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Proposed EQS for Water Framework Directive Annex VIII substances: tetrachloroethane

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The UK Technical Advisory Group (UKTAG) supporting the implementation of the Water Framework Directive (2000/60/EC) is a partnership of UK environmental and conservation agencies. It also includes partners from the Republic of Ireland. This report is the result of research commissioned and funded on behalf of UKTAG by the Scotland & Northern Ireland Forum for Environmental Research (SNIFFER) and the Environment Agency's Science Programme.

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Steve Killeen

Head of Science

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.

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 tetrachloroethane (TCE) using the methodology described in Annex V of the Directive. There is no existing EQS for TCE.

The PNECs described in this report are based on a technical assessment of the available ecotoxicity data for TCE, 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.

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 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

Tetrachloroethane is a water-soluble, volatile chlorinated solvent. It is lost from water through volatilisation, but the rate at which this occurs depends on the local characteristics of the water body, ambient temperature and wind speed.

Tetrachloroethane can be broken down by a number of processes; it hydrolyses in water, particularly at high pH, and can be degraded anaerobically.

Its toxicity to aquatic life arises through non-specific narcosis affecting membrane function. This is likely to occur in a wide range of species.

Availability of data

A moderate number of ecotoxicity studies with TCE are available but these cover only a limited range of taxonomic groups. Studies with freshwater species include both acute and chronic studies; chronic data are represented by studies with algae, crustaceans and fish, and acute data are available for the same taxa plus bacteria.

By comparison, saltwater toxicity data are restricted to acute studies on bacteria, crustaceans, fish and algae.

Derivation of PNECs

Long-term PNEC for freshwaters

The lowest reliable no observed effect concentration (NOEC) of 4,931 $\mu\text{g l}^{-1}$ is for survival of flagfish (*Jordanella floridae*) following a 10-day exposure to TCE. A similar NOEC (6,900 $\mu\text{g l}^{-1}$) was also generated from a reliable study with the water flea, *Daphnia magna*. The available evidence suggests that fish are more sensitive to TCE than algae and crustaceans. Therefore, an assessment factor of 50 applied to the flagfish NOEC of 4,931 $\mu\text{g l}^{-1}$ is recommended following the Annex V guidance, resulting in a PNEC_{freshwater_lt} of 99 $\mu\text{g l}^{-1}$.

Short-term PNEC for freshwaters

Good quality data are available from acute studies with fish, algae and crustaceans, with flagfish again being the most sensitive species of those tested (96-hour LC50 of 18,480 $\mu\text{g l}^{-1}$). The availability of data for this range of taxa permits the use of an assessment factor smaller than the default factor of 100. As a result, a factor of 10 applied to the flagfish 96-hour LC50 is recommended, resulting in a PNEC_{freshwater_st} of 1,848 $\mu\text{g l}^{-1}$.

Long-term PNEC for saltwaters

The absence of reliable chronic saltwater toxicity data means the saltwater PNEC is based on freshwater data. This assumes that freshwater and saltwater species share a similar distribution of sensitivities to TCE. This is considered acceptable since the non-specific mode of action of this substance should not result in systematically greater sensitivity of any particular taxonomic group. However, the greater taxonomic diversity of marine organisms compared with those living in freshwaters introduces greater uncertainty into the prediction of a saltwater PNEC. Together with the paucity of data, these considerations invite a higher safety factor to be applied. Consequently, an assessment factor of 500 applied to the flagfish 10-day NOEC is recommended, resulting in a PNEC_{saltwater_lt} of 10 $\mu\text{g l}^{-1}$.

The large assessment factor recommended by the Annex V methodology reflects the high uncertainty in extrapolating from the limited dataset available. However, some of the uncertainty could be reduced, and hence the size of the assessment factor, if chronic toxicity data were to be generated, e.g. for marine fish, algal and invertebrate species.

Short-term PNEC for saltwaters

Although several studies with marine organisms have been reported, none were of sufficient quality to form the basis of a PNEC. As a result, a saltwater short-term PNEC is based on freshwater data. Again, this assumes that freshwater and saltwater species share a similar distribution of sensitivities to TCE. This assumption is considered acceptable given the non-specific mode of action of this substance. An assessment factor of 100 applied to the flagfish 96-hour LC50 is recommended, reflecting the high level of uncertainty associated with extrapolating from a small freshwater dataset. A PNEC_{saltwater_st} of 184 $\mu\text{g l}^{-1}$ results. Again, some of this uncertainty, and hence the size of the assessment factor, could be reduced if reliable acute toxicity data were to be generated, e.g. for marine fish, algal and invertebrate species.

PNECs for sediment and secondary poisoning

Tetrachloroethane is not sufficiently lipophilic or bioaccumulative to warrant the development of PNECs for sediment and secondary poisoning

Summary of proposed PNECs

Receiving medium/exposure scenario	Proposed PNEC ($\mu\text{g l}^{-1}$)	Existing EQS
Freshwater/long-term	99	No standard
Freshwater/short-term	1,848	No standard
Saltwater/long-term	10	No standard
Saltwater/short-term	184	No standard

Analysis

Current analytical methodologies employing extraction/preconcentration gas chromatography/mass spectrometry (GC-MS) are capable of achieving detection limits as low as 1 ng l^{-1} . This is sufficiently sensitive to analyse TCE in water for compliance purposes.

Implementation issues

Before EQSs can be recommended, the following issues would need to be addressed:

1. A lack of reliable ecotoxicological data introduces a high level of uncertainty into the extrapolation process, encouraging the use of large assessment factors (particularly for the freshwater long-term and saltwater PNECs). This uncertainty would be reduced by undertaking additional ecotoxicity testing, which could lead to a change in the proposed PNECs.
2. Such an approach is recommended but, since there are no existing EQSs, an interim standard is not possible.

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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 environment 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 tetrachloroethane (TCE) using the methodology described in Annex V of the Directive. There is no existing EQS for TCE.

The PNECs described in this report are based on a technical assessment of the available ecotoxicity data for TCE, 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. 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.

This report provides a data sheet for TCE.

1.1 Properties and fate in water

Tetrachloroethane is a water-soluble, volatile chlorinated solvent. It is lost from water through volatilisation, but the rate at which this occurs depends on the local characteristics of the water body, ambient temperature and wind speed.

Tetrachloroethane can be broken down by a number of processes; it hydrolyses in water, particularly at high pH, and can be degraded anaerobically.

Its toxicity to aquatic life arises through non-specific narcosis affecting membrane function. This is likely to occur in a wide range of species.

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.

2. Results and observations

2.1 Identity of substance

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

Table 2.1 Isomers covered by this report

Name	CAS Number
1,1,1,2-Tetrachloroethane	630-20-6
1,1,2,2-Tetrachloroethane	79-34-5

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 [31].

Section 2.6 summarises the effects data identified from the literature for TCE. 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	1,848 $\mu\text{g l}^{-1}$ (see Section 3.1.1)	-	ND
Freshwater long-term	99 $\mu\text{g l}^{-1}$ (see Section 3.1.1)	Lack of data	ND
Saltwater short-term	184 $\mu\text{g l}^{-1}$ (see Section 3.1.2)	-	ND
Saltwater long-term	10 $\mu\text{g l}^{-1}$ (see Section 3.1.2)	Lack of data	ND
Freshwater sediment short-term	NA	-	-
Freshwater sediment long-term	NA	Lack of data	-
Saltwater sediment short-term	NA	-	-
Saltwater sediment long-term	NA	Lack of data	-
Freshwater secondary poisoning	NA	-	-
Saltwater secondary poisoning	NA	-	-

AF = assessment factor

NA = not applicable

ND = no data

SSD = species sensitivity distribution

2.3 Hazard classification

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

Table 2.3 Hazard classification

CAS Number	Substance	R-phrases and labelling
79-34-5	1,1,2,2-tetrachloroethane	T+; R26/27 - N; R51-53 + R26/27: very toxic by inhalation and in contact with skin + R51/53: Toxic to aquatic organisms, may cause long-term adverse effects in the aquatic environment.
630-20-6	1,1,1,2-tetrachloroethane	No data available

2.4 Physical and chemical properties

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

Table 2.4 Physical and chemical properties of tetrachloroethane

Property	1,1,2,2-Tetrachloroethane	1,1,1,2-Tetrachloroethane	Reference
Molecular formula	C ₂ H ₂ Cl ₄	C ₂ H ₂ Cl ₄	
Molecular weight	167.85	167.85	[1]
Composition	C: 14.31; H: 1.20; Cl: 84.49	C: 14.31; H: 1.20; Cl: 84.49	[1]
Appearance	Colourless liquid	Colourless liquid	[2]
Melting point (°C)	-44	-70.2	[1]
Boiling point (°C)	146.5	130.5	[1]
Vapour pressure	5 mmHg at 20°C	12 mmHg at 20°C	[2]
Water solubility (mg l ⁻¹)	2,900 at 20°C	1,070 at 20°C	[2]
Henry's Law constant	2.56 x 10 ⁻⁴ atm-m ³ /mol at 25°C	2.45 x 10 ⁻⁴ atm-m ³ /mol at 25°C	[2]
Dissociation constant (pKa)	-	-	
Octanol–water partition coefficient (log Kow)	2.39	2.93	[2]

2.5 Environmental fate and partitioning

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

Table 2.5 Environmental fate and partitioning of tetrachloroethane

Property	Value	Reference
Abiotic degradation	TCE released into water will primarily be lost by volatilisation in a matter of days to weeks.	[3]
	The volatilisation half-lives from a model river and a model pond have been estimated to be 6.3 hours and 3.5 days, respectively.	[3]
	Under alkaline conditions, TCE may be expected to hydrolyse.	[4]
	TCE undergoes base-catalysed hydrolysis in water at commonly encountered environmental pH values to form trichloroethene. Half-life at 25°C and pH 7.0 calculated at 102 days. Hydrolysis is very pH dependent, with calculated half-lives (based on measured hydrolytic rate constants) of 1, 11 and 111 days for pH 9, 8, and 7, respectively.	[3, 5, 6]
	Empirical half-disappearance times (including abiotic and biotic removal) of 573 days at pH 6.05 and 36 days at pH 7.01 have been reported.	[5]
	In pore water extracted from sediments, a 29.1 day half-life for TCE has been reported at pH 7–7.5.	[6]
	In wastewater treatment plants, air stripping is an important mechanism for transferring the chemical from the water into air.	[6]
Biodegradation	No significant degradation has been found under aerobic conditions where TCE was incubated in sewage seed for 7 days.	[7]
	However, other work has reported a 41% degradation rate in 24 days in an acclimated modified shake flask biodegradation test.	[8]
	A continuous flow biofilm column operating under anaerobic conditions with a sewage inoculum achieved 97% steady state removal during 4 months of operation.	[4]
	A 19% loss was obtained in a 5-day river die-away test using an acclimated system with an initial concentration of 17.3 ppm.	[3]
	Some biodegradation may occur in situations where evaporation is extremely slow and the body of water is rich in microorganisms, such as a eutrophic lake. Biodegradation in groundwater is possible but the biodegradation products of TCE are resistant to further biodegradation.	[4]

Property	Value	Reference
Partition coefficients	A measured Koc of 79 in a silt loam indicates TCE will be highly mobile in soil.	[9]
	Results suggest that 1,1,2,2-tetrachloroethane will not adsorb appreciably to soil, suspended solids or sediment.	[3]
	TCE is not expected to partition from the water column to organic matter contained in sediments and suspended solids.	[4]
	Octanol–water partition coefficients (log Kow) of 2.39 for 1,1,2,2-tetrachloroethane and 2.93 for 1,1,1,2-tetrachloroethane have been reported.	[2]
Bioaccumulation BCF	Bioconcentration factor (BCF) of 8.0 in a 16-day experiment with bluegill sunfish.	[10]
	BCF of 2.0 in fathead minnows.	[11]

Tetrachloroethane is a relatively soluble, volatile chlorinated solvent. One of the most important routes for loss from the aquatic environment is via volatilisation, with an estimated half-life of between a few hours to several days depending on conditions such as wind speed, temperature, surface area of water body, etc. (Table 2.5).

Hydrolysis of TCE is pH dependent and significantly faster under basic conditions with half-lives varying by two orders of magnitude between pH 7 and 9 (Table 2.5).

Anaerobic biodegradation provides an additional degradation pathway for TCE, particularly in groundwater. The anaerobic degradation products include (in decreasing amounts) [4]:

- 1,2-dichloroethene
- 1,1,2-trichloroethene
- trichloroethane
- 1,1-dichloroethene
- vinyl chloride.

The organic carbon–water partition coefficient for TCE is low (reflecting its solubility). Therefore, TCE has only a weak affinity for soil and sediment. The low octanol–water partition coefficient of TCE along with its low bioaccumulation factors in aquatic biota (Table 2.5) indicate that bioaccumulation in aquatic organisms will be minimal.

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.

Freshwater and marine toxicity values were retrieved from the US Environmental Protection Agency (US EPA) ECOTOX database.³

Additional water column toxicity data, sediment toxicity data and mammalian or avian chronic oral toxicity data were sought from ScienceDirect®⁴ and Web of Science®.⁵

In addition, data were also sought from:

- Hazardous Substances Data Bank (HSDB®) database of the US National Library of Medicine [4];
- US EPA Integrated Risk Information System (IRIS) database,⁶
- World Health Organization (WHO) Environmental Health Criteria (EHC) documents.⁷

Approximately 60 acute and chronic effects data were located for 1,1,1,2-tetrachloroethane and 1,1,2,2-tetrachloroethane. However, more than 90 per cent of the data related to the 1,1,2,2-tetrachloroethane isomer.

Effect concentrations ranged from 181–35,000 µg l⁻¹ for 1,1,1,2-tetrachloroethane and from 253–344,000 µg l⁻¹ for 1,1,2,2-tetrachloroethane. Given that the majority of the data on TCE were for the 1,1,2,2-isomer and the effect data ranges for this chemical effectively encompass the data for 1,1,1,2-tetrachloroethane, PNECs based on the 1,1,2,2 isomer will be protective of exposures to 1,1,1,2-tetrachloroethane.

Toxicity data for sediments (on a mg/kg sediment basis) were not available.

2.6.1 Toxicity to freshwater organisms

Short-term (st) toxicity data from single species tests were available for four taxonomic groups, i.e. algae, bacteria, crustaceans and fish. Chronic toxicity data were available for algae, crustaceans and fish. The species range in the freshwater dataset (short and long term) is fairly limited, with only two algal, one bacterial, one crustacean and five fish species being represented. No field data were identified for TCE.

Diagrammatic representations of the available freshwater data (cumulative distribution functions) for TCE 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 derive the TCE PNECs. The lowest critical freshwater data for TCE are presented in Tables 2.6 and 2.7.

³ <http://www.epa.gov/ecotox/>

⁴ <http://www.sciencedirect.com/>

⁵ <http://scientific.thomson.com/products/wos/>

⁶ <http://www.epa.gov/iris/index.html>

⁷ <http://www.inchem.org/pages/ehc.html>

Figure 2.1 Cumulative distribution function of freshwater long-term data ($\mu\text{g l}^{-1}$) for tetrachloroethane

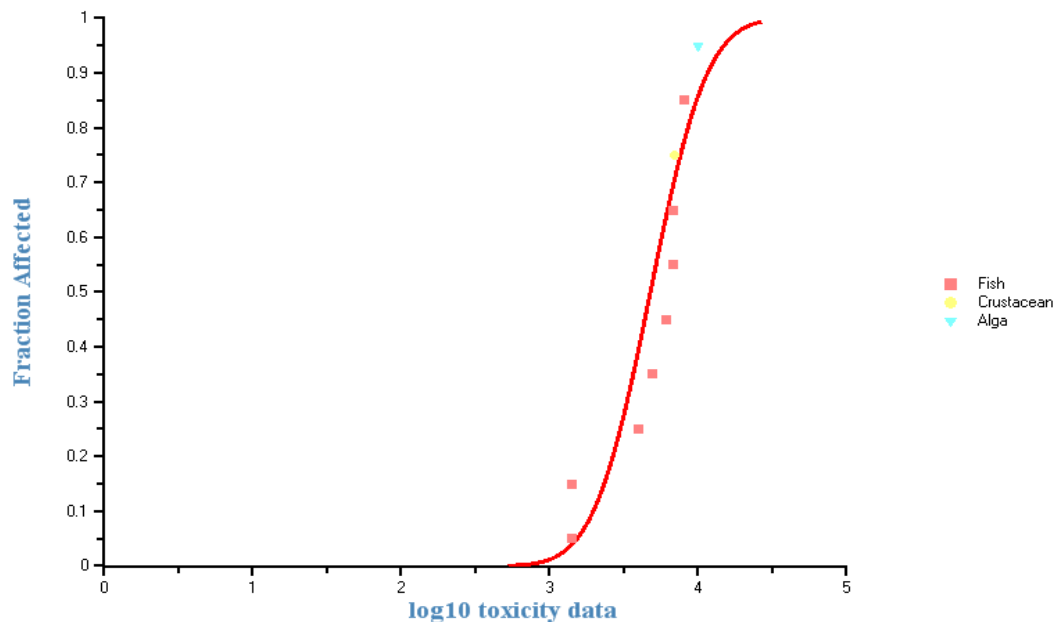


Figure 2.2 Cumulative distribution function of freshwater short-term data ($\mu\text{g l}^{-1}$) for tetrachloroethane

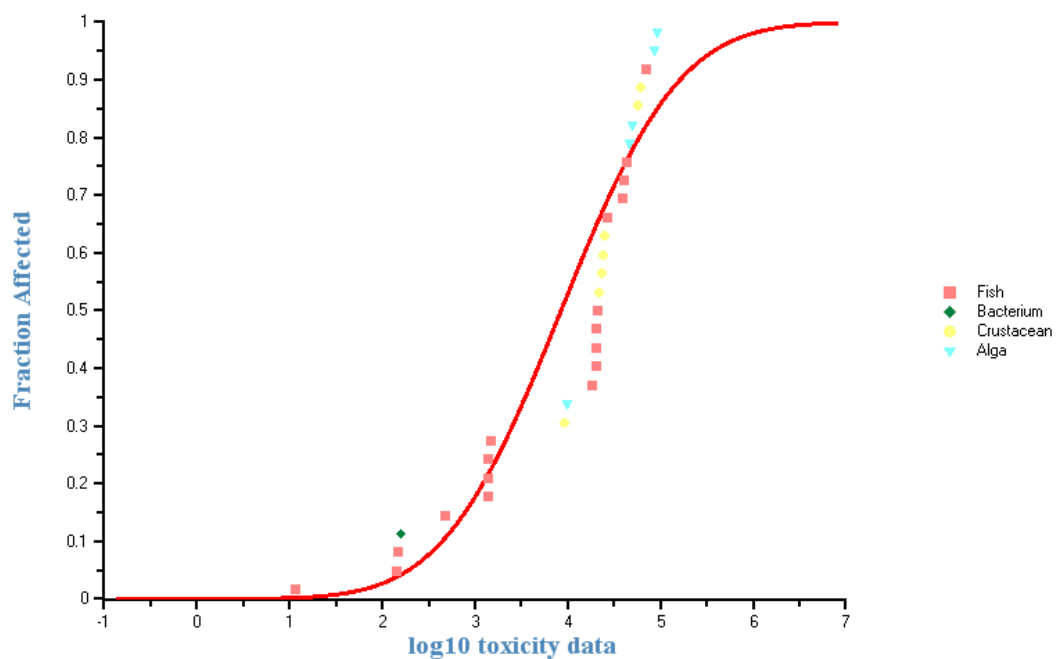


Table 2.6 Most sensitive long-term freshwater toxicity data for organisms exposed to tetrachloroethane

Test substance	Species	Taxonomic group	Endpoint	Effect	Test duration	Conc. ($\mu\text{g l}^{-1}$)	Toxicant analysis ¹	Comments	Reliability index ²	Reference
1,1,2,2-Tetrachloroethane	<i>Selenastrum capricornutum</i>	Algae	NOEC	Chlorophyll content	96 hours	<10,000	n	ND	4	[12]
1,1,2,2-Tetrachloroethane	<i>Daphnia magna</i>	Crustaceans	NOEC	Number of young	28 days	6,900	y	pH 7.3; 20°C; hardness 44.7 mg l ⁻¹ CaCO ₃	1	[13]
1,1,2,2-Tetrachloroethane	<i>Daphnia magna</i>	Crustaceans	LOEC	Number of young	28 days	14,000	y	pH 7.3; 20°C; hardness 44.7 mg l ⁻¹ CaCO ₃	1	[13]
1,1,2,2-Tetrachloroethane	<i>Jordanella floridae</i>	Fish	NOEC	Mortality	10 days	4,931	y	pH 6.9; 25°C; hardness 48 mg l ⁻¹ CaCO ₃	1	[14]
1,1,2,2-Tetrachloroethane	<i>Jordanella floridae</i>	Fish	LOEC	Mortality	10 days	10,597	y	pH 6.9; 25°C; hardness 48 mg l ⁻¹ CaCO ₃	1	[14]
1,1,2,2-Tetrachloroethane	<i>Jordanella floridae</i>	Fish	NOEC	Mortality	28 days	6,147	y	pH 6.9; 25°C; hardness 48 mg l ⁻¹ CaCO ₃	1	[14]
1,1,2,2-Tetrachloroethane	<i>Jordanella floridae</i>	Fish	LOEC	Mortality	28 days	11,663	y	pH 6.9; 25°C; hardness 48 mg l ⁻¹ CaCO ₃	1	[14]
1,1,2,2-Tetrachloroethane	<i>Pimephales promelas</i>	Fish	No effect on growth	Growth	32 days	1,400	y	25°C	2	[15]
1,1,2,2-Tetrachloroethane	<i>Pimephales promelas</i>	Fish	Significant decrease in growth	Growth	32 days	4,000	y	25°C	2	[15]
1,1,2,2-Tetrachloroethane	<i>Oncorhynchus mykiss</i>	Fish	MATC	Growth	32 days	1,400	ND	pH 7.4; 25°C; hardness 45 mg l ⁻¹ CaCO ₃	4	[16]
1,1,2,2-Tetrachloroethane	<i>Oryzias latipes</i>	Fish	No effect	Tumour	3 months	8,000	ND	ND	3	[17]

¹ Toxicant analysis: y = measured; n = nominal.

² See Annex 1.

LOEC = lowest observed effect concentration; MATC = maximum allowable toxicant concentration; NOEC = no observed effect concentration

ND = no data

Table 2.7 Most sensitive short-term freshwater toxicity data for organisms exposed to tetrachloroethane

Test substance	Species	Taxonomic group	Endpoint	Effect	Test duration (hours)	Conc. ($\mu\text{g l}^{-1}$)	Toxicant analysis ¹	Comments	Reliability index ²	Reference
1,1,1,2-Tetrachloroethane	<i>Daphnia magna</i>	Crustaceans	NOEC	Mortality	48	>10,000	n	pH 8; 22°C; hardness 173 mg l ⁻¹ CaCO ₃	3	[18]
1,1,1,2-Tetrachloroethane	<i>Lepomis macrochirus</i>	Fish	LC50	Mortality	96	20,000	n	pH 7.5; 22°C; hardness 33 mg l ⁻¹ CaCO ₃	3	[19]
1,1,2,2-Tetrachloroethane	<i>Nitrosomonas</i>	Bacteria	IC50	Population growth	24	156	n	ND	3	[20]
1,1,2,2-Tetrachloroethane	<i>Scenedesmus subspicatus</i>	Algae	EC50	Population growth	96	47,000	y	ND	2	[21]
1,1,2,2-Tetrachloroethane	<i>Daphnia magna</i>	Crustaceans	NOEC	Mortality	48	<1,700	n	pH 8; 22°C; hardness 173 mg l ⁻¹ CaCO ₃	3	[18]
1,1,2,2-Tetrachloroethane	<i>Daphnia magna</i>	Crustaceans	LC50	Mortality	48	6,800 (lowest of 50 repeats) (9,300 mean of 50 repeats)	n	pH 8; 22°C; hardness 173 mg l ⁻¹ CaCO ₃	3	[18]
1,1,2,2-Tetrachloroethane	<i>Daphnia magna</i>	Crustaceans	EC50	Immobilisation	48	23,000	y	pH 7.3; 20°C; hardness 44.7 mg l ⁻¹ CaCO ₃	1	[13]
1,1,2,2-Tetrachloroethane	<i>Jordanella floridae</i>	Fish	LC50	Mortality	72	18,480	y	28°C	1	[14]
1,1,2,2-Tetrachloroethane	<i>Jordanella floridae</i>	Fish	LC50	Mortality	96	18,480	y	28°C	1	[14]
1,1,2,2-Tetrachloroethane	<i>Pimephales promelas</i>	Fish	LC50	Mortality	96	20,300	y	pH 7.3; 25.6°C; hardness 45 mg l ⁻¹ CaCO ₃	NQA	[23]
TCE	<i>Oryzias latipes</i>	Fish	LC50	Mortality	48	39,000	n	30°C	NQA	[22]

¹Toxicant analysis: y = measured; n = nominal. ² See Annex 1.

NOEC = no observed effect concentration; EC50 = concentration effective against 50% of the organisms tested

LC50 = concentration lethal to 50% of the organisms tested; IC50 = concentration at which the population effect of the organisms tested is inhibited by 50%

ND = no data; NQA = not quality assessed as lower values available

2.6.2 Toxicity to saltwater organisms

Short-term saltwater toxicity data from single species tests were available for four taxonomic groups, i.e. algae, crustaceans, fish and bacteria. No chronic saltwater toxicity data were identified. The species range and number of data points in the saltwater dataset were very limited, with only one algal, one bacterial, two crustacean and one fish species being represented. Data from field studies with marine organisms were also not available.

A diagrammatic representation of the available saltwater data (cumulative distribution functions) for TCE is presented in Figure 2.3. This diagram includes all data regardless of quality and provides an overview of the spread of the available data. These diagrams are not species sensitivity distributions and have not been used to derive the TCE PNECs. The lowest critical saltwater data for TCE are presented in Table 2.8.

An analysis of the relative sensitivity of the species tested is not possible due to the limited size of the dataset.

Figure 2.3 Cumulative distribution function of saltwater short-term data ($\mu\text{g l}^{-1}$) for tetrachloroethane

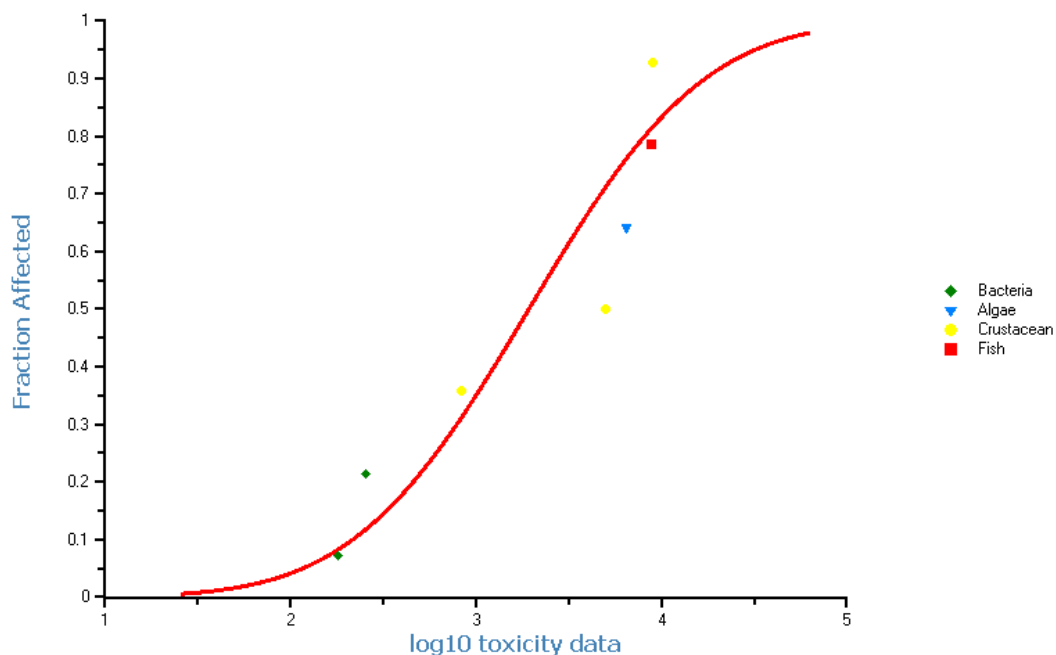


Table 2.8 Most sensitive short-term saltwater toxicity data for organisms exposed to tetrachloroethane

Test substance	Species	Taxonomic group	Endpoint	Effect	Test duration	Conc. ($\mu\text{g l}^{-1}$)	Toxicant analysis ¹	Comments	Reliability index ²	Reference
1,1,1,2-Tetrachloroethane	<i>Vibrio fischeri</i>	Bacteria	IC50	Population response	5 min	181	n	ND	3	[20]
1,1,2,2-Tetrachloroethane	<i>Vibrio fischeri</i>	Bacteria	IC50	Population response	5 min	253	n	ND	3	[20]
1,1,2,2-Tetrachloroethane	<i>Skeletonema costatum</i>	Algae	EC50	Physiology	96 hours	6,440	n	ND	4	[12]
1,1,2,2-Tetrachloroethane	<i>Americamysis bahia</i>	Crustaceans	LC50	Mortality	96 hours	9,020	n	ND	4	[12]
1,1,2,2-Tetrachloroethane	<i>Artemia salina</i>	Crustaceans	LC50	Mortality	24 hours	839	n	pH 8.6; 25°C; salinity 35‰	3	[24]
1,1,2,2-Tetrachloroethane	<i>Artemia salina</i>	Crustaceans	LC50	Mortality	24 hours	5,035	n	pH 8.6; 25°C; salinity 35‰	3	[24]
1,1,2,2-Tetrachloroethane	<i>Cyprinodon variegatus</i>	Fish	NOEC	Mortality	96 hours	8,800	n	25°C; salinity 20‰	3	[25]

¹ Toxicant analysis: y = measured; n = nominal.

² See Annex 1.

NOEC = no observed effect concentration

EC50 = concentration effective against 50% of the organisms tested

LC50 = concentration lethal to 50% of the organisms tested

IC50 = concentration at which the population effect of the organisms tested is inhibited by 50%

ND = no data

2.6.3 Toxicity to sediment-dwelling organisms

Toxicity data for TCE concentrations in sediment (on, for example, a mg/kg sediment basis) were not found.

2.6.4 Endocrine-disrupting effects

No information was found to suggest that TCE has endocrine-disrupting effects.

2.6.5 Mode of action of tetrachloroethane

Tetrachloroethane is a non-specific narcotic that acts by arresting the activity of protoplasmic structures [26]. Neutral organic chemicals such as TCE partition into the lipid portion of biological membranes, swelling the membrane and disrupting cellular structure and function. This results in narcosis-lethargy, unconsciousness and, in extreme cases, death [26].

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

Chronic toxicity data were available for three taxonomic groups, i.e. algae, crustaceans and fish.

Algae appear to be the least sensitive taxonomic group to the effects of TCE. A 96-hour no observed effect concentration (NOEC) (chlorophyll content) of $<10 \text{ mg l}^{-1}$ was reported in *Selenastrum capricornutum* [12]. This was the highest tested concentration. However, this value was regarded as unreliable due to a lack of experimental information and measured effect concentrations. No other long-term algal data could be located.

The lowest chronic data point for crustaceans was a 28-day NOEC (number of young) of 6.9 mg l^{-1} in *Daphnia magna* exposed to 1,1,2,2-tetrachloroethane [13]. The corresponding lowest observed effect concentration (LOEC) was 14 mg l^{-1} . This was a semi-static test with measured concentrations performed to a standard methodology. Consequently, it was regarded as reliable and valid for PNEC derivation. No other chronic data points were available for crustaceans.

The US EPA ECOTOX database reported a 32-day maximum allowable toxicant concentration (MATC) of 1.4 mg l^{-1} in rainbow trout (*Oncorhynchus mykiss*) exposed to 1,1,2,2-tetrachloroethane. However, when the original report was obtained, it did not contain data for rainbow trout [16]. As the reliability of the MATC is not assignable, this data point is excluded from the PNEC derivation.

The same study did report a significant decrease in growth in the fathead minnow (*Pimephales promelas*) following 32-day exposure to 4 mg l^{-1} of 1,1,2,2-tetrachloroethane. The lowest exposure concentration with no significant effect on growth was reported as 1.4 mg l^{-1} , which can be regarded as a NOEC [15]. Based on these data a MATC of $2,366 \mu\text{g l}^{-1}$ (geometric mean of the NOEC and LOEC) can be calculated. In the same study, a significant effect on survival (80 per cent mortality) was seen at 13.7 mg l^{-1} . These fish data are regarded as valid with restriction because, although the values are based on measured data, the report provided only limited information (a single page summary) on the test methodology [15].

The next lowest long-term test result is a 10-day NOEC of 4,931 $\mu\text{g l}^{-1}$ (corresponding LOEC of 10,597 $\mu\text{g l}^{-1}$) for mortality in flagfish (*Jordanella floridae*) [14]. In the same study, a higher NOEC (mortality) of 6,147 $\mu\text{g l}^{-1}$ (LOEC 11,663 $\mu\text{g l}^{-1}$) was reported in the same species after a 28-day exposure period. These data were generated in flow-through studies with measured exposure concentrations. The 10-day study was well documented and so is valid for use in PNEC derivation. In the 28-day study control, however, survival was only 65 per cent. Therefore, this study should be treated with caution and is used only to support the 10-day study.

No other chronic data for fish were available.

Good quality NOEC data were available for crustaceans and fish. The lowest reliable study was the 10-day NOEC of 4,931 $\mu\text{g l}^{-1}$ (corresponding LOEC of 10,597 $\mu\text{g l}^{-1}$) for mortality in flagfish (*Jordanella floridae*) [14]. This study is supported by additional NOECs for fathead minnow and *Daphnia magna*. In addition, fish also appear to be the most sensitive taxa to short-term exposures of TCE. Therefore, in line with the guidance in the TGD [31], use of an assessment factor of 50 is appropriate resulting in a PNEC of 98.6 $\mu\text{g l}^{-1}$.

$$\text{PNEC}_{\text{freshwater_It}} = 4931 \mu\text{g l}^{-1}/\text{AF (50)} = 99 \mu\text{g l}^{-1} \text{ TCE}$$

PNEC accounting for transient concentration peaks

Short-term toxicity data were available for four taxonomic groups, i.e. bacteria, crustaceans, fish and algae.

The lowest short-term bacterial value available for TCE was a 24-hour IC50 of 156 $\mu\text{g l}^{-1}$ for *Nitrosomonas* [20]. However, there was no chemical analysis in this study and few data were available to assess its quality. Consequently, this study was deemed unreliable.

Only limited algae data were available with a reported 96-hour EC50 (population growth) of 47 mg l^{-1} in *Scenedesmus subspicatus* [21]. The *S. subspicatus* study was carried out in closed vessels and concentrations were determined analytically. It is therefore considered as valid. No other short-term algal data could be located.

The lowest reported short-term toxicity data for crustaceans was a 48-hour NOEC (mortality) of <1.7 mg l^{-1} and a 48-hour LC50 of 6.8 mg l^{-1} in *Daphnia magna* [18]. However, this study was conducted using open exposure vessels and the exposure concentrations were not analytically determined. Consequently, these data were not suitable for the PNEC assessment. The same study also reported a 48-hour NOEC (mortality) of <10 mg l^{-1} for *Daphnia* exposed to 1,1,1,2-tetrachloroethane, but this value was also excluded for the same reasons.

The next lowest reported toxicity data point for crustaceans, which is valid for the PNEC assessment, is a 48-hour EC50 of 23 mg l^{-1} for *Daphnia magna* [13]. This was a well-documented study with data generated in sealed vessels and measured exposure concentrations.

Figure 2.2 shows that there are a number of endpoints for fish at very low TCE concentrations [27–29]. All of these endpoints relate to uptake studies with TCE and are

reported as physiology endpoints. However, there were no adverse effects in these studies and all of the data are non-significant up to the highest concentrations tested. Consequently, these values have been excluded from the PNEC derivation.

The lowest acute data point for fish was a 96-hour LC50 for 1,1,2,2-tetrachloroethane of 18,480 $\mu\text{g l}^{-1}$ in the flagfish (*Jordanella floridae*) [14]. This test was carried out under flow-through conditions with measured exposure concentrations. Consequently it was regarded as the lowest reliable acute toxicity value.

In summary, good quality short-term data were available for algae, crustaceans and fish. Therefore, the 'base set' of data required by the TGD [31] is met. As with the chronic dataset, fish are the most sensitive freshwater organisms. Consequently, a PNEC based on the lowest fish LC50 of 18,480 $\mu\text{g l}^{-1}$ in the flagfish (*Jordanella floridae*) [14] is likely to be protective of freshwater organisms. Therefore, it is justifiable to use a lower assessment factor of 10 (instead of the standard factor of 100) resulting in the following PNEC:

$$\text{PNEC}_{\text{freshwater_st}} = 18,480 \mu\text{g l}^{-1}/\text{AF (10)} = 1,848 \mu\text{g l}^{-1} \text{ TCE}$$

PNEC based on outdoor simulated ecosystem studies

No data from outdoor simulated ecosystems were found for TCE.

3.1.2 PNECs for saltwaters

Only limited data were available for saltwater species (two short-term data points for bacteria, three for crustaceans and one for fish and algae). The available data do not allow firm conclusions to be drawn on the comparative sensitivity of the marine species to freshwater species. However, visual comparison of the data points available indicates that the sensitivity of marine species does not differ obviously from the range of values reported for freshwater species. This assumption is supported by the non-specific mode of action of TCE. As a result of this and in accordance with recommendations in the TGD [31], the freshwater data have been used in conjunction with the saltwater dataset.

PNEC accounting for the annual average concentration

No long-term (lt) effects data were available for saltwater organisms. Consequently, the saltwater PNEC has been based on freshwater data.

Good quality NOEC data were available for crustaceans and fish. The lowest reliable study was the 10-day NOEC of 4,931 $\mu\text{g l}^{-1}$ (corresponding LOEC of 10,597 $\mu\text{g l}^{-1}$) for mortality in flagfish (*Jordanella floridae*) [14]. This study is supported by additional NOECs for fathead minnow and *Daphnia magna*. In addition, fish also appear to be the most sensitive taxa to short-term exposures of TCE. Therefore, in accordance with the guidance in the TGD [31], an assessment factor of 500 is appropriate, due to a lack of data for additional marine taxonomic groups, resulting in the following PNEC:

$$\text{PNEC}_{\text{saltwater_lt}} = 4,931 \mu\text{g l}^{-1}/\text{AF (500)} = 10 \mu\text{g l}^{-1} \text{ TCE}$$

PNEC accounting for transient concentration peaks

Limited bacterial data were available obtained using the rapid screening toxicity test Microtox®. Five-minute IC50 values of 181 and 253 $\mu\text{g l}^{-1}$ for 1,1,1,2-tetrachloroethane and 1,1,2,2-tetrachloroethane, respectively, have been reported for this system [20].

However, the values were generated in open vessels without chemical analysis. In addition, the TGD [31] regards Microtox data as unsuitable for PNEC derivation. Consequently, these data have been discarded from the analysis.

Only one marine algal value was available for TCE, with a 96-hour EC50 (physiology) of 6,440 µg l⁻¹ for *Skeletonema costatum* exposed to 1,1,2,2-tetrachloroethane [12]. The original report contained few experimental details and the results were based on nominal concentrations. Consequently, this study was considered unreliable and excluded from the analysis.

The lowest available data point for marine crustaceans was a 24-hour LC50 of 839 µg l⁻¹ for *Artemia salina* exposed to 1,1,2,2-tetrachloroethane [24]. However, the data were generated in open vessels and were based on nominal concentrations. Consequently, this data point was excluded from the analysis. For the same reason the higher LC50 of 5,035 µg l⁻¹, generated in the same study, was also considered unreliable.

The only other data point for crustaceans was a 96-hour LC50 of 9,020 µg l⁻¹ for *Americamysis bahia* exposed to 1,1,2,2-tetrachloroethane [12]. However, the original report contained few experimental details and the results were based on nominal concentrations. Consequently, this study could not be assessed for reliability.

The lowest acute data point for fish was a 96-hour NOEC (mortality) of 8,800 µg l⁻¹ for the sheepshead minnow (*Cyprinodon variegates*) [25]. This value was generated in a static system using nominal concentrations and with no mention of sealed containers to prevent volatilisation. Consequently, this study was also considered unreliable and excluded from the PNEC assessment.

Since all the available short-term data for saltwater organisms were found to be of unacceptable quality, the saltwater PNEC derivation was based on the freshwater data. The lowest reliable short-term data point is a 96-hour LC50 of 18,480 µg l⁻¹ for flagfish (see Section 3.1.1). The TGD [31] does not provide specific guidance for assessment of acute effects of intermittent releases to marine water bodies. Therefore, the saltwater PNEC derivation for short-term exposure to TCE was based on the general guidance given in the TGD on the effects assessment for intermittent releases and derived using the highest short-term assessment factor of 100, resulting in the following PNEC:

$$\text{PNEC}_{\text{saltwater_st}} = 18,480 \mu\text{g l}^{-1}/\text{AF (100)} = 184 \mu\text{g l}^{-1} \text{ TCE}$$

The assessment factor for saltwater intermittent releases was not reduced to 10, as in freshwaters, due to a lack of data for additional marine taxonomic groups.

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

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

3.3 Derivation of existing EQSs

There are no current standards derived for TCE.

3.4 Derivation of PNECs for sediment

Based on the log Kow values for TCE of 2.39–2.93 [2], the TGD criterion [31] for setting sediment standards is not met. Consequently, there is no requirement to derive a quality standard for sediments.

3.5 Derivation of PNECs for secondary poisoning of predators

3.5.1 Mammalian and avian toxicity data

The TGD [31] requires the use of mammalian/avian no-effect data for the assessment of secondary poisoning. Such data are widely used by regulatory bodies such as the WHO to set standards for the protection of human health.

In setting such standards, the available mammalian no observed adverse effect levels (NOAELs) undergo a strict quality assessment, with the lowest high quality data point being used to set the standard. Consequently, a NOAEL used to set a tolerable daily intake (TDI) value or reference dose should be of suitable quality for use in the assessment of secondary poisoning. A summary of the no-effect values used by international organisations to set human health standards for TCE is given in Table 3.1.

Table 3.1 Mammalian oral toxicity data relevant for the assessment of non-compartment specific secondary poisoning

Chemical	End-point	Value (mg/kg bw/day)	Species	Duration	Effect	Regulatory body*
1,1,2,2-tetrachloroethane	NOAEL	56	Rat	?	Reduced body weight	ATSDR
1,1,2,2-tetrachloroethane	LOAEL	43	Female rat	78 weeks	Respiratory effect	ATSDR
1,1,1,2-tetrachloroethane	LOAEL	89.3	Rat	103 weeks	Reduced survival, heat stress, kidney and liver toxicity	US EPA

* Regulatory body using the respective NOAEL to set a human health standard.

ATSDR = Agency for Toxic Substances and Disease Registry

bw = body weight

LOAEL = lowest observable adverse effect level

3.5.2 PNECs for secondary poisoning of predators

The available BCF data for TCE (see Section 2.5) are below the TGD trigger value of 100. In addition, the log Kow values for TCE (see Section 2.5) are also below the TGD criterion for setting secondary poisoning standards. Consequently, there is no requirement to derive a quality standard for the protection of top predators.

4. Analysis and monitoring

Analysis of TCE is routinely performed using purge and trap methods followed by gas chromatography (GC) with mass spectrometry (MS), flame ionisation, electron capture, electrolytic conductivity or microcoulometric detection.

These methods are generally used for water as well as sediment, soil or other solid samples.

Reported detection limits range from 0.001–5 $\mu\text{g l}^{-1}$ for water and from 1–5 $\mu\text{g/kg}$ for soil and sediment samples [3]. Detection limits of 0.01 $\mu\text{g l}^{-1}$ have been reported for solid-phase microextraction coupled with gas chromatography/ion trap mass spectrometry analysis for water [30]. Gas chromatography, often in combination with mass spectrometry, is commonly used for quantifying TCE in biological samples, with detection limits of 400 $\mu\text{g/kg}$ in tissues and 5–500 ng l^{-1} in blood [3].

Proposed PNECs derived for TCE range from 10 $\mu\text{g l}^{-1}$ for long-term protection of pelagic communities to 1,848 $\mu\text{g l}^{-1}$ for short-term protection of freshwater communities. 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. Using this criterion, it is evident that analytical methodologies employing extraction/preconcentration GC-MS are capable of achieving detection limits as low as 1 ng l^{-1} . This suggests that current analytical methods offer adequate performance to analyse TCE in water for compliance purposes.

5. Conclusions

5.1 Availability of data

A moderate number of ecotoxicity studies with TCE are available but these cover only a limited range of taxonomic groups. Studies with freshwater species include both acute and chronic studies; chronic data are represented by studies with algae, crustaceans and fish, and acute data are available for the same taxa plus bacteria.

By comparison, saltwater toxicity data are restricted to acute studies on bacteria, crustaceans, fish and algae.

5.2 Derivation of PNECs

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

5.2.1 Long-term PNEC for freshwaters

The lowest reliable NOEC of $4,931 \mu\text{g l}^{-1}$ is for survival of flagfish (*Jordanella floridae*) following a 10-day exposure to TCE. A similar NOEC ($6,900 \mu\text{g l}^{-1}$) was also generated from a reliable study with the water flea, *Daphnia magna*. The available evidence suggests that fish are more sensitive to TCE than algae and crustaceans. Therefore, an assessment factor of 50 applied to the flagfish NOEC of $4,931 \mu\text{g l}^{-1}$ is recommended following the Annex V guidance, resulting in a $\text{PNEC}_{\text{freshwater_lt}}$ of $99 \mu\text{g l}^{-1}$.

5.2.2 Short-term PNEC for freshwaters

Good quality data are available from acute studies with fish, algae and crustaceans, with flagfish again being the most sensitive species of those tested (96-hour LC50 of $18,480 \mu\text{g l}^{-1}$). The availability of data for this range of taxa permits the use of an assessment factor smaller than the default factor of 100. As a result, a factor of 10 applied to the flagfish 96-hour LC50 is recommended, resulting in a $\text{PNEC}_{\text{freshwater_st}}$ of $1,848 \mu\text{g l}^{-1}$.

5.2.3 Long-term PNEC for saltwaters

The absence of reliable chronic saltwater toxicity data means the saltwater PNEC is based on freshwater data. This assumes that freshwater and saltwater species share a similar distribution of sensitivities to TCE. This is considered acceptable since the non-specific mode of action of this substance should not result in systematically greater sensitivity of any particular taxonomic group. However, the greater taxonomic diversity of marine organisms compared with those living in freshwaters introduces greater uncertainty into the prediction of a saltwater PNEC. Together with the paucity of data, these considerations invite a higher safety factor to be applied. Consequently, an assessment factor of 500 applied to the flagfish 10-day NOEC is recommended, resulting in a $\text{PNEC}_{\text{saltwater_lt}}$ of $10 \mu\text{g l}^{-1}$.

The large assessment factor recommended by the Annex V methodology reflects the high uncertainty in extrapolating from the limited dataset available. However, some of the

uncertainty could be reduced, and hence the size of the assessment factor, if chronic toxicity data were to be generated, e.g. for marine fish, algal and invertebrate species.

5.2.4 Short-term PNEC for saltwaters

Although several studies with marine organisms have been reported, none were of sufficient quality to form the basis of a PNEC. As a result, a saltwater short-term PNEC is based on freshwater data. Again, this assumes that freshwater and saltwater species share a similar distribution of sensitivities to TCE. This assumption is considered acceptable given the non-specific mode of action of this substance. An assessment factor of 100 applied to the flagfish 96-hour LC50 is recommended, reflecting the high level of uncertainty associated with extrapolating from a small freshwater dataset. A $PNEC_{\text{saltwater_st}}$ of $184 \mu\text{g l}^{-1}$ results. Again, some of this uncertainty, and hence the size of the assessment factor, could be reduced if reliable acute toxicity data were to be generated, e.g. for marine fish, algal and invertebrate species.

5.2.5 PNECs for sediment and secondary poisoning

Tetrachloroethane is not sufficiently lipophilic or bioaccumulative to warrant the development of PNECs for sediment and secondary poisoning

Table 5.1 Summary of proposed PNECs

Receiving medium/exposure scenario	Proposed PNEC ($\mu\text{g l}^{-1}$)	Existing EQS
Freshwater/long-term	99	No standard
Freshwater/short-term	1,848	No standard
Saltwater/long-term	10	No standard
Saltwater/short-term	184	No standard

5.3 Analysis

Current analytical methodologies employing extraction/preconcentration GC-MS are capable of achieving detection limits as low as 1 ng l^{-1} . This is sufficiently sensitive to analyse TCE in water for compliance purposes.

5.4 Implementation issues

Before EQSs can be recommended, the following issues would need to be addressed:

1. A lack of reliable ecotoxicological data introduces a high level of uncertainty into the extrapolation process, encouraging the use of large assessment factors (particularly for the freshwater long-term and saltwater PNECs). This uncertainty would be reduced by undertaking additional ecotoxicity testing, which could lead to a change in the proposed PNECs.
2. Such an approach is recommended but, since there are no existing EQSs, an interim standard is not possible.

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List of abbreviations

AA	annual average
AF	assessment factor
ASTM	American Society for Testing and Materials
ATSDR	Agency for Toxic Substances and Disease Registry
BCF	bioconcentration factor
bw	body weight
CAS	Chemical Abstracts Service
DO	dissolved oxygen
EC50	concentration effective against 50% of the organisms tested
ECB	European Chemicals Bureau
EHC	Environmental Health Criteria
EQS	Environmental Quality Standard
GC	gas chromatography
GC-MS	gas chromatography/mass spectrometry
GLP	Good Laboratory Practice (OECD)
HSDB	Hazardous Substances Data Bank
IC50	concentration at which the population effect of the organisms tested is inhibited by 50%
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
MATC	maximum allowable toxicant concentration
MS	mass spectrometry
NA	not applicable
ND	no data
NOAEL	no observed adverse effect level
NOEC	no observed effect concentration
NQA	not quality assessed as lower values available
OECD	Organisation for Economic Co-operation and Development
PNEC	predicted no-effect concentration

SEPA	Scottish Environment Protection Agency
SNIFFER	Scotland & Northern Ireland Forum for Environmental Research
SSD	species sensitivity distribution
st	short term
TCE	tetrachloroethane
TDI	tolerable daily intake
TGD	Technical Guidance Document
UKTAG	UK Technical Advisory Group
US EPA	US Environmental Protection Agency
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	12
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Information on the test species	
Test species used	<i>Americamysis bahia</i> , <i>Skeletonema costatum</i> and <i>Selenastrum capricornutum</i>
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 (expressed as 1,1,2,2-tetrachloroethane)
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	Nominal
Measurement of water quality parameters	Not stated
Test validity criteria satisfied	Not stated
Water quality criteria satisfied	Not stated
Endpoint comment	Very few details available in the original paper. The report states that only the results had to be published and that a final report was not required for the research.
Study conducted to GLP	Not stated

Reliability of study	Unknown
Relevance of study	Unknown
Klimisch Code	4

Reference number	13 (acute)
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Information on the test species	
Test species used	<i>Daphnia magna</i>
Source of the test organisms	Lab culture
Holding conditions prior to test	Held in filtered lake water at 20°C.
Life stage of the test species used	<24 hour daphnids

Information on the test design	
Methodology used	ASTM 1980*
Form of the test substance	1,1,2,2,-tetrachloroethane
Source of the test substance	Aldrich 95–99% pure
Type and source of the exposure medium	Filtered lake water
Test concentrations used	Six concentrations (0.036–0.1 mg l ⁻¹)
Number of replicates per concentration	4
Number of organisms per replicate	5
Nature of test system (static, semi-static or flow-through, duration, feeding)	Static (sealed vessels)
Measurement of exposure concentrations	Measured
Measurement of water quality parameters	pH, dissolved oxygen (DO) and temperature
Test validity criteria satisfied	Yes
Water quality criteria satisfied	Yes
Endpoint comment	Valid endpoint
Study conducted to GLP	Not stated

Reliability of study	Reliable
Relevance of study	Relevant
Klimisch Code	1

* American Society for Testing and Materials (ASTM), 1980 *Standard practise for conducting static acute toxicity tests with fishes, macroinvertebrates and amphibians*. Standard E 729-80. West Conshohocken, PA: ASTM.

Reference number	13 (chronic)
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Information on the test species	
Test species used	<i>Daphnia magna</i>
Source of the test organisms	Lab culture
Holding conditions prior to test	Held in filtered lake water at 20°C.
Life stage of the test species used	<24 hour daphnids

Information on the test design	
Methodology used	ASTM 1978* (proposed method) with modification to prevent volatile losses.
Form of the test substance	1,1,2,2,-tetrachloroethane
Source of the test substance	Aldrich 95–99% pure
Type and source of the exposure medium	Filtered lake water
Test concentrations used	Six concentrations
Number of replicates per concentration	7–10
Number of organisms per replicate	1
Nature of test system (static, semi-static or flow-through, duration, feeding)	Semi-static (three times per week)
Measurement of exposure concentrations	Measured
Measurement of water quality parameters	pH, DO and temperature
Test validity criteria satisfied	Yes
Water quality criteria satisfied	Yes
Endpoint comment	Valid endpoint. NOEC taken as highest concentration not significantly different from controls. LOEC was significantly different.
Study conducted to GLP	Not stated

Reliability of study	Reliable
Relevance of study	Relevant
Klimisch Code	1

* American Society for Testing and Materials (ASTM), 1978 *Proposed standard practice for conducting renewal life cycle toxicity tests with the Daphnid Daphnia magna*. Draft No. 4 (ed. R Comotto). Philadelphia, PA: ASTM.

Reference number	14 (acute)
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Information on the test species	
Test species used	<i>Jordanella floridae</i>
Source of the test organisms	Laboratory stocks
Holding conditions prior to test	Not stated
Life stage of the test species used	Juveniles

Information on the test design	
Methodology used	Followed standard US EPA methodology.
Form of the test substance	1,1,2,2-tetrachloroethane
Source of the test substance	Not stated
Type and source of the exposure medium	Dechlorinated tap water
Test concentrations used	5–6 concentrations, using acetone as a carrier
Number of replicates per concentration	Tests run in duplicate
Number of organisms per replicate	5
Nature of test system (static, semi-static or flow-through, duration, feeding)	Semi-static (24 hours duration) and flow-through
Measurement of exposure concentrations	Semi-static nominal and flow-through measured (three times during test)
Measurement of water quality parameters	pH, DO and temperature
Test validity criteria satisfied	Not stated
Water quality criteria satisfied	Not stated
Endpoint comment	Flow-through study is valid.
Study conducted to GLP	Not stated

Reliability of study	Reliable
Relevance of study	Relevant
Klimisch Code	1

Reference number	14 (chronic)
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Information on the test species	
Test species used	<i>Jordanella floridae</i>
Source of the test organisms	Laboratory stocks
Holding conditions prior to test	Not stated
Life stage of the test species used	Embryo larvae and 2-week-old fry

Information on the test design	
Methodology used	Followed standard US EPA methodology.
Form of the test substance	1,1,2,2-tetrachloroethane
Source of the test substance	Not stated
Type and source of the exposure medium	Dechlorinated tap water
Test concentrations used	5–6 concentrations, using acetone as carrier
Number of replicates per concentration	Tests run in duplicate
Number of organisms per replicate	25 (eggs), 25 (fry)
Nature of test system (static, semi-static or flow-through, duration, feeding)	Flow-through
Measurement of exposure concentrations	Measured (five days a week)
Measurement of water quality parameters	pH, DO and temperature
Test validity criteria satisfied	Control survival only 65% in 28-day study but 100% in 10-day study
Water quality criteria satisfied	Not stated
Endpoint comment	10-day study appears valid. Control survival only 65% in 28-day study.
Study conducted to GLP	Not stated

Reliability of study	Reliable
Relevance of study	Relevant
Klimisch Code	1

Reference number	15
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Information on the test species	
Test species used	<i>Pimephales promelas</i>
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 (expressed as 1,1,2,2-tetrachloroethane)
Source of the test substance	Not stated
Type and source of the exposure medium	Lake water
Test concentrations used	5 (1.4–28.4 µg l ⁻¹)
Number of replicates per concentration	2
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	Measured
Measurement of water quality parameters	Temperature
Test validity criteria satisfied	Not stated
Water quality criteria satisfied	Not stated
Endpoint comment	Few details available. Flow-through study using measured concentrations. A significant effect on growth at the 95% level at 4 mg l ⁻¹ . Significant effect on survival only at 13.7 mg l ⁻¹ .
Study conducted to GLP	Not stated

Reliability of study	Reliable
Relevance of study	Relevant
Klimisch Code	2

Reference number	16
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Information on the test species	
Test species used	<i>Oncorhynchus mykiss?</i> (more likely to be <i>Pimephales promelas</i>)
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 (expressed as 1,1,2,2-tetrachloroethane)
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	Flow-through
Measurement of exposure concentrations	Measured
Measurement of water quality parameters	pH, hardness and temperature
Test validity criteria satisfied	Not stated
Water quality criteria satisfied	Not stated
Endpoint comment	This data was taken from the US EPA ECOTOX database. However, the actual report makes no mention of long-term studies with rainbow trout and the only long-term fish data were for fathead minnow exposed for 32 days to 1,1,2,2-tetrachloroethane. In that study, a concentration of 4 mg l ⁻¹ had a significant effect on growth, but not mortality. These data are also identical to that of DeFoe 1980 [15] and is likely to be the same study (see above for details). Effects in fathead minnow at 1.4 mg l ⁻¹ were reported for tetrachloroethene but not TCE.
Study conducted to GLP	Not stated

Reliability of study	Unreliable
Relevance of study	Relevant
Klimisch Code	4

Reference number	17
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Information on the test species	
Test species used	<i>Oryzias latipes</i>
Source of the test organisms	Not stated
Holding conditions prior to test	Not stated
Life stage of the test species used	9–10 days old

Information on the test design	
Methodology used	Not stated
Form of the test substance	TCE
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	Nominal
Measurement of water quality parameters	Not stated
Test validity criteria satisfied	Not stated
Water quality criteria satisfied	Not stated
Endpoint comment	No effect up to highest concentration tested.
Study conducted to GLP	Not stated

Reliability of study	Unreliable
Relevance of study	Relevant
Klimisch Code	3

Reference number	18
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Information on the test species	
Test species used	<i>Daphnia magna</i>
Source of the test organisms	Laboratory stocks
Holding conditions prior to test	Held in deionised well water
Life stage of the test species used	<24 hours old

Information on the test design	
Methodology used	Followed standard US EPA methodology.
Form of the test substance	1,1,2,2- and 1,1,1,2-tetrachloroethane
Source of the test substance	Purity >80%
Type and source of the exposure medium	Not stated
Test concentrations used	5–8 concentrations
Number of replicates per concentration	Tests run in triplicate
Number of organisms per replicate	5
Nature of test system (static, semi-static or flow-through, duration, feeding)	Static (48 hours duration)
Measurement of exposure concentrations	Nominal
Measurement of water quality parameters	pH, DO and temperature
Test validity criteria satisfied	Not stated
Water quality criteria satisfied	Reported for all tests carried out (>50). Not possible to establish if met for TCE test.
Endpoint comment	Endpoint appears valid, although based on nominal concentrations.
Study conducted to GLP	Not stated

Reliability of study	Unreliable
Relevance of study	Relevant
Klimisch Code	3

Reference number	19
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Information on the test species	
Test species used	<i>Lepomis macrochirus</i>
Source of the test organisms	Commercial fishery
Holding conditions prior to test	Held in well water
Life stage of the test species used	0.3–1.2 g

Information on the test design	
Methodology used	Followed standard US EPA methodology.
Form of the test substance	>80% pure substance
Source of the test substance	1,1,1,2- and 1,1,2,2,-tetrachloroethane
Type and source of the exposure medium	Well water
Test concentrations used	Not stated
Number of replicates per concentration	Not stated
Number of organisms per replicate	10
Nature of test system (static, semi-static or flow-through, duration, feeding)	Static test in sealed jars
Measurement of exposure concentrations	Nominal
Measurement of water quality parameters	pH, DO
Test validity criteria satisfied	Not stated
Water quality criteria satisfied	Yes
Endpoint comment	No information on replicates or concentration range. The chemicals were recorded as soluble, but concentrations were nominal.
Study conducted to GLP	Not stated

Reliability of study	Unreliable
Relevance of study	Relevant
Klimisch Code	3

Reference number	20
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Information on the test species	
Test species used	<i>Vibrio fischeri</i> I and <i>Nitrosomonas</i>
Source of the test organisms	Not stated
Holding conditions prior to test	Not stated
Life stage of the test species used	Population

Information on the test design	
Methodology used	Microtox standard method
Form of the test substance	Not stated (expressed as 1,1,2,2-tetrachloroethane)
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)	Open vessels for Microtox
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
Endpoint comment	Very few details available. Tests run in open vessels with no mention of analysis of exposure concentrations.
Study conducted to GLP	Not stated

Reliability of study	Unreliable
Relevance of study	Unknown
Klimisch Code	3

Reference number	21
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Information on the test species	
Test species used	<i>Scenedesmus subspicatus</i>
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	1,1,2,2-tetrachloroethane
Source of the test substance	Purity >90%
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 (closed vessel)
Measurement of exposure concentrations	Measured
Measurement of water quality parameters	No data
Test validity criteria satisfied	Not stated
Water quality criteria satisfied	Not stated
Endpoint comment	Endpoint appears valid, although only limited data with which to assess validity.
Study conducted to GLP	Not stated

Reliability of study	Reliable
Relevance of study	Relevant
Klimisch Code	2

Reference number	24
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Information on the test species	
Test species used	<i>Artemia salina</i>
Source of the test organisms	Encysted eggs bought from Bay Brand (San Francisco)
Holding conditions prior to test	Held for 3–4 days prior to testing in synthetic seawater (cysts held at 4°C and nauplii at 25°C).
Life stage of the test species used	Nauplii

Information on the test design	
Methodology used	Not carried out to a standardised methodology, but a well-documented study.
Form of the test substance	1,1,2,2-tetrachloroethane (analysis grade)
Source of the test substance	Sigma chemicals
Type and source of the exposure medium	Artificial seawater
Test concentrations used	Various, but not stated
Number of replicates per concentration	4
Number of organisms per replicate	10
Nature of test system (static, semi-static or flow-through, duration, feeding)	Static
Measurement of exposure concentrations	Nominal
Measurement of water quality parameters	Not stated
Test validity criteria satisfied	Not stated
Water quality criteria satisfied	Not stated
Endpoint comment	Different aged nauplii exposed. The 72-hour organisms were an order of magnitude more sensitive than the 24- and 48-hour organisms. All concentrations were nominal and there was no mention of sealed vessels to prevent volatilisation.
Study conducted to GLP	Not stated

Reliability of study	Unreliable
Relevance of study	Relevant
Klimisch Code	3

Reference number	25
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Information on the test species	
Test species used	<i>Cyprinodon variegatus</i>
Source of the test organisms	Laboratory reared
Holding conditions prior to test	Held in seawater at 25–31°C
Life stage of the test species used	14–28 days old

Information on the test design	
Methodology used	Followed standard US EPA methodology.
Form of the test substance	1,1,2,2-tetrachloroethane
Source of the test substance	Commercial
Type and source of the exposure medium	Filtered natural seawater
Test concentrations used	Not stated
Number of replicates per concentration	Not stated
Number of organisms per replicate	10
Nature of test system (static, semi-static or flow-through, duration, feeding)	Static
Measurement of exposure concentrations	Nominal
Measurement of water quality parameters	pH, DO
Test validity criteria satisfied	Yes
Water quality criteria satisfied	Not stated
Endpoint comment	Nominal concentrations, with no mention of sealed containers to prevent volatilisation. In addition, jars were only three-quarters full with water so a headspace was available.
Study conducted to GLP	Not stated

Reliability of study	Unreliable
Relevance of study	Relevant
Klimisch Code	3

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