicity test
Form of the test substance	1. Aqueous solution of microencapsulated permethrin (925:75 *cis*/*trans*) suspended in an aromatic solvent emulsifier, the capsule wall is a polyamide polyurea polymer and the average size of the capsules is 30 µm 2. EC permethrin, 50% v/v active ingredient of 40:60 *cis*/*trans* permethrin
Source of the test substance	1. Pennwalt Corp., France 2. Chipman Chemical, Canada
Type and source of the exposure medium	Daphnia: Filtered (30 µm) well water Insects: Filtered (30 µm) river water, pH 7.8
Test concentrations used	Daphnia: 7 concentration levels plus control Insects: 5 concentration levels plus control
Number of replicates per concentration	Daphnia: 5 Insects: 2
Number of organisms per replicate	Daphnia: 5 Insects: not reported
Nature of test system (static, semi-static or flow-through, duration, feeding)	Daphnia: static, feeding 12 hours before test Insects: recirculating chamber No feeding during tests
Measurement of exposure concentrations	Nominal concentrations

Measurement of water quality parameters	At least prior to test. Tests conducted in environmental chambers under controlled light and temperature conditions
Test validity criteria satisfied	Not stated
Water quality criteria satisfied	Not stated
Study conducted to GLP	Not stated
Overall comment on quality	Good but reference to nominal concentrations of specific permethrin formulations.

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

Reference number	39

Information on the test species	
Test species used	*Daphnia magna* *Daphnia pulex*
Source of the test organisms	*Daphnia magna* and *Daphnia pulex* were obtained from laboratory cultures at the University of Guelph.
Holding conditions prior to test	Daphnia cultures were maintained in an environmental chamber at 18.5°C and a photoperiod of 16:8 light:dark. Laboratory grown *Chlamydomonas reinhardtii* were given as food.
Life stage of the test species used	Daphnia: neonates <48 hours

Information on the test design	
Methodology used	Sub-chronic daphnia test
Form of the test substance	Aqueous solution of microencapsulated permethrin (925:75 *cis*/*trans*) suspended in an aromatic solvent emulsifier, the capsule wall is a polyamide polyurea polymer and the average size of the capsules is 30 µm
Source of the test substance	Pennwalt Corp., France
Type and source of the exposure medium	Daphnia*:* Filtered (30 µm) well water
Test concentrations used	Daphnia: 7 concentration levels plus control
Number of replicates per concentration	Daphnia: 5
Number of organisms per replicate	Daphnia: 5
Nature of test system (static, semi-static or flow-through, duration, feeding)	Static (because of the slow release nature of the microcapsules). Feeding every 2 days with *Chlamydomonas reindardii*.
Measurement of exposure concentrations	Not measured because no analytical procedure to measure the residual concentration of the penncapthrin microcapsule formulation in aqueous media has been developed.
Measurement of water quality parameters	At least prior to test. Tests conducted in environmental chambers under controlled light and temperature conditions
Test validity criteria satisfied	Not stated
Water quality criteria satisfied	Not stated
Study conducted to GLP	Not stated
Overall comment on quality	Moderate – subchronic test with reference to nominal concentrations of specific permethrin

	formulations.

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

Reference number	40

Information on the test species	
Test species used	*Pimephales promelas* (fathead minnow) *Heliosoma trivolvis* (snail)
Source of the test organisms	*Pimephales* and *Heliosoma*: cultures from US EPA Environmental Research Laboratory, Dultuth, MN
Holding conditions prior to test	Not mentioned
Life stage of the test species used	*P. promelas*: <1-day-old larvae *H. trivolvis*: individuals of 0.09–0.3 g weight

Information on the test design	
Methodology used	*P. promelas*: early life stage test for 32 days *H. trivolvis*: 28 day test
Form of the test substance	Technical grade permethrin, 92% a.i.
Source of the test substance	ICI Americas, Inc. Goldsboro, NC
Type and source of the exposure medium	Lake Superior water filtered through sand, sterilised with ultraviolet light and heated to 25 ± 2°C for *P. promelas* and 15 ± 2°C for *H. trivolvis*.
Test concentrations used	Five plus control. Saturated solutions of permethrin were used to avoid the use of solvent chemicals. A concentration of 16 µg l^{-1} was maintained in the saturator. Toxicant solution was delivered to the diluter system via fluid metering pumps to produce desired concentrations.
Number of replicates per concentration	4 (fish) 2 (snails)
Number of organisms per replicate	15 (fish) 10 (snails)
Nature of test system (static, semi-static or flow-through, duration, feeding)	Continuous flow mini-diluter exposure system as described by Benoit *et al.* 1982.* Flow rate in each exposure chamber of 7 cm width, 19 cm length and 4.5 cm depth was 12.5 ml per min.
Measurement of exposure concentrations	Yes, by GC
Measurement of water quality parameters	Yes, according to APHA *et al.* 1980** methods: hardness, alkalinity, acidity, pH, dissolved oxygen, temperature
Test validity criteria satisfied	Not stated
Water quality criteria satisfied	Not stated
Study conducted to GLP	Not stated
Overall comment on quality	Good

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

* Benoit D A, Mattson V R and Olson D L, 1982 *A continuous-flow mini-diluter system for toxicity testing*. Water Research, **16**, 457–464.
** American Public Health Association (APHA), American Water Works Association (AWWA) and Water Pollution Control Federation (WPCF), 1980 *Standard methods for the examination of water and wastewater*. 15th ed. pp. 1134. Washington, DC: APHA.

Reference number	42

Information on the test species	
Test species used	Indigenous stream invertebrates of the insect orders Plecoptera and Ephemeroptera, indigenous fish fry (*Salvelinus malma*) and periphyton.
Source of the test organisms	Indigenous to the stream used as test site.
Holding conditions prior to test	Natural conditions at test site.
Life stage of the test species used	Life stage of the insect larvae not further specified; two size classes of fish fry used (2 and 5 cm length)

Information on the test design	
Methodology used	Field test
Form of the test substance	Pounce 0.5% EC
Source of the test substance	FMC Corporation, Princeton, NJ
Type and source of the exposure medium	Natural stream water
Test concentrations used	Contamination in stream after spraying of trees located at stream banks monitored. 5 hours post-application 0.05 µg l^{-1}, 8–11 hours post-application 0.14 µg l^{-1}, 14-hours post-application 0.02 µg l^{-1}. At two control sites 800 m upstream and 500 m downstream of the treatment area, no permethrin residues (i.e. <0.01 µg l^{-1}) were found in water.
Number of replicates per concentration	Active biomonitoring by placing exposure devices into the stream at the two control sites and the treatment area was performed. Two replicates at each site were exposed for each of the two size classes of fish and the benthic insects actively exposed.
Number of organisms per replicate	10 for each insect species 3–5 for fish
Nature of test system (static, semi-static or flow-through, duration, feeding)	Field test in a natural stream. Actively exposed fish were feed with stream invertebrates during the test.
Measurement of exposure concentrations	Yes
Measurement of water quality parameters	No (except nutrients such as P- and N-species and the cations Ca, Mg, Mn, Na, K, Si). The nutrient measurements were intended to detect an impact on periphyton viability.
Test validity criteria satisfied	Not stated
Water quality criteria satisfied	Not stated

Study conducted to GLP	Not stated
Overall comment on quality	Good

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

Reference number	**43**

Information on the test species	
Test species used	Natural pelagic community of a pond ecosystem, in particular: *Ceratium hirundinella* (Dinoflagellatae*)* *Chaoborus flavicans*
Source of the test organisms	Indigenous in pond used for testing
Holding conditions prior to test	Natural conditions in pond
Life stage of the test species used	all (*C. hirundinella*) pelagic larvae (*C. flavicans*)

Information on the test design	
Methodology used	Enclosures (stainless steel frame covered with polyethylene film; 1 m diameter, 3.8 m deep)
Form of the test substance	EC permethrin (mixture of *cis*- and *trans*-isomer)
Source of the test substance	Not stated
Type and source of the exposure medium	Eutrophic water of a natural pond (1.4 µg l^{-1} PO_4-P, 7.4 µg l^{-1} NO_3-N, 1.36 µg l^{-1} NO_2-N, 13.6 µg l^{-1} NH_4-N, dissolved oxygen 8.0 mg l^{-1} near surface – 0.03 mg l^{-1} near bottom, 23–30°C)
Test concentrations used	Enclosure 1: 0 (control) Enclosure 2: first treatment 0.75 µg l^{-1}, second treatment (14 days later) 10 µg l^{-1}. Enclosure 3: first treatment 1.5 µg l^{-1}, second treatment (14 days later) 1.5 µg l^{-1}.
Number of replicates per concentration	1
Number of organisms per replicate	*C. hirundinella*: >500 to >1,000 l^{-1} (control) *C. flavicans*: 12,700 ± 700 (control)
Nature of test system (static, semi-static or flow-through, duration, feeding)	Enclosure
Measurement of exposure concentrations	Measurement of permethrin residues in water and sediment by GC 2–5 days post-application
Measurement of water quality parameters	pH, dissolved oxygen and temperature
Test validity criteria satisfied	Not stated
Water quality criteria satisfied	Not applicable
Study conducted to GLP	Not stated
Overall comment on quality	

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

Reference number	44

Information on the test species	
Test species used	*Cyprinodon variegatus* (sheepshead minnow)
Source of the test organisms	Eggs were obtained from 11 hormone-injected females and fertilised by using 5 or more males as described in a cited publication.
Holding conditions prior to test	Not stated
Life stage of the test species used	1.5–24-hour-old embryos

Information on the test design	
Methodology used	Early life stage test
Form of the test substance	Technical grade permethrin, 93% purity. Stock solutions were made by dissolving permethrin in triethylene glycol.
Source of the test substance	ICI Americas, Inc.
Type and source of the exposure medium	Sea water with a salinity ranging from 22–32‰
Test concentrations used	6 plus control
Number of replicates per concentration	4
Number of organisms per replicate	20
Nature of test system (static, semi-static or flow-through, duration, feeding)	Intermittent-flow system. Feeding with *Artemia nauplii*.
Measurement of exposure concentrations	Yes, at least weekly by GC
Measurement of water quality parameters	According to ASTM 1980 guidelines*
Test validity criteria satisfied	Not stated
Water quality criteria satisfied	Not stated
Study conducted to GLP	According to ASTM 1980 guidelines
Overall comment on quality	Good

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

*American Society for Testing and Materials (ASTM), 1980 *Standard practise for conducting static acute toxicity tests with fishes, macroinvertebrates and amphibians*. Standard E729-80. West Conshohocken, PA: ASTM.

Reference number	47

Information on the test species	
Test species used	*Chironomus riparius* (midge)
Source of the test organisms	Laboratory stock
Holding conditions prior to test	Egg masses were maintained under constant temperature (20 ± 2°C) and light (16 hours light/8 hours dark) conditions. Larvae were reared in 8-litre aquaria with 2 cm substrate of fine acid-washed quartz-sand with 6 cm overlying culture water (groundwater). Larvae were fed every 48h with finely ground TetraMin® fish food.
Life stage of the test species used	First instar larvae

Information on the test design	
Methodology used	Laboratory sediment test
Form of the test substance	Not stated
Source of the test substance	Not stated
Type and source of the exposure medium	Natural sediment from an uncontaminated experimental pond
Test concentrations used	Nominal concentrations (ng/g) of 0 (control), 200, 400, 800 and 1600,
Number of replicates per concentration	3
Number of organisms per replicate	21
Nature of test system (static, semi-static or flow-through, duration, feeding)	Static sediment toxicity test, test ended 5 days after last emergence in control vessels, test animals fed with 0.5 mg ground tetramin fish food per larva per day for the first 10 days and 1 mg ground tetramin fish food per larva per day for the remainder of the test
Measurement of exposure concentrations	Yes, (in sediment samples at beginning of test).
Measurement of water quality parameters	Yes (Temperature, pH and dissolved oxygen at beginning and end of test)
Test validity criteria satisfied	Yes (100% emergence in control vessels)
Water quality criteria satisfied	Not stated
Study conducted to GLP	Not stated
Overall comment on quality	Good,

Reliability of study	**Reliable (-)**
Relevance of study	**Relevant**
Klimisch Code	**2**

Reference	62

Information on the test species	
Test species used	*Pseudokirchneriella subcapitata*
Source of the test organisms	In house cultures
Holding conditions prior to test	Nutrient media (ATCC 22662)
Life stage of the test species used	Growth phase

Information on the test design	
Methodology used	The method is well described in the report.
Form of the test substance	Analytical material (>96% purity)
Source of the test substance	Sigma-Aldrich, Dorset, UK
Type and source of the exposure medium	Nutrient media
Test concentrations used	0 (control), 0.32, 1.0, 3.2, 10, 32, 100 and 320 µg l^{-1} (nominal permethrin concentrations)
Number of replicates per concentration	Six (for controls) and three (for treatments)
Number of organisms per replicate	Initial starting density = 5 x 10^3 cells/ml
Nature of test system (Static, semi-static or flow through, duration, feeding)	Static, 72 hours
Measurement of exposure concentrations	The test concentrations were analysed at the beginning and end of the test (measured values were 51% of nominal concentrations).
Measurement of water quality parameters	Yes (pH and temperature)
Test validity criteria satisfied	Yes (204 - 228 times increase in controls)
Water quality criteria satisfied	Yes
Study conducted to GLP	The study was carried out to the principles of GLP
Comments	The study was well conducted, is of good quality and the exposure concentrations used were measured.

Reliability of study	Reliable
Relevance of study	Relevant
Klimisch Code	2

Reference	63

Information on the test species	
Test species used	*Skeletonema costatum*
Source of the test organisms	In house cultures
Holding conditions prior to test	Nutrient media
Life stage of the test species used	Growth phase

Information on the test design	
Methodology used	The method is well described in the report.
Form of the test substance	Technical grade
Source of the test substance	Shell Chemical Company, San Ramon, California
Type and source of the exposure medium	Nutrient media
Test concentrations used	Not stated
Number of replicates per concentration	Three for controls and for treatments
Number of organisms per replicate	Not stated
Nature of test system (Static, semi-static or flow through, duration, feeding)	Static, 96 hours
Measurement of exposure concentrations	Not stated
Measurement of water quality parameters	Yes (temperature)
Test validity criteria satisfied	Not stated
Water quality criteria satisfied	Not stated
Study conducted to GLP	No
Comments	The study was well conducted but the exposure concentrations used were not measured.

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

ANNEX 2 Data sheets: water column data

Ordered and identified by reference numbers as listed in References & Bibliography.

Reference	**12**
CAS number	52645-53-1
Chemical	Permethrin
Chemical species	
Organism (common name)	Caddisfly Stonefly
Organism (scientific name)	*Brachycentrus americanus* (caddisfly) *Pteronarcys dorsata* (stonefly)
Life stage (e.g. egg, embryo, ELS, adult)	Larvae
Exposure regime (e.g. static, renewal, etc.)	Flow-through
Test method	Flow-through toxicity test
Analysis (measured or nominal)	Measured, accuracy check of analytical results
Temperature	15 ± 0.6°C
Hardness	46–48 mg l^{-1} (as $CaCO_3$)
pH	7.6–7.8
Salinity	Unfiltered Lake Superior water
Exposure duration	Up to 28 days
Endpoint (e.g. NOEC, EC50)	LC50, NOEC, effects on behaviour (mobility, feeding)
Effect (e.g. reproduction, survival, growth)	*Brachycentrus americanus*: No mortality or abnormal behaviour in control during entire test. No effect on behaviour at 0.03 µg l^{-1} for up to 28 days exposure. After 48 hours at 0.064 µg l^{-1}, 100% of the exposed animals showed behavioural changes (loss of feeding position, abnormal leg movements, etc.). The 21-day LC50 was 0.17 µg l^{-1}. However, after 28 days exposure at 0.03 µg l^{-1}, 55% of the exposed animals were dead (~10% mortality after 10 days at 0.03 µg l^{-1}). *Pteronarcys dorsata*: Within 2 hours, 25% of the animals were immobilised upon exposure to ≥0.21 µg l^{-1} (90% after 5 hours). At 0.12 µg l^{-1}, 65% were immobile after 96 hours. 21 days of exposure to 0.042 µg l^{-1} resulted in immobility of 100% of the exposed individuals. At 0.029 µg l^{-1}, no abnormal behaviour or other adverse effects were seen during the 28-day duration of the experiment (NOEC 0.03 µg l^{-1}). Immobilised animals did not often die (maximum death rate at any concentration 3 out of 10 animals exposed). Death presumably due to starvation as result of paralysis of the animals. Larvae were analysed in 10 groups for permethrin content. BCF values ranged from 43 to 570 with an average of 183 and a standard deviation of 171.
Concentration	See 'Effect'
Initial quality assessment (e.g. good, moderate, poor)	Good
Comments	

Reference	**15**
CAS number	52645-53-1
Chemical	Permethrin
Chemical species	
Organism (common name)	Mesocosm (pond community) Midge Pond weed
Organism (scientific name)	*Chironomus riparius* *Elodea canadensis*
Life stage (e.g. egg, embryo, ELS, adult)	Larvae
Exposure regime (e.g. static, renewal, etc.)	Mesocosm
Test method	Static
Analysis (measured or nominal)	Nominal concentrations in water, measured in sediment
Temperature	20–30°C at the surface, 10–15°C at the bottom
Hardness	Not reported
pH	9–10.5
Salinity	River Thames water
Exposure duration	Single application, post-application observation period 52 days
Endpoint (e.g. NOEC, EC50)	No specific endpoint mentioned. Description of observed effects.
Effect (e.g. reproduction, survival, growth)	*Elodea canadensis* rapidly recolonised the ponds and no differences in weed density could be observed at the end of the study. Knockdown of aquatic invertebrates, particularly hemipterans, was observed immediately after spraying of the ponds dosed with the highest concentrations (100, 50 and 10 µg l^{-1}). On day 2 post dosing, dead chironomid larvae were found in sediment grab samples from ponds dosed at 50 and 100 µg l^{-1}. No emergence of chironomid adults was seen in ponds dosed with 50 and 100 µg l^{-1} until days 24 and 31, respectively. At 10 µg l^{-1}, insects were collected at all sampling dates but numbers were much and significantly reduced relative to the control until day 24 post treatment. Chironomid emergence at 1 µg l^{-1} was similar to the control. Regression analysis revealed that dose had a significant effect on abundance of the chironomids.
Concentration	1 µg l^{-1} can be considered as study NOEC
Initial quality assessment (e.g. good, moderate, poor)	Good, but no measurement of toxicant concentration in the water body
Comments	

Reference	**15**
CAS number	52645-53-1
Chemical	Permethrin
Chemical species	
Organism (common name)	Midge
Organism (scientific name)	*Chironomus riparius*
Life stage (e.g. egg, embryo, ELS, adult)	8–10-day-old larvae
Exposure regime (e.g. static, renewal, etc.)	Static
Test method	Single species static sediment toxicity test
Analysis (measured or nominal)	Nominal concentrations
Temperature	20 ± 1°C
Hardness	Not reported
pH	Not reported
Salinity	Groundwater
Exposure duration	10 days
Endpoint (e.g. NOEC, EC50)	LC50
Effect (e.g. reproduction, survival, growth)	Survival of larvae
Concentration	2.11 mg/kg (dry wt)
Initial quality assessment (e.g. good, moderate, poor)	Good, but no measurement of toxicant concentration in sediment and overlying water body
Comments	

Reference	**16**
CAS number	52645-53-1
Chemical	Permethrin
Chemical species	
Organism (common name)	Opossum shrimp Pink shrimp
Organism (scientific name)	*Americamysis bahia* *Pennaeus duorarum*
Life stage (e.g. egg, embryo, ELS, adult)	*M. bahia*: ≤24-hour-old postrelease juveniles *P. duorarum*: 3–5-day-old postlarvae
Exposure regime (e.g. static, renewal, etc.)	Static
Test method	Static acute toxicity test according to ASTM 1988 guidelines for macronivertebrates
Analysis (measured or nominal)	Stock solutions measured by GC prior to dosing
Temperature	25°C
Hardness	Not reported but mentioned that all water quality parameters remained within limits set by ASTM
pH	
Salinity	Sand and 1 µm fibre-filtered natural sea water adjusted to 25‰ salinity with deionised water
Exposure duration	96 hours
Endpoint (e.g. NOEC, EC50)	LC50
Effect (e.g. reproduction, survival, growth)	Mortality
Concentration	*P. duorarum*: 0.17 (95% CI: 0.15–0.19) µg l^{-1} *A. bahia*: 0.095 (95% CI: 0.077–0.12) µg l^{-1}
Initial quality assessment (e.g. good, moderate, poor)	Good
Comments	

CI = confidence interval

Reference	**18**
CAS number	52645-53-1
Chemical	Permethrin
Chemical species	
Organism (common name)	Green algae
Organism (scientific name)	*Chlamydomonas reinhardtii*
Life stage (e.g. egg, embryo, ELS, adult)	Cells in vegetative (exponential) growth phase
Exposure regime (e.g. static, renewal, etc.)	Static
Test method	Alga test
Analysis (measured or nominal)	Nominal
Temperature	25°C
Hardness	Not reported
pH	Not reported
Salinity	Synthetic growth medium not further specified
Exposure duration	72 hours
Endpoint (e.g. NOEC, EC50)	Inhibition of cell growth (in percentage of cell number of control).
Effect (e.g. reproduction, survival, growth)	Inhibition of cell growth (in percentage of cell number of control). EC0 is 4.7 mg l^{-1} (1.2×10^{-5} M), EC100 391 mg l^{-1} (10^{-3} M). From Figure 1 of the publication, an EC10 of 5.1 mg l^{-1} (1.3×10^{-5} M) can be inferred.
Concentration	From Figure 1 of the publication, an EC10 of 5.1 mg l^{-1} (1.3×10^{-5} M) can be inferred.
Initial quality assessment (e.g. good, moderate, poor)	Moderate
Comments	No description of growth medium, no measurement of toxicant concentrations in test.

Reference	**22**
CAS number	52645-53-1
Chemical	Permethrin
Chemical species	
Organism (common name)	White sucker
Organism (scientific name)	*Catostomus commersoni*
Life stage (e.g. egg, embryo, ELS, adult)	Larvae, 13, 20 or 26 days old. Fed or unfed during 2 hours of exposure and subsequent 94-hour observation period.
Exposure regime (e.g. static, renewal, etc.)	Static
Test method	Static acute toxicity test
Analysis (measured or nominal)	Measured by GLC
Temperature	20.5°C
Hardness	384 mg l^{-1} (as $CaCO_3$)
pH	8.09
Salinity	Not stated
Exposure duration	2-hour pulse exposure plus 94 hours observation time
Endpoint (e.g. NOEC, EC50)	LC50
Effect (e.g. reproduction, survival, growth)	Mortality
Concentration	96-hour LC50: larvae, 13 days old: 184 µg l^{-1} (fed); 2 µg l^{-1} (unfed) larvae, 20 days old: 10 µg l^{-1} (fed); 1 µg l^{-1} (unfed) larvae, 26 days old: 3668 µg l^{-1} (fed); 172 µg l^{-1} (unfed)
Initial quality assessment (e.g. good, moderate, poor)	Good
Comments	

Reference	**24**
CAS number	52645-53-1
Chemical	Permethrin
Chemical species	
Organism (common name)	Crayfish
Organism (scientific name)	*Procambarus clarkii*
Life stage (e.g. egg, embryo, ELS, adult)	8–12 mm length 25–35 mm length 45–55 mm length 65–75 mm length
Exposure regime (e.g. static, renewal, etc.)	Static
Test method	Static acute toxicity test
Analysis (measured or nominal)	Nominal
Temperature	21–23°C
Hardness	100 mg l^{-1} (as $CaCO_3$)
pH	7.9–8.8
Salinity	Not stated
Exposure duration	96 hours
Endpoint (e.g. NOEC, EC50)	LC50
Effect (e.g. reproduction, survival, growth)	Mortality
Concentration	96-hour LC50: 8–12 mm length: 0.499, 0.282, 0.532 µg l^{-1} 25–35 mm length: 1.047, 0.695, 0.819 µg l^{-1} 45–55 mm length: 1.368, 1.266, 1.266 µg l^{-1} 65–75 mm length: 0.803; 0.645, 0.992 µg l^{-1}
Initial quality assessment (e.g. good, moderate, poor)	Good, but reference to nominal concentrations.
Comments	

Reference	**25**
CAS number	52645-53-1
Chemical	Permethrin
Chemical species	
Organism (common name)	White River crayfish Red Swamp crayfish
Organism (scientific name)	*Procambarus zonangulus* *Procambarus clarkii*
Life stage (e.g. egg, embryo, ELS, adult)	Entire populations of the crayfish species.
Exposure regime (e.g. static, renewal, etc.)	Static
Test method	Field test. One single application to each earthen experimental pond to yield nominal initial concentrations 1–3 µg l^{-1} permethrin. Nine untreated ponds adjacent to the testing area served as controls.
Analysis (measured or nominal)	No analysis of permethrin concentrations
Temperature	Natural ambient (ca. 19°C)
Hardness	220 ± 66 mg l^{-1} as $CaCO_3$
pH	7.6
Salinity	Freshwater
Exposure duration	One single application to each pond, observation for 7 days post-application
Endpoint (e.g. NOEC, EC50)	No specific endpoint. Description of observations
Effect (e.g. reproduction, survival, growth)	Mortality
Concentration	Initial nominal concentrations of 1–3 µg l^{-1} permethrin caused crayfish mortalities of 54–83% 7 days post-application. *Procambarus zonangulus* mortality was 100% 7 days post-application.
Initial quality assessment (e.g. good, moderate, poor)	Moderate
Comments	No analytical monitoring of toxicant concentrations

Reference	**32**
CAS number	52645-53-1
Chemical	Permethrin
Chemical species	
Organism (common name)	Water flea
Organism (scientific name)	*Daphnia magna* *Ceriodaphnia dubia*
Life stage (e.g. egg, embryo, ELS, adult)	Neonates <24 hours
Exposure regime (e.g. static, renewal, etc.)	Static
Test method	Toxicity test
Analysis (measured or nominal)	Analysed by GC
Temperature	25 ± 1°C
Hardness	Reconstituted moderately hard water prepared by a (cited) standard procedure
pH	Not reported
Salinity	Freshwater
Exposure duration	48 hours
Endpoint (e.g. NOEC, EC50)	LC50
Effect (e.g. reproduction, survival, growth)	Mortality
Concentration	*Daphnia magna*: 48-hour LC50 1.25 µg l^{-1} *Ceriodaphnia dubia*:48-hour LC50 0.55 µg l^{-1}
Initial quality assessment (e.g. good, moderate, poor)	Good – reference to measured concentrations. LC50s determined on the basis of nominal concentrations by probability analysis or binomial test (where data were not sufficient for probability analysis). Use of US EPA TOXDAT programs.
Comments	

Reference	**35**
CAS number	52645-53-1
Chemical	Permethrin
Chemical species	
Organism (common name)	Yellow fever mosquito
Organism (scientific name)	*Aedes aegypti*
Life stage (e.g. egg, embryo, ELS, adult)	Larvae used for testing were generally 3rd instars 72-hour post-hatch.
Exposure regime (e.g. static, renewal, etc.)	Static
Test method	Static acute toxicity test
Analysis (measured or nominal)	Residues of technical grade permethrin were analysed to determine disappearance. This was not possible for microencapsulated permethrin due to difficulties in separating permethrin in solution from that remaining in capsules.
Temperature	25°C
Hardness	Not stated
pH	7.8–8.0
Salinity	City of Guelph tap water, chlorine removed
Exposure duration	24 hours
Endpoint (e.g. NOEC, EC50)	LC50
Effect (e.g. reproduction, survival, growth)	Mortality
Concentration	Technical grade permethrin: 24-hour LC50 0.45 µg l^{-1} Microencapsulated permethrin: 24-hour LC50 21.6 µg l^{-1}
Initial quality assessment (e.g. good, moderate, poor)	Good
Comments	

Reference	**36**
CAS number	52645-53-1
Chemical	Permethrin
Chemical species	
Organism (common name)	Blackflies Mayflies Caddisflies Stoneflies Dragonflies Crayfish
Organism (scientific name)	*Simulium* sp. *Isonychia* sp. *Pycnopsyche* sp. *Acroneuria* sp. *Ophiogomphus* sp. *Orconectes* sp.
Life stage (e.g. egg, embryo, ELS, adult)	Insect larvae. Life stage of larvae and the crayfish species *Orconectes* not further specified.
Exposure regime (e.g. static, renewal, etc.)	Flow-through with natural creek water. Water velocity in the troughs was 0.18 m s^{-1} at a flow rate of 33 ml s^{-1}.
Test method	
Analysis (measured or nominal)	Nominal
Temperature	8 (nightly low) to 16°C (daily high)
Hardness	60 mg l^{-1}
pH	6.5–7.5
Salinity	Freshwater
Exposure duration	1-hour pulse exposure followed by 47-hour observation period
Endpoint (e.g. NOEC, EC50)	LC50 NOEC
Effect (e.g. reproduction, survival, growth)	LC50: mortality NOEC: drift
Concentration	*Simulium* sp.: LC50 3.8 µg l^{-1} *Isonychia* sp.: LC50 4.4 µg l^{-1} *Pycnopsyche* sp.: LC50 7.0 µg l^{-1} *Acroneuria* sp.: LC50 2.0 µg l^{-1} *Ophiogomphus* sp.: LC50 7.1 µg l^{-1} *Orconectes* sp.: LC50 3.0 µg l^{-1} NOEC drift*:* 0.5 µg l^{-1}
Initial quality assessment (e.g. good, moderate, poor)	Good, but reference to nominal concentration
Comments	

Reference	**37**
CAS number	52645-53-1
Chemical	Permethrin
Chemical species	
Organism (common name)	Freshwater fish: Lahontan cut-throat trout Greenback cut-throat trout Apache trout Rainbow trout Saltwater fish: Leon Springs pupfish) Sheepshead minnow
Organism (scientific name)	Freshwater fish: *Oncorhynchus clarki henshawi* (lahontan cut-throat trout) *Oncorhynchus clarkii stomias* (greenback cut-throat trout) *Oncorhynchus gilae apache* (apache trout) *Oncorhynchus mykiss* (rainbow trout) Saltwater fish: *Cyprinodon bovinus* (Leon Springs pupfish) *Cyprinodon variegatus* (sheepshead minnow)
Life stage (e.g. egg, embryo, ELS, adult)	Juveniles; ca. 0.2–1 g bw
Exposure regime (e.g. static, renewal, etc.)	Static
Test method	Static acute toxicity test
Analysis (measured or nominal)	Stock solutions were analysed by GC as a confirmation of nominal concentrations. The average nominal concentrations was 111% (*n* = 9).
Temperature	12°C (freshwater salmonids); 20°C (saltwater fish)
Hardness	160–180 mg l^{-1} as $CaCO_3$ (freshwater)
pH	Not stated but according to cited guideline
Salinity	Saltwater 2‰; freshwater (reconstituted water: hardness 160–180 mg l^{-1} $CaCO_3$)
Exposure duration	96 hours (with observations of mortality as endpoint at 12, 24, 48, 72 and 96 hours)
Endpoint (e.g. NOEC, EC50)	LC50
Effect (e.g. reproduction, survival, growth)	Mortality
Concentration	Freshwater fish: Lahontan cut-throat trout: 96-hour LC50 1.6 µg l^{-1} Greenback cut-throat trout: 96-hour LC50 <1 µg l^{-1} Apache trout: 96-hour LC50 1.7 µg l^{-1} Rainbow trout: 96-hour LC50 3.3 µg l^{-1} Saltwater fish: Leon Springs pupfish: 96-hour LC50 21 µg l^{-1} Sheepshead minnow: 96-hour LC50 17 µg l^{-1}

Initial quality assessment (e.g. good, moderate, poor)	Good
Comments	

Reference	**38**
CAS number	52645-53-1
Chemical	Permethrin
Chemical species	
Organism (common name)	Opossum shrimp Pink shrimp Sheepshead minnow Atlantic silverside
Organism (scientific name)	*Americamysis (Mysidopsis) bahia* *Pennaeus duorarum* *Cyprinodon variegatus* *Menidia menidia*
Life stage (e.g. egg, embryo, ELS, adult)	*M. bahia*: ≤24-hour old individuals Not stated for other test organisms
Exposure regime (e.g. static, renewal, etc.)	Flow-through
Test method	Flow-through acute toxicity test according to ASTM 1980 guidelines
Analysis (measured or nominal)	Yes, by GC
Temperature	Not reported
Hardness	Not reported
pH	Not reported
Salinity	Filtered sea water
Exposure duration	96 hours
Endpoint (e.g. NOEC, EC50)	LC50
Effect (e.g. reproduction, survival, growth)	Mortality
Concentration	*Americamysis (Mysidopsis) bahia*: 96-hour LC50 0.02 µg l^{-1} *Pennaeus duorarum*: 96-hour LC50 0.22 µg l^{-1} *Cyprinodon variegatus*: 96-hour LC50 7.8 µg l^{-1} *Menidia menidia*: 96-hour LC50 2.2 µg l^{-1}
Initial quality assessment (e.g. good, moderate, poor)	Good
Comments	

Reference	**39**
CAS number	52645-53-1
Chemical	Permethrin
Chemical species	
Organism (common name)	Waterflea Caddisfly Blackfly Mayfly
Organism (scientific name)	*Daphnia magna* *Daphnia pulex* *Hydropsyche* spp. *Simulium vittatum* *Isonychia bicolor*
Life stage (e.g. egg, embryo, ELS, adult)	Daphnia: neonates <48 hours Insects: life stages as collected in the field
Exposure regime (e.g. static, renewal, etc.)	Daphnia: static Insects: recirculating system
Test method	Toxicity test
Analysis (measured or nominal)	Nominal
Temperature	Daphnia: 18.5°C Insects: 14°C
Hardness	Not reported
pH	Daphnia: not reported Insects: 7.8
Salinity	Daphnia: filtered (30 µm) well water Insects: filtered (30 µm) river water
Exposure duration	Short-term toxicity tests: Daphnia: 72 hours (*Daphnia pulex*) or 96 hours (*Daphnia magna*) Insects: 1-hour pulse exposure in flowing water, then provision of clean recirculating water. Observation up to 96 hours post-treatment at 24-hour intervals. Long-term toxicity tests: Exposure of *Daphnia magna* for 40 days and *Daphnia pulex* for 32 days
Endpoint (e.g. NOEC, EC50)	LC50 NOEC
Effect (e.g. reproduction, survival, growth)	Short-term tests: Mortality (an organism was considered dead when no visible signs of movement were apparent in response to agitation) Long-term tests: (with Daphnia): Mortality, time to first brood, number of broods, mean brood size, total young

Concentration	<u>Short-term tests:</u> *Daphnia magna*: 96-hour LC50 0.59–21.81 µg l^{-1} (six individual tests with the microencapsulated permethrin formulation) *Daphnia pulex*: 72-hour LC50: 6.8–22.57 µg l^{-1} (four individual tests with the EC permethrin formulation) *Hydropsyche* spp.: 1-hour LC50 3,560–5,610 µg l^{-1} (four individual tests with the microencapsulated permethrin formulation) *Simulium vittatum*: 1-hour LC50 1,410–3,580 µg l^{-1} (three individual tests with the microencapsulated permethrin formulation) *Isonychia bicolor*: 1-hour LC50 12,810–14,010 µg l^{-1} (two individual tests with the microencapsulated permethrin formulation) <u>Long-term tests:</u> *Daphnia magna*: NOEC 1µg l^{-1} (mortality; LOEC 5 µg l^{-1}, >50% mortality); *Daphnia pulex*: NOEC <1 µg l^{-1} (LOEC 1 µg l^{-1}, this concentration, however, caused >90% mortality and more than 50% diminished brood size compared with the control)
Initial quality assessment (e.g. good, moderate, poor)	Short-term: good Long-term: moderate: static exposure, no analytical verification of exposure concentrations, exposure to microencapsulated toxicant
Comments	

Reference	**40**
CAS number	52645-53-1
Chemical	Permethrin
Chemical species	
Organism (common name)	Fathead minnow Snail
Organism (scientific name)	*Pimephales promelas* *Heliosoma trivolvis*)
Life stage (e.g. egg, embryo, ELS, adult)	Fish: 4–5-day-old larvae Snails: individuals of 0.09–0.3 g weight
Exposure regime (e.g. static, renewal, etc.)	Flow-through
Test method	Fish: early life stage for 28 days Snail: 28-day test
Analysis (measured or nominal)	Measured by GC
Temperature	Fish: 25 ± 2°C Snail: 15 ± 2°C
Hardness	34–48 mg l^{-1} as $CaCO_3$
pH	7.4–7.9
Salinity	Lake Superior water
Exposure duration	Fish: 32 days Snail: 28 days
Endpoint (e.g. NOEC, EC50)	Fish: NOEC
Effect (e.g. reproduction, survival, growth)	Fish: survival, growth, embryo hatchability
Concentration	Permethrin significantly reduced survival and impaired swimming ability at a concentration of 1.4 µg l^{-1}. One day after hatch, survival of larvae at this concentration was reduced to 37%. Most larvae that survived were convulsive. Four days after hatch, only one larva remained alive at 1.4 µg l^{-1}. No significant effects on survival were seen at permethrin concentrations of 0.66 µg l^{-1} or less. Hatchability, normal appearance and growth of embryos were not decreased at any concentration tested after the 32-day test. Snail survival was not significantly decreased up to the highest concentration tested for 28 days (0.33 µg l^{-1}). Snails exposed to the highest concentration responded more slowly when probed than snails exposed to lower concentrations. However, this condition disappeared after the first week of exposure. BCFs in *P. promelas* were 2,800 ± 700 after 32 days exposure and the 28-day BCF in snails was 800 ± 150.
Initial quality assessment (e.g. good, moderate, poor)	Good
Comments	

Reference	**42**
CAS number	52645-53-1
Chemical	Permethrin
Chemical species	
Organism (common name)	Stonefly larvae Mayfly larvae Dolly Varden trout Benthic algae
Organism (scientific name)	*Plecoptera* *Ephemeroptera* *Salvelinus malma* *Periphyton*
Life stage (e.g. egg, embryo, ELS, adult)	Life stage of the insect larvae not further specified; two size classes of fish fry used (2 and 5 cm length)
Exposure regime (e.g. static, renewal, etc.)	Exposure in natural stream
Test method	Field test
Analysis (measured or nominal)	Contamination in stream after spraying of trees located at stream banks monitored by analysis.
Temperature	Natural ambient
Hardness	Natural ambient
pH	Natural ambient
Salinity	Natural ambient
Exposure duration	5 hours post-application 0.05 µg l^{-1}, 8–11 hours post-application 0.14 µg l^{-1}, 14 hours post-application 0.02 µg l^{-1}. At two control sites 800 m upstream and 500 m downstream of the treatment area, no permethrin residues (i.e. <0.01 µg l^{-1}) were found in water.
Endpoint (e.g. NOEC, EC50)	Description of observations upon use of permethrin
Effect (e.g. reproduction, survival, growth)	Drift of stream invertebrates Mortality of actively exposed indigenous stream invertebrates and fish Impact on periphyton
Concentration	Periphyton was not affected by permethrin. Invertebrate drift, however, increased significantly fourfold 3 hours after permethrin use at the treatment site but, within 9 hours, declined to levels observed before spray application. Dipteran (Chironomidae) larvae and trichopteran (Limnophilidae) larvae accounted for the increase in drift. Aquatic invertebrates and fish fry caged in the biomonitors did not exhibit increased mortality because of the permethrin treatment. Only one plecopteran died within the treatment site within 24 hours of treatment and no mortality was observed at sites above or below the treatment area.
Initial quality assessment (e.g. good, moderate, poor)	Good
Comments	

Reference	**43**
CAS number	52645-53-1
Chemical	Permethrin
Chemical species	
Organism (common name)	Natural pelagic community of a pond, in particular: Dinoflagellate algae A predatory insect larvae Pelagic crustacean species A rotifer
Organism (scientific name)	*Ceratium hirundinella* *Chaoborus flavicans*
Life stage (e.g. egg, embryo, ELS, adult)	All (*C. hirundinella*) Pelagic larvae (*C. flavicans*)
Exposure regime (e.g. static, renewal, etc.)	Enclosure (static)
Test method	
Analysis (measured or nominal)	Permethrin concentrations (residues) measured in water and sediment 2–5 days post-application by gas chromatography)
Temperature	23–30°C
Hardness	Not reported
pH	Not reported
Salinity	Natural freshwater pond water
Exposure duration	30 days post first application (2nd application 18 days after 1st)
Endpoint (e.g. NOEC, EC50)	No specific endpoint reported but rather description of changes in community structure
Effect (e.g. reproduction, survival, growth)	Abundance of selected species, photosynthesis, respiration
Concentration	Significant concentration dependent reduction of the dinoflagellate *Ceratium hirundinella*, the cladoceran *Daphnia rosea* and the midge larvae *Chaoborus flavicans*. Upon these changes in community composition, some other crustacean and a rotifer species benefited.
Initial quality assessment (e.g. good, moderate, poor)	Good
Comments	

Reference	**44**
CAS number	52645-53-1
Chemical	Permethrin
Chemical species	
Organism (common name)	Sheepshead minnow
Organism (scientific name)	*Cyprinodon variegatus*
Life stage (e.g. egg, embryo, ELS, adult)	1.5–24-hour-old embryos
Exposure regime (e.g. static, renewal, etc.)	Intermittent flow
Test method	28-day early life stage test
Analysis (measured or nominal)	Yes, at least weekly by GC
Temperature	30 ± 1.5°C
Hardness	Not reported
pH	Not reported
Salinity	Sea water with a salinity ranging from 22–32‰.
Exposure duration	28 days
Endpoint (e.g. NOEC, EC50)	NOEC
Effect (e.g. reproduction, survival, growth)	Embryo development, hatching success and survival and growth of hatched fish
Concentration	28-day NOEC fry survival: 10 µg l^{-1} (measured); fry survival at 10 µg l^{-1}: 99% 28-day LOEC fry survival: 22 µg l^{-1} (measured); fry survival at 22 µg l^{-1}: 1%
Initial quality assessment (e.g. good, moderate, poor)	Good
Comments	

Reference	**62**
CAS number	52645-53-1
Chemical	Permethrin
Chemical species	-
Organism (common name)	Green algae
Organism (scientific name)	*Pseudokirchneriella subcapitata*
Life stage (e.g. egg, embryo, ELS, adult)	Growth phase
Exposure regime (e.g. static, renewal, etc.)	Static
Test method	72 hour growth inhibition test
Analysis (measured or nominal)	Yes, the test concentrations were analysed at the beginning and end of the test (measured values were 52 to 83% of nominal concentrations)
Temperature	$22 \pm 2^{o}C$
Hardness	Not reported
pH	Not reported
Salinity	Nutrient media
Exposure duration	3 days
Endpoint (e.g. NOEC, EC50)	NOEC, EC50
Effect (e.g. reproduction, survival, growth)	Growth inhibition
Concentration	3-day NOEC (growth inhibition): 160 µg l^{-1} 3-day EC50 (growth inhibition): >160 µg l^{-1}
Initial quality assessment (e.g. good, moderate, poor)	Good
Comments	

Reference	**63**
CAS number	52645-53-1
Chemical	Permethrin
Chemical species	
Organism (common name)	Marine diatom
Organism (scientific name)	*Skeletonema costatum*
Life stage (e.g. egg, embryo, ELS, adult)	Growth phase
Exposure regime (e.g. static, renewal, etc.)	Static
Test method	96 hour growth inhibition test
Analysis (measured or nominal)	Not stated
Temperature	$22 \pm 2^{o}C$
Hardness	Not relevant
pH	8.1
Salinity	Sea water with a salinity of 30‰.
Exposure duration	4 days
Endpoint (e.g. NOEC, EC50)	EC50
Effect (e.g. reproduction, survival, growth)	Growth inhibition
Concentration	4-day EC50 (growth inhibition): 68 µg l^{-1}
Initial quality assessment (e.g. good, moderate, poor)	Good
Comments	