

# ***Common Implementation Strategy for the Water Framework Directive***

**Environmental Quality Standards (EQS)**

**Substance Data Sheet**

**Priority Substance No. 32**

**Trichloromethane**

**CAS-No. 67-66-3**

***Final version  
Brussels, 31 July 2005***

**Disclaimer**

*This data sheet provides background information on the setting of the Environmental Quality Standard in accordance with Article 16 of the Water Framework Directive (2000/60/EC). The information was compiled, evaluated and used as outlined in the Manual<sup>[4]</sup> and has been discussed in a consultative process with the Expert Advisory Forum on Priority Substances and the Expert Group on Quality Standards. Furthermore, it has been peer-reviewed by the SCTEE<sup>[12]</sup>. The substance data sheet may, however, not necessarily represent the views of the European Commission.*

*New upcoming information was considered and included up to the date of finalisation of this data sheet. Information becoming available after finalisation of this document will be evaluated in the review process of priority substances according to Art. 16(4) of the Water Framework Directive. If necessary, the Environmental Quality Standard substance data sheets will then be revised in the light of technical and scientific progress.*

## 1 Identity of substance

Priority Substance No: 32	Trichloromethane
CAS-Number:	67-66-3
Classification WFD Priority List <sup>*</sup> :	PS

\* PS: priority substance; PHS: priority hazardous substance; PSR: priority substance under review according to Decision 2455/2001.

## 2 Proposed quality standards

### 2.1 Overall quality standards

Ecosystem	Quality Standard	Quality Standard "rounded values"	Comment
AA-QS all surface waters covered by the WFD	2.5 µg/l corresponding to 12µg/kg sediment (wet wt)	<b>2.5 µg/l</b> <b>corresponding to</b> <b>12µg/kg sediment (wet wt)</b>	see section 8.2
MAC-QS (ECO) <sup>*</sup>	266 µg/l	<b>270 µg/l</b>	see section 8.1

\* The proposal by the Commission may include a MAC-QS value which is based on the calculation of 12 \* AA-EQS. This derivation is based on the minimum annual frequency of monitoring of priority substances in accordance with the Water Framework Directive. The derivation of such a MAC-QS is based on monitoring, compliance and reporting considerations rather than derived from effect data as presented in this EQS datasheet.

### 2.2 Specific quality standards

Protection Objective <sup>#</sup>	Quality Standard	Comment
Pelagic community (freshwater & saltwater)	146 µg/l	see section 8.1
Benthic community (sediment)	12 µg/kg sediment (wet wt) corresponding concentration in water: 2.5 µg/l	see section 8.2
Predators (secondary poisoning)	Derivation of QS not required	trigger for derivation of QS not met see section 8.3
Food uptake by man	Suspected carcinogen, derivation of QS required but not possible based on the data/information given in the EU-RAR	see sections 7 & 8.4
Abstraction of water intended for human consumption (AWIHC)	No DW abstraction standard set by CD 75/440/EEC. As removal efficiency in drinking water processing is presumably higher than 80% the derivation of a standard specifically addressing drinking water abstraction is not deemed necessary.	see section 8.5
Water intended for human consumption (WIHC)	< 100 µg/l	drinking water limit value set by CD 98/83/EC for Σ trihalomethanes is 100 µg/l

# If justified by substance properties or data available, QS for the different protection objectives are given independently for freshwater environments, transitional waters or coastal and territorial waters

### 3 Classification

R-Phrases and Labelling	Reference
<p>1 % ≤ conc. &lt; 5 % 5% ≤ conc. &lt; 20 %  conc. ≥ 20 %</p> <p><b>R 40</b> [Limited evidence of a carcinogenic effect] <b>R 22</b> [Harmful if swallowed] - <b>40-48/20/22</b> [Harmful: danger of serious damage to health by prolonged exposure through inhalation and if swallowed] <b>R 22-38</b> [Irritating to skin] <b>40-48/20/22</b></p>	[1]

### 4 Physical and chemical properties

Property	Value	Ref.
Molecular weight	119.5 g/mol	[1]
Vapour pressure	209 hPa at 20°C	[1]
Henry's law constant	275 Pa.m <sup>3</sup> /mol at 20°C	[1]
Solubility in water	8,700 mg/l at 20°C	[1]

### 5 Environmental fate and partitioning

Property	Value	Ref.	Comments
Hydrolysis			[1]: hydrolysis is an unimportant fate process at a neutral pH value
Photolysis			[1]It is concluded that direct photolysis is not an important fate process
<u>Biodegradation</u> aerobic:  anaerobic:  sediment:	DT50: 15 d (k <sub>sed</sub> = 0.046 d <sup>-1</sup> )	[1]	<p>[1]: ... degradation of chloroform occurs only under certain aerobic conditions by methane-utilising bacteria. However, the respective results cannot be used in the generic assessment. The first order rate constant for aerobic biodegradation in soil and sediment is 0 d<sup>-1</sup>..</p> <p>[1]: ... In conclusion, although a certain biodegradation can be mentioned to take place under some anaerobic conditions, chloroform is not considered readily biodegradable in water systems.</p> <p>[1]: ... biodegradation is observed in anaerobic sediment. Based on these results, half lives determined by Van Beelen and Van Keulen (1990) are assumed to be valid for the anaerobic part of the sediment. In the TGD, it is proposed to consider that 9/10 of the sediment is anaerobic. Therefore, a half live of 15 days is used in this assessment for the sediment.</p>
<u>Partition coefficients</u> Octanol – Water	Log Kow 1.97	[1]	[1]: used in the risk assessment
Koc (organic carbon-water)	185	[1]	[1]: used in the assessment
K <sub>p</sub> <sub>susp</sub>	18.5 l/kg	[1]	K <sub>p</sub> <sub>susp</sub> , K <sub>susp-water</sub> , K <sub>sed-water</sub> are estimated using standard OC contents and algorithms given in the TGD
K <sub>susp-water</sub>	5.53 m <sup>3</sup> /m <sup>3</sup>	[1]	
K <sub>sed-water</sub>	5.42 m <sup>3</sup> /m <sup>3</sup>	[1]	
<u>Bioaccumulation</u> Bioconcentration Factor (BCF) Fish	1.4 – 13 13	[1] [1]	Worst case BCF used in the RA

### 5.1 Natural sources of trichloromethane

At the meeting of the Expert Group on Quality Standards (12-16 May, Brussels), EUROCHLOR suggested to take the natural background concentration of trichloromethane, resulting from significant emissions by natural sources, into account when deriving a water quality standard for this substance. EUROCHLOR considered the Added Risk Approach as used to derive EQSs for some metals as suitable for trichloromethane as well.

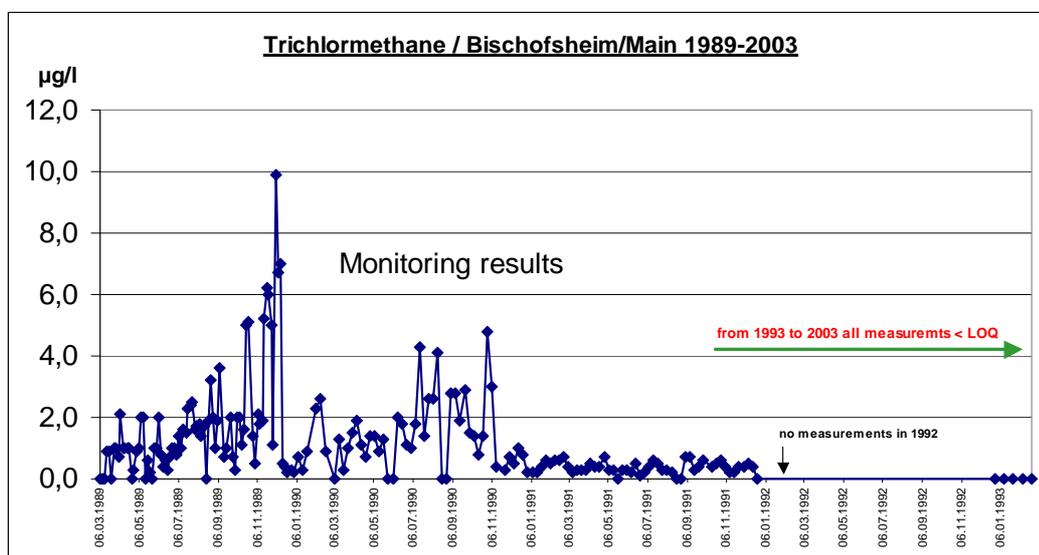
Chloroform is released by both natural and anthropogenic processes. Approximately 90% of the emissions may originate from non-anthropogenic sources<sup>[9]</sup>. However, due to the high volatility of chloroform, these natural releases will mainly influence the final air concentration. The equilibrium distribution to air is greater than 99%<sup>[9]</sup>. The mean concentration in European rivers is calculated to be 0.5 µg/l and concentrations reported for the open ocean range from 0.007 – 1.09 µg/l<sup>[9]</sup>.

The contribution of natural sources to the environmental concentration of trichloromethane is considered and discussed as well in the ongoing risk assessment for this substance. The conclusion drawn in the current draft of the RAR is as follows:

*... although one can not deny that chloroform might be released by natural processes, the global contribution of these phenomena to chloroform emissions to the air and the terrestrial compartments can not be assessed. All available studies are actually giving empirical calculations based on specific measurements. Therefore, natural emissions of chloroform will be neglected in this risk assessment.<sup>[1]</sup>*

The French Rapporteur also recommends to neglect a possible natural background concentration of trichloromethane for the purpose of deriving a surface water quality standard for this substance<sup>[8]</sup>. This position is corroborated by an analysis of monitoring data conducted by the German Federal Environmental Agency. The data (see figure 1) clearly show that concentrations downstream of a trichloromethane emitting industrial plant dropped below the limit of quantification (0.1 – 0.3 µg/l) after a properly working sewage treatment plant went into operation<sup>[10]</sup>.

Figure 1:



Taken all above mentioned facts together, it seems not necessary to take natural background concentrations of trichloromethane into account for the purpose of quality standard setting because in relation to the concentrations that are proposed (respectively will be set) as quality standards the natural background levels are insignificantly low.

## 6. Effect data (aquatic environment) <sup>[1]</sup>

Aquatic toxicity data assessed and used in the risk assessment report <sup>[1]</sup> are listed in Annex 1 to this data sheet.

### 6.1 Predicted no-effect concentrations (aquatic environment)

Table 6.1: PNECs

Compartment	Value	Reference
Surface water	146 µg/l	[1]
Sediment	12 µg/kg wet wt; 55 µg/kg dry wt	[1]
PNEC <sub>food</sub> (secondary poisoning)	an assessment of non-specific exposure has not been carried out because of the low BCF of trichloromethane	[1]

#### 6.1.1 Calculation of the PNEC surface water <sup>[1]</sup>

Fish :

NOEC-6/9 months: 1.463 mg/l (*Oryzias latipes*, Toussaint *et al.*, 2001)

Invertebrate:

NOEC-21d: 6.3 mg/l (*Daphnia magna*, Kühn *et al.*, 1989)

Algae:

72h-EC 10: 3.61 mg/l (*Chlamydomonas reinhardtii*, Brack & Rottler, 1994)

There are three long-term NOECs from species representing three trophic levels. Therefore, the PNEC is derived using an assessment factor of 10 to the lowest NOEC.

$$\text{PNEC}_{\text{aqua}} = 1.463 / 10 = 146 \text{ µg/l}$$

#### 6.1.2 Calculation of the PNEC sediment <sup>[1]</sup>

Van Vlaardingen & van Beelen (1992) studied the toxicity of chloroform to the methanogenesis. Chloroform solution as a dilution in methanol was added to a sediment / water suspension. The sediment was primarily composed of methanogenic mud. Test bottles were incubated for 11 days at 20°C in a rotary shaker. Methane production in the contaminated bottles was measured at the end of the experiment and compared to the methane production of the blank bottles. Then EC 10 and EC 50 could be calculated. Although some details on experimental conditions are lacking, the EC10 (see Table A1.6 in Annex 1) can be used as a long term toxicity test result as methanogenesis is an important route of degradation of organic matter.

There are two methods of determination of PNEC<sub>sed</sub> :

##### 1) Determination of the PNEC<sub>sed</sub> using the sediment toxicity test

An assessment factor of 100 should be applied to the EC10 from the test on the inhibition of methane production:

$$\text{PNEC}_{\text{sed}} (1) = 55 \text{ µg/kg (dw)}$$

## 2) Determination of the $PNEC_{sed}$ using the Equilibrium partitioning method

According to the TGD,  $PNEC_{sed}(ww) = \frac{K_{susp-water}}{RHO_{susp}} * PNEC_{aquatic} * 1000$

$K_{susp-water}$  = suspended matter\_water partition coefficient =  $5.53 \text{ m}^3 \cdot \text{m}^{-3}$

Therefore:  $PNEC_{sed} = 702 \text{ } \mu\text{g} \cdot \text{kg}^{-1}$  (ww)  
 $PNEC_{sed} = 3230 \text{ } \mu\text{g} \cdot \text{kg}^{-1}$  (dw)

The result with the Equilibrium partitioning method is much higher than the result based on the toxicity to methanogenic bacteria. However, this value based on experimental results will be preferred:  **$PNEC_{sed} = 55 \text{ } \mu\text{g}/\text{kg}$  (dw) and  $PNEC_{sed} = 12.0 \text{ } \mu\text{g}/\text{kg}$  (ww)**

### 6.1.3 Calculation of the PNEC for non compartment specific effects relevant for the food chain (secondary poisoning)<sup>[1]</sup>

Because of the low bioaccumulation potential of chloroform (BCF = 13), the potential for secondary poisoning can be considered to be negligible. This is confirmed by the monitoring data available from marine aquatic biota as well as in birds.

## 6.2 Summary on endocrine disrupting potential

A (suspected) potential of trichloromethane to exert adverse effects on endocrine regulation is not mentioned in the RA<sup>[1]</sup>. Further, trichloromethane is not mentioned in the "Community Strategy for Endocrine Disrupters"<sup>[6]</sup>.

## 7 Effect data (human health)

An NOAEL of 17 mg/kg/d was assumed for overt toxicity on the liver and the kidneys in rats and mice by inhalation. By proceeding to physiologically-based pharmacokinetic models, the NOAEL for human exposure associated with hepatocellular toxicity was estimated at 4,600 mg/kg/d<sup>[1]</sup>.

No threshold levels (e.g. NOAEL values) with relevance to oral uptake of trichloromethane are mentioned in the draft RAR<sup>[1]</sup> or have been submitted by other Member States or by NGOs.

However, as the risk assessment is showing that there are emissions to air and water, an estimation of the exposure of man via the environment is ongoing but not yet completed<sup>[1]</sup>. Results already available suggest that the highest exposures are to be expected through intake of drinking water, intake of fish and through intake of air. Toxicity by inhalation seems to be the critical end point for man exposed indirectly via the environment.

The substance is already heavily regulated for human exposure and therefore it cannot be expected that additional risk reduction measures will taken on the basis of a detailed RAR. This was discussed and agreed by the CAs.

## 8 Calculation of quality standards

### 8.1 Quality standards for water

#### *Freshwater*

The  $PNEC_{water}$  as identified in <sup>[1]</sup> (146 µg/l, see section 6.1.1 of this data sheet) is proposed as the quality standard for freshwater.

$$QS_{freshwater} = 146 \mu\text{g Trichloromethane / l}$$

The  $\log Kp_{susp}$  is <3 and therefore the trigger criterion to calculate a corresponding  $QS_{SPM.freshwater}$  referring to the concentration of trichloromethane in suspended particulate matter (SPM) is not met.

#### *Transitional, coastal and territorial waters*

Toxicity data on marine organisms are available in the RAR <sup>[1]</sup> for fish, crustaceans and algae (see annex 1 to this data sheet). The sensitivities of saltwater and freshwater species belonging to the same taxonomic groups are comparable. The same conclusion has been drawn from an evaluation of the available toxicity data in the Euro Chlor risk assessment for trichloromethane in the marine environment <sup>[2]</sup>. It is therefore suggested to calculate the  $QS_{saltwater}$  from the same data set as used for the derivation of the  $QS_{freshwater}$ . To this end, the TGD assessment factor method as proposed for the marine effects assessment is used (section 4.3 in chapter 3b of volume II of the draft revised TGD <sup>[3]</sup>).

The acutely most sensitive species has an LC50 value lower than the lowest NOEC value. Therefore the QS would normally be derived by applying an assessment factor of 1000 to the lowest LC50 resulting in a  $QS_{saltwater}$  of 0.39 µg/l trichloromethane. However, given the fact that this LC50 has been obtained with a marine species and that trichloromethane is believed to have a non-specific narcotic mode of toxic action <sup>[2, 7]</sup>, it appears not reasonable to expect a significantly greater sensitivity of other saltwater species. It is therefore suggested to use the same assessment factor as used to derive the freshwater quality standard (10). Hence, the resulting saltwater quality standard is equal to the freshwater standard.

$$QS_{saltwater} = QS_{freshwater} = 146 \mu\text{g Trichloromethane / l}$$

The  $\log Kp_{susp}$  is <3 and therefore the trigger criterion to calculate a corresponding  $QS_{SPM.saltwater}$  referring to the concentration of trichloromethane in suspended particulate matter (SPM) is not met.

#### *Quality standard accounting for transient concentration peaks (MAC-QS)*

It is suggested to derive the MAC-QS on the basis of the lowest acute toxicity test available that is considered valid in the risk assessment. This is the 72 h EC50 of 13.3 mg/l reported for the green algae *Chlamydomonas reinhardtii*. For substances with an acute to chronic ratio lower than 10, the French Rapporteur recommends to use an AF of 50 instead of 100 to derive the MAC-QS <sup>[8]</sup>. This recommendation is followed:

$$MAC-QS = 13300 \mu\text{g/l / AF (50)} = 266 \mu\text{g Trichloromethane / l}$$

## 8.2 Quality standard for sediment

The log  $K_{p\_susp}$  is <3 and therefore the trigger criterion to derive a  $QS_{sediment}$  is not met.

However, some concern for the sediment compartment has been identified in the RAR on the basis of the experimental data available. Additional testing of benthic organisms has therefore been asked for under article 10 (2) of regulation 793/93 (April 2005). The tests have not been conducted yet.

It is therefore suggested to use current  $PNEC_{sediment}$  derived in the risk assessment<sup>[1]</sup> (see section 6.1.2 of this data sheet) as sediment quality standard.

$$QS_{sediment} = PNEC_{sediment} = 12 \mu\text{g Trichloromethane/ kg sediment (wet weight)}$$

For the reasons explained in section 8.1 it is not expected that marine sediment, respectively marine benthic organisms, are more sensitive towards chloroform than freshwater benthos. Therefore, the same QS of 12  $\mu\text{g/l}$  may apply for freshwater sediments as well as for those of transitional, coastal and territorial waters.

The corresponding concentration of the  $QS_{sediment}$  in water can be calculated with the Equilibrium Partitioning method.

$$QS_{water} = \frac{QS_{sediment} (12\mu\text{g/kg ww}) * RHO_{susp} (1150 \text{ kg/m}^3)}{K_{susp-water} (5.53 \text{ m}^3/\text{m}^3) * 1000} = 2.5 \mu\text{g trichloromethane/l}$$

with:

$K_{susp\_water}$       suspended matter - water partition coefficient  
 $RHO_{susp}$         density of suspended particulate matter

Hence the concentration in water corresponding to a  $QS_{sediment}$  of 12  $\mu\text{g/kg}$  is a  **$QS_{water}$  of 2.5  $\mu\text{g/l}$** .

Consequently, the protection of the bentic community requires a concentration in water that is lower than that necessary for the protection of the pelagic community. Further to the finalisation of the additional testing carried out in the context of the risk assessment report, the proposed  $QS_{sediment}$  may need to be reviewed.

## 8.3 Secondary poisoning of top predators

The BCF of trichloromethane is <100 and therefore the trigger criterion to derive a quality standard addressing secondary poisoning of predators ( $QS_{secpois}$ ) is not met (see also section 6.1.3 of this data sheet).

## 8.4 Quality standard referring to food uptake by humans

Trichloromethane is classified as carcinogen of category 3 (R40) and therefore the derivation of a quality standard addressing the protection of human health from adverse effects due to the uptake of food originating from aquatic environments is required (trigger criteria met, see table 1b in<sup>[4]</sup>).

An estimation of the exposure of man via the environment is conducted in the context of the ongoing EU risk assessment but not yet completed<sup>[1]</sup>. Results already available suggest that the highest exposures are to be expected through intake of drinking water, intake of fish and through intake of air. Toxicity by inhalation seems to be the critical end point for man exposed indirectly via the environment.

Based on the data / information currently available in the RAR it is not possible to calculate a QS referring to the protection from adverse health effects due to the uptake of fishery products.

The US-EPA recently concluded that trichloromethane is not likely to be carcinogenic to humans by any routes of exposure at a dose level that does not cause cytotoxicity and cell regeneration. This conclusion is supported by the finding that trichloromethane is not a strong mutagen and is not likely to cause cancer through a genotoxic mode of action. The US-EPA concluded that daily exposure to 10 µg/kg body weight per day is safe from the point of view of carcinogenicity (reference<sup>[11]</sup>, as cited in<sup>[9]</sup>).

### 8.5 Quality standard for drinking water abstraction

No "A1- guide value" has been set in the context of Council Directives 75/440/EEC. The drinking water standard set in CD 98/83/EC is 100 µg/l (maximum acceptable level for Σ-Trihalomethanes). Hence the drinking water standard is lower than the quality standard required to protect the freshwater community from adverse effects (146 µg/l).

An assessment by experts in drinking water technology with regard to the question which percentage of the amount of trichloromethane present in raw water can be removed by usual simple treatment procedures might be helpful. If the respective percentage was known, this figure could be used together with the drinking water standard to estimate the maximum level in surface water that can be removed during drinking water production by simple treatment.

According to a calculation with the SIMPLETREAT model conducted by the French Rapporteur, removal in STPs is 83.1 % (high volatile substance). That leads to an indicative QS<sub>drinking water before simple treatment</sub> of 592 µg/L<sup>[8]</sup>.

### 8.6 Overall quality standard

The current water quality objective for trichloromethane established by Council Directive 86/280/EEC is 12 µg/l.

Protection of human health from adverse effects due to the cancerogenic properties of trichloromethane is the objective that is not yet properly addressed in this data sheet. However, as the cancerogenic potential of trichloromethane appears to be rather low (see sections 7 and 8.5) and the substance is not liable to bioaccumulation, it is not very probable that human health related protection objectives (protection of drinking water abstraction, protection from the occurrence of adverse health effects due to the uptake of fishery products) are those that might require the lowest levels in the aquatic environment.

The quality standard derived for the protection of the benthic community (12 µg/kg sediment corresponding to 2.5 µg/l) is the lowest. Therefore, this value is suggested as overall AA-QS.

## 9 References

- [1] European Union Risk Assessment Report: Chloroform (CAS No: 67-66-3 EINECS No: 200-663-8) Draft report, August 2003, France (file: R047\_0308\_env.doc. ). The (final) report may be available from the internet-site of the European Chemicals Bureau: <http://ecb.jrc.it/existing-chemicals/>
- [2] Zok, S et al., 1998 : Euro Chlor Risk Assessment for the Marine Environment, OSPARCOM Region North Sea – Chloroform. Environmental Monitoring and Assessment 53(3), pp 401-424
- [3] Technical Guidance Document on Risk Assessment in Support of Commission Directive 93/67/EEC on Risk Assessment for New Notified Substances and Commission Regulation (EC) No 1488/94 on Risk Assessment for Existing Substances and Directive 98/8/EC of the European Parliament and the Council Concerning the placing of biocidal products on the market. Part II. European Commission Joint Research Centre, EUR 20418 EN/2, © European Communities 2003. Available at the internet-site of the European Chemicals Bureau: <http://ecb.jrc.it/existing-chemicals/>
- [4] Manual of the Methodological Framework Used to Derive Environmental Quality Standards for Priority Substances of the Water Framework Directive. Peter Lepper, Fraunhofer-Institute Molecular Biology and Applied Ecology, 15 November 2004. Available at the internet-site of the European Commission: [http://europa.eu.int/comm/environment/water/water-dangersub/pri\\_substances.htm](http://europa.eu.int/comm/environment/water/water-dangersub/pri_substances.htm)
- [5] Council Directive 86/280/EEC on limit values and quality objectives for discharges of certain dangerous substances included in List I of the Annex to Directive 76/464/EEC. OJ L 181, 04/07/1986, p. 16-27
- [6] COM(2001)262 final: Communication from the Commission to the Council and the European Parliament on the implementation of the Community Strategy for Endocrine Disrupters – a range of substances suspected of interfering with the hormone system of humans and wildlife.
- [7] De Bruijn, J. et al.: Environmental Risk Limits in The Netherlands. National Institute of Public Health and the Environment (RIVM), Bilthoven. RIVM Report No. 601 640 001, Parts I-III, Appendix C – section on trichloromethane
- [8] Personal communication: Additional French comments for the derivation of Quality Standards – Chloroform. E-mail by Vincent Bonomet, INERIS, 29.07.2003
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- [10] Personal communication. Dr. Christiane Markard, German Federal Environmental Agency, Berlin. E-mail of 09.07.2003
- [11] U.S. Environmental Protection Agency, 2001: Toxicological Review of Chloroform (CAS No. 67-66-3) in Support of Summary Information on the Integrated Risk Information System (IRIS), Report No. EPA/635/R-01/001. Reference cited in<sup>[9]</sup>
- [12] Opinion of the Scientific Committee on Toxicity, Ecotoxicity and the Environment (SCTEE) on “The Setting of Environmental Quality Standards for the Priority Substances included in Annex X of Directive 2000/60/EC in Accordance with Article 16 thereof ”, adopted by the SCTEE during the 43<sup>rd</sup> plenary meeting of 28 May 2004, European Commission Health & Consumer Protection Directorate General, Brussels. [http://europa.eu.int/comm/health/ph\\_risk/committees/sct/documents/out230\\_en.pdf](http://europa.eu.int/comm/health/ph_risk/committees/sct/documents/out230_en.pdf)

## Annex 1: Aquatic toxicity data assessed and used in the risk assessment report <sup>[1]</sup>

### Meaning of the reliability indices given in the tables:

Reliability index 1 : Method and description in accordance with test guidelines and with accurate actual concentrations measurements

Reliability index 2 : Falling short of highest standards concerning protocol or reporting

Reliability index 3 : Not valid

Reliability index 4 : Validity cannot be established due to missing information

Table A1.1: Acute toxicity results with fish

Species	Method	Endpoint (mg/l)	Reliability index	Remarks	Reference
<i>Limanda limanda</i>		LC 50-96 h = 28 mg/l	4	Flow-through system, analytical monitoring  No details on the experimental conditions	Pearson & McConnell, 1975
<i>Lepomis macrochirus</i>		LC 50-96 h = 18 mg/l (mean LC 50 of 5 tests)	3	Daily analytical monitoring. No clear dose-effect relation.  High mortality due to <i>Columnaris</i> infection in the control and the lower concentration	Anderson & Lusty, 1980
<i>Poecelia reticulata</i>	US EPA, 1971	LC 50-96 h = 300 mg/l	3	No analytical monitoring  static test, insufficient documentation	Hazdra et al., 1979
<i>Leuciscusidus</i>	DIN 38412	LC 0-48 h = 51 mg/l LC 50-48 h = 92 mg/l LC 100-48 h = 151 mg/l	3	The test system is not appropriate for volatile substances	Knie et al., 1983
<i>Leuciscusidus melatonus</i>	DIN 38412, 48 h	LC 0-48 h = 147 mg/l LC 50-48 h = 162 mg/l LC 100-48 h = 176 mg/l	3	No analytical monitoring  The test system is not appropriate for volatile substances	Juhnke & Lüdemann, 1978
<i>Oncorhynchus mykiss</i>		LC 50-48 h = 20 mg/l	3	No analytical monitoring. Insufficient documentation	Slooff, 1979, 1984
<i>Brachydanio rerio</i>		LC 50-48 h = 100 mg/l	3	No analytical monitoring. Insufficient documentation	Slooff, 1979, 1984
<i>Oryzias latipes</i>	Testing methods for industrial and waste water, Japan	LC 50-48 h = 117 mg/l	3	No analytical monitoring  Semi-static system	MITI, 1992

Species	Method	Endpoint (mg/l)	Reliability index	Remarks	Reference
<i>Cyprinus carpio</i>		LC 50-3-5 d = 97 mg/l (toxicity to carp embryos)	2	Semi-static system 2 initial concentration are measured. LC 50 value is corrected with estimated mean concentration during the static period	Mattice et al., 1981
<i>Ictalurus punctatus</i> (juvenile catfish)		LC 50-96 h = 75 mg/l	2	Daily analytical monitoring. Flow-through toxicant delivery system	Anderson & Lusty, 1980
<i>Pimephales promelas</i>	US-EPA, 96 h	LC 50-96 = 71 mg/l	2	Flow-through system, daily analytical monitoring	Geiger et al., 1990
<i>Pimephales promelas</i>	ASTM, 1980,	Fry : LC 50-96 h = 129 mg/l Juvenile : LC 50-96 h = 171 mg/l Subadults : LC 50-96 h = 103 mg/l	2	static, closed system , no analytical monitoring	Mayes et al., 1983
<i>Oncorhynchus mykiss</i>		LC 50-96 h = 18 mg/l (mean LC 50 of 5 tests)	2	Daily analytical monitoring. Flow-through toxicant delivery system	Anderson & Lusty, 1980
<i>Micropterus salmoides</i>		LC 50-96 h = 51 mg/l (mean LC 50 of 3 tests)	2	Daily analytical monitoring. Flow-through toxicant delivery system	Anderson & Lusty, 1980
<i>Poecelia reticulata</i>		experimental : LC 50-14 d = 102 mg/l calculated : LC 50-14d = 154 mg/l	2	No analytical monitoring Semi-static system Use of solvent	Könemann, 1981
<i>Brachydanio rerio</i>	OECD 203	LC50-96h =121 mg/l	1	Flow-through test (6 renewals per day) with analytical monitoring	Röderer, 1990

Table A1.2: Chronic toxicity results with fish and amphibians

Species	Method	Endpoint (95 % confidence limit)	Reliability index	Remarks	Reference
<i>Pimephales promelas</i> :		LC 50-9 d > 58 mg/l	3	flow through; exposure period: 9 days; analytical monitoring	Black et al., 1982
<i>Oncorhynchus mykiss</i>		LC 10 = 83.2 µg/l (9.4 - 251.4 µg/l)	3	flow through; exposure period : 27 days; analytical monitoring	cited by Black et al., 1982
<i>Oncorhynchus mykiss</i>		LC 1-27 d = 0.0062 mg/l LC50-27d=2.03mg/l (water hardness = 48 mg/l)  LC 1-27 d = 0.0049 mg/l LC50-27d=1.24mg/l (water hardness = 210 mg/l)	3	flow through; exposure period : 27 days; analytical monitoring	Birge et al., 1979
<i>Oncorhynchus mykiss</i>	24 h flow-through system	LOEC = 20 mg/l  (increasing of the respiration frequency)	3	No analytical monitoring, Uncommon endpoint  Flow-through closed dynamic system	Slooff, 1979
<i>Poecilia sphenops</i>	60 d	NOEC < 1.5 mg/l (mortality, distress, inhibition of growth and fatty change of the liver)	3	Semi-static system (complete renewal every 2 weeks)  No analytical monitoring  2 concentrations, no replicate, only 6 fish per concentration	Loekle et al., 1983
<i>Oryzias latipes</i>		LOEC-6/9 months = 1.464 mg/l  NOEC-6/9 months = 0.151 mg/l  NOEC-6/9 months > 1.463 mg/l	1	Flow-through exposure system with weekly analyses  Lesions in gallbladder and abnormalities of the bile ducts  length, growth	Toussaint <i>et al.</i> , 2001
<i>Brachydanio rerio</i>	14 d	LOEC=13 mg/l,  NOEC = 6.1 mg/l (position of the fish in the aquaria)	1	Flow-through system (6 renewals per day) with analytical monitoring	Röderer, 1990
<i>Rana temporaria</i> *		LC 50-5 d = 16.95 mg/l (11.05 – 28.91 mg/l)	3	flow through; exposure period: 5 days; analytical monitoring	Black et al., 1982
<i>Ambystoma gracile</i> *		LC 50-5 d = 21.58 mg/l (13.25 – 41.77 mg/l)	3	flow through; exposure period: 5 days; analytical monitoring	Black et al., 1982
<i>Xenopus laevis</i> *		LC 50-5 d > 68 mg/l	3	flow through; exposure period: 5 days; analytical monitoring	Black et al., 1982
<i>Hyla crucifer</i> *		LC 50-7 d = 0.27 mg/l (0.19 – 0.37 mg/l)  LC 10-7 d = 17.7 µg/l (9.9 – 28.1 µg/l)  LC 1-7 d = 1.9 µg/l (0.8 - 3.9 µg/l)	3	flow through; exposure period: 7 days; analytical monitoring	Birge et al., 1980
<i>Bufo fowleri</i> *		LC 50-7 d = 35.14 mg/l (18.37 – 92.25 mg/l)	3	flow through; exposure period: 7 days; analytical monitoring	Birge et al., 1980

Species	Method	Endpoint (95 % confidence limit)	Reliability index	Remarks	Reference
<i>Rana pipiens</i> *		LC50-9d = 4.16mg/l (1.96 – 7.06 mg/l)  LC 10-9 d = 383.4 µg/l (60.1 - 985 µg/l)  LC 1-9 d = 54.9 µg/l (3.1 – 225 µg/l)	3	flow through; exposure period: 9 days; analytical monitoring	Birge et al., 1980
<i>Rana palustris</i> *		LC 50-8 d = 20.55 mg/l (11.53 - 43.83 mg/l)	3	flow through; exposure period: 8 days; <i>analytical monitoring</i>	Birge et al., 1980

Table A1.3: Acute toxicity results with invertebrates

Species	Method	Endpoint (mg/l)	Reliability index	Remarks	Reference
<i>Panaeus duorarum</i>		LC 50-96 h = 81.5 mg/l	4	Insufficient documentation on test method	US-EPA, 1980
<i>Daphnia magna</i>	static three-brood test, Cowgill & Milazzo, 1989	LC 50-48 h = 353 mg/l	3	No analytical monitoring. Volatility is not sufficiently taken into account  Organisms are fed during the test	Cowgill and Milazzo, 1991
<i>Ceriodaphnia dubia</i>	static three-brood test, Cowgill & Milazzo, 1989	LC 50-48 h = 290 mg/l	3	No analytical monitoring. Volatility is not sufficiently taken into account  Organisms are fed during the test	Cowgill and Milazzo, 1991
<i>Daphnia magna</i>	ASTM subcommittee on safety to aquatic organisms	LC 50-48 h = 65.7 mg/l (geometric mean of 3 results)	3	No analytical monitoring Volatility is not sufficiently taken into account	Gersich, et al., 1986
<i>Daphnia magna</i>	DIN 38412	LC 50 24h = 290 mg/l  LC 0-24 h = 62 mg/l  LC 100-24 h = 500 mg/l	3	No analytical monitoring.  Test system is not appropriate to volatile substances	Knie et al., 1983
<i>Crassostrea virginica</i>		LC 50-48 h = 0.385 mg/l (estimated from a graph)	3	Analytical monitoring at a median concentration: 100µg/L The result is based on a calculated time-weighted mean concentration that is taking into account the loss of chloroform)	Stewart <i>et al.</i> , 1979

Species	Method	Endpoint (mg/l)	Reliability index	Remarks	Reference
<i>Crassostrea gigas</i>		EC 50-48h = 152.5 mg/L NOEC = 50.4 mg/L	1	Analytical monitoring at every tested concentration (48h losses were below 12%). Larvae with incompletely developed shells were counted dead	WRc, 2002
<i>Daphnia magna</i>	US-EPA-660/3, 1975	LC 50-48 h = 29 mg/l	2	No analytical monitoring closed vessels	Le Blanc, 1980
<i>Daphnia magna</i>	Bobra et al., 1983	LC 50-48 h = 79 mg/l	2	No analytical monitoring static closed test, no air-spaces in exposure chambers to minimize volatilisation daphnids 4-5 days old	Abermethyl et al., 1986
<i>Daphnia magna</i>	DIN 38412	LC 50-24 h = 79 mg/l LC 0-48 h = 48 mg/l	2	Nominal concentration static closed test	Kühn et al., 1989
<i>Artemia salina</i>		EC-50-24 h = 31.1 mg/l (25% ASW) EC-50-24 h = 37 mg/l (25% ASW) (immobilisation of stage II nauplii)	2	No monitoring but the volatility is sufficiently taken into consideration ASW = Artificial Sea Water	Foster and Tullis, 1985

Table A1.4: Chronic toxicity results with invertebrates

Species	Method	Endpoint (mg/l)	Reliability	Remarks	Reference
<i>Daphnia magna</i>	static three-brood test, Cowgill & Milazzo, 1989	NOEC = 120 mg/l (mortality, brood size and progeny in 10 days test)	3	No analytical monitoring. Volatility is not sufficiently taken into account. Organisms are fed during the test	Cowgill and Milazzo, 1991
<i>Cerio-daphnia dubia</i>	static three-brood test, Cowgill & Milazzo, 1989	NOEC = 3.4 mg/l (mortality in 9 days test)	3	No analytical monitoring. Volatility is not sufficiently taken into account Organisms are fed during the test	Cowgill and Milazzo, 1991
<i>Daphnia magna</i>	Hermens, 1984	EC 50-16 d = 59.9 mg/l NOEC = 15 mg/l (growth)	2	endpoint : length (uncommon) analytical monitoring	Hermens et al., 1985
<i>Daphnia magna</i>	German Federal Environmental Agency, 1984	NOEC = 6.3 mg/l (21 days test)	1	Analytical monitoring NOEC refers to the parent animal mortality the reproduction rate and the appearance of first offsprings.	Kühn et al., 1989

Table A1.5: Toxicity results with algae

Species	Method	Endpoint	Reliability	Remarks	Reference
<i>Haematococcus pluvialis</i>	Warburg apparatus, 1983	EC 10-4 h = 440 mg/l (reduction of O <sub>2</sub> production)	4	Static test. No analytical monitoring. No indication on volatility consideration	Knier et al., 1983
<i>Skeletonema costatum</i>	EPA	NOEC = 216 mg/l EC 50-5 days = 437-477 mg/l	3	No analytical monitoring Closed bottles Low growth in the controls	Cowgill et al., 1989
<i>Skeletonema costatum</i>	Erickson et al., 1970-1972	EC 50-7 days > 32 mg/l (biomass measured by turbidity)	3	No analytical monitoring Volatility is not sufficiently taken into account	Erickson & Freeman, 1977
<i>Thalassiosira pseudonana</i>	Erickson et al., 1970-1972	EC 50-7 days > 32 mg/l (biomass measured by turbidity)	3	No analytical monitoring Volatility is not sufficiently taken into account	Erickson & Freeman, 1977
<i>Scenedesmus quadricauda:</i>	Concentration of algal suspension is measured turbidimetrically	NOEC = 1100 mg/l (8 days test)	2	No analytical monitoring closed system Determination of the Toxicity Threshold	Bringmann & Kühn, 1977-1980
<i>Microcystis aeruginosa</i>	Concentration of algal suspension is measured turbidimetrically	NOEC = 185 mg/l (8 days test)	2	No analytical monitoring closed system Determination of the Toxicity Threshold	Bringmann & Kühn, 1975-1978
<i>Scenedesmus subspicatus</i>	DIN 38412, Part 9 Concentration of algal suspension is measured turbidimetrically	Biomass : EC 50 48 h = 560 mg/l EC 10 48 h = 225 mg/l Growth rate : EC 50-48h = 950 mg/l EC 10-48h = 360 mg/l	2	No analytical monitoring closed system Validity criteria are fulfilled	Kühn & Pattard, 1990
<i>Chlamydomonas reinhardtii</i>	Modified protocol to provide sufficient CO <sub>2</sub> concentration. Guideline validity criteria are fulfilled	EC 50 72h = 13.3 mg/l EC 10 72h = 3.61 mg/l (biomass)	1	Analytical monitoring, closed system using bipartite vessels	Brack & Rottler, 1994

Table A1.6: Toxicity results with sediment dwelling organisms

Species	Method	Endpoint	Reliability	Remarks	Reference
Methanogenic bacteria (sediment from the estuary of the river Rhine)		EC 10-11 days = 5.5 mg/kg (dw)  EC 50-11 days = 6.9 mg/kg (dw)  (inhibition of methane production)	2	Theoretical toxicant concentration  Sterile incubation closed bottles. No indication of the number of concentration and the final volume of methanol.	van Vlaardingen & van Beelen, 1992