CLH report

Proposal for Harmonised Classification and Labelling

**Based on Regulation (EC) No 1272/2008 (CLP Regulation),
Annex VI, Part 2**

Chemical name:

2,2',6,6'-tetra-*tert*-butyl-4,4'-methylenediphenol

EC Number: 204-279-1

CAS Number: 118-82-1

Index Number: -

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on behalf of the Austrian Competent Authority (Austrian Federal Ministry for Climate Action, Environment, Energy, Mobility, Innovation and Technology. Radetzkystraße 2, 1030 Vienna, Austria)

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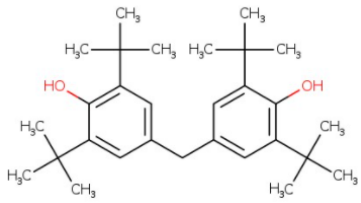
ABBREVIATIONS

AC	Article Category
%AR	Applied Radioactivity
B / vB	Bioaccumulative / very bioaccumulative
BCF	Bioconcentration Factor
BMF	Biomagnification Factor
BOD	Biochemical Oxygen Demand
CAS	Chemical Abstract Service
COD	Chemical Oxygen Demand
d	Day
DS	Dossier submitter
Drg	Danger
DW	Dry Weight
GLP	Good Laboratory Practice
HCB	Hexachlorbenzol
Kow	Partition coefficient octanol/water
LC50	Lethal concentration, 50%
m/f	Male/female
MITI	Ministry of International Trade and Industry, Japan
NER	Non-extractable residues
OECD	Organisation for Economic Co-operation and Development
P / vP	Persistent / very persistent
PC	Product Category
TG	Test Guideline

1 IDENTITY OF THE SUBSTANCE

1.1 Name and other identifiers of the substance

Table 1: Substance identity and information related to molecular and structural formula of the substance

Name(s) in the IUPAC nomenclature or other international chemical name(s)	2,2',6,6'-tetra-*tert*-butyl-4,4'-methylenediphenol
Other names (usual name, trade name, abbreviation)	2,6-di-*tert*-butyl-4-[(3,5-*ditert*-butyl-4-hydroxyphenyl)methyl]phenol Phenol, 4,4'-methylenebis[2,6-bis(1,1-dimethylethyl)- TBMD
ISO common name (if available and appropriate)	-
EC number (if available and appropriate)	204-279-1
EC name (if available and appropriate)	2,2',6,6'-tetra-*tert*-butyl-4,4'-methylenediphenol
CAS number (if available)	118-82-1
Other identity code (if available)	-
Molecular formula	C29H44O2
Structural formula	 (**source:** European Chemicals Agency, http://echa.europa.eu/)
SMILES notation (if available)	CC(C)(C)C1=CC(=CC(=C1O)C(C)(C)C)CC2=CC(=C(C(=C2)C(C)(C)C)O)C(C)(C)C
Molecular weight or molecular weight range	424.7
Information on optical activity and typical ratio of (stereo) isomers (if applicable and appropriate)	-
Description of the manufacturing process and identity of the source (for UVCB substances only)	-
Degree of purity (%) (if relevant for the entry in Annex VI)	-

1.2 Composition of the substance

2,2',6,6'-tetra-*tert*-butyl-4,4'-methylenediphenol (TBMD) is a mono-constituent substance.

Impurities do not contribute to the classification. For further information see confidential annex to this document.

Table 2: Constituents (non-confidential information)

Constituent (Name and numerical identifier)	**Concentration range (% w/w minimum and maximum in multi-constituent substances)**	**Current CLH in Annex VI Table 3 (CLP)**	**Current self-classification and labelling (CLP)**
2,2',6,6'-tetra-*tert*-butyl-4,4'-methylenediphenol EC 204-279-1	CONF	-	Not classified

Detailed information on the test substance (if available) is given in the study descriptions.

2 PROPOSED HARMONISED CLASSIFICATION AND LABELLING

2.1 Proposed harmonised classification and labelling according to the CLP criteria

Table 3: For substance with no current entry in Annex VI of CLP

	Index No	Chemical name	EC No	CAS No	Classification		Labelling			Specific Conc. Limits, M-factors and ATEs	Notes
					Hazard Class and Category Code(s)	Hazard statement Code(s)	Pictogram, Signal Word Code(s)	Hazard statement Code(s)	Suppl. Hazard statement Code(s)		
Current Annex VI entry	No current Annex VI entry										
Dossier submitter's proposal	TBD	2,2',6,6'-tetra-*tert*-butyl-4,4'-methylenediphenol	204-279-1	118-82-1	Aquatic Chronic 1	H410	GHS09 Dgr	H410		M = 10000	

Table 4: Reason for not proposing harmonised classification and status under consultation

Hazard class	Reason for no classification	Within the scope of consultation
Explosives	*hazard class not assessed in this dossier*	No
Flammable gases (including chemically unstable gases)	*hazard class not assessed in this dossier*	No
Oxidising gases	*hazard class not assessed in this dossier*	No
Gases under pressure	*hazard class not assessed in this dossier*	No
Flammable liquids	*hazard class not assessed in this dossier*	No
Flammable solids	*hazard class not assessed in this dossier*	No
Self-reactive substances	*hazard class not assessed in this dossier*	No
Pyrophoric liquids	*hazard class not assessed in this dossier*	No
Pyrophoric solids	*hazard class not assessed in this dossier*	No
Self-heating substances	*hazard class not assessed in this dossier*	No
Substances which in contact with water emit flammable gases	*hazard class not assessed in this dossier*	No
Oxidising liquids	*hazard class not assessed in this dossier*	No
Oxidising solids	*hazard class not assessed in this dossier*	No
Organic peroxides	*hazard class not assessed in this dossier*	No
Corrosive to metals	*hazard class not assessed in this dossier*	No
Acute toxicity via oral route	*hazard class not assessed in this dossier*	No
Acute toxicity via dermal route	*hazard class not assessed in this dossier*	No
Acute toxicity via inhalation route	*hazard class not assessed in this dossier*	No
Skin corrosion/irritation	*hazard class not assessed in this dossier*	No
Serious eye damage/eye irritation	*hazard class not assessed in this dossier*	No
Respiratory sensitisation	*hazard class not assessed in this dossier*	No
Skin sensitisation	*hazard class not assessed in this dossier*	No
Germ cell mutagenicity	*hazard class not assessed in this dossier*	No
Carcinogenicity	*hazard class not assessed in this dossier*	No
Reproductive toxicity	*hazard class not assessed in this dossier*	No
Specific target organ toxicity-single exposure	*hazard class not assessed in this dossier*	No
Specific target organ toxicity-repeated exposure	*hazard class not assessed in this dossier*	No
Aspiration hazard	*hazard class not assessed in this dossier*	No
Hazardous to the aquatic environment	Aquatic Chronic 1, H410	Yes
Hazardous to the ozone layer	*hazard class not assessed in this dossier*	No

3 HISTORY OF THE PREVIOUS CLASSIFICATION AND LABELLING

The substance has no harmonized classification so far.

The substance has 723 C&L notifications. 528 thereof indicate no classification. Other notifiers give following self-classifications (summary): Acute Tox. 4, H312; Skin Irrit 2, H315; Eye Irrit 2, H319; STOT SE 3, H335; Aquatic Chronic 2, H411; Aquatic Chronic 4, H413 [ECHA dissemination site, accessed 03/2022].

4 JUSTIFICATION THAT ACTION IS NEEDED AT COMMUNITY LEVEL

Justification that action is needed at Community level is required.

Based on the available data a classification for environmental toxicity in combination with a high M-factor is indicated. Due to disagreement by the DS with the current self-classification of registrants and notified self-classifications in the C&L inventory a harmonized classification for this endpoint is proposed.

5 IDENTIFIED USES

Table 5: The following uses are indicated at ECHA dissemination site [accessed 03/2022]:

Categories	Use(s)	Technical function
Manufacture	Manufacture of the substance	-
Formulation	Formulation with lubricant additives, lubricants and greases (PC 17, 24, 25) Formulation (PC 1, 9a, 9b, 9c, 14, 16, 17, 20, 21, 24, 25, 27, 28, 35)	-
Uses at industrial sites	Industrial use of lubricants and greases containing additives (antioxidant) (PC 14, 16, 17, 24, 25) Industrial use in construction of aircraft engines Industrial coatings and inks (PC 9a) Washing and cleaning products (PC 35) Plant protection products (PC 27) Intermediate (PC 19)	antioxidant, stabilizer
Uses by professional workers	Professional use of lubricants and greases containing additives (antioxidant) in vehicles and machinery (PC 16, 24) Professional use of coatings and inks (PC 9a) Lubrication of aircraft engines Professional use of washing and cleaning products (PC 35) Professional use of lubricants (PC 17, 24, 25)	antioxidant, stabilizer
Consumer Uses	Consumer use of lubricants (PC 24)	antioxidant,

	General consumer use of lubricants and greases containing additive (antioxidant) (PC 24) Consumer use of coatings and inks (PC 9a) General consumer use of lubricants and greases in vehicles or machinery (PC 24) Consumer use of washing and cleaning products (PC 35)	stabilizer
Article service life	AC 1: Vehicles AC 2: Machinery, mechanical appliances, electrical/electronic articles AC 3: Electrical batteries and accumulators AC 4: Stone, plaster, cement, glass and ceramic articles AC 7: Metal articles	antioxidant, stabilizer

6 DATA SOURCES

The dossier is based on information from ECHA dissemination site (Substance Information - ECHA (europa.eu)) as well as original study reports provided by registrants. In addition, scientific literature served as information source.

Please see Chapter 14 for details.

7 PHYSICOCHEMICAL PROPERTIES

Table 6: Summary of physicochemical properties (ECHA dissemination site, accessed 03/2022]

Property	Value	Reference	Comment (e.g. measured or estimated)
Physical state at 20°C and 101,3 kPa	Solid, powder lite yellow, odourless	ECHA dissemination site	-
Melting/freezing point	156.4°C ±0.1°C	ECHA dissemination site	OECD 102
Boiling point	175°C at 0.48mbar	ECHA dissemination site	OECD 103; decomposition at atmospheric pressure
Relative density	1.06 at 20.6°C	ECHA dissemination site	EU Method A.3
Vapour pressure	2.9 x 10^{-6} Pa at 25°C	ECHA dissemination site	OECD 104
Surface tension	-	ECHA dissemination site	Data waiving
Water solubility	0.032 µg/l at 20°C, pH 7	ECHA dissemination site	OECD 105

Property	Value	Reference	Comment (e.g. measured or estimated)
Partition coefficient n-octanol/water (log K_{ow})	8.99	ECHA dissemination site	Estimate (KOWWIN v.1.68)
Flash point	-	ECHA dissemination site	waiving
Flammability	-	ECHA dissemination site	waiving
Explosive properties	-	ECHA dissemination site	waiving
Self-ignition temperature	-	ECHA dissemination site	waiving
Oxidising properties	-	ECHA dissemination site	waiving
Granulometry	92% < 100µm 2.39% < 5.5 µm	ECHA dissemination site	OECD 110
Stability in organic solvents and identity of relevant degradation products	-	ECHA dissemination site	waiving
Dissociation constant	Strongest pKa (acid): 11 ± 0.4 Strongest pKa (base): No base pKa	ECHA dissemination site	calculation
Viscosity	-	ECHA dissemination site	waiving

8 EVALUATION OF PHYSICAL HAZARDS

Not evaluated in this report.

9 TOXICOKINETICS (ABSORPTION, METABOLISM, DISTRIBUTION AND ELIMINATION)

Not evaluated in this report.

10 EVALUATION OF HEALTH HAZARDS

Not evaluated in this report.

11 EVALUATION OF ENVIRONMENTAL HAZARDS

11.1 Rapid degradability of organic substances

Table 7: Summary of relevant information on rapid degradability

Method	Results	Remarks	Reference
Ready biodegradability			
BIOWIN v4.10 (EPI SuiteTM)	Overall, not readily biodegradable. Biowin 1 = 0.0959 (<0.5) Biowin 2 = 0.0005 (<0.5) Biowin 3 = 1.4501 (<2.5) Biowin 4 = 2.6321 (<2.26 (-2.75)) Biowin 5= -0.1702 (<0.5) Biowin 6 = 0.0014 (<0.5) Biowin 7 = -1.7978 (<0.5) Ready biodegradability prediction: **NO**	The substance is in the applicability domain of the used models, results are considered valid. Biowin 1 and 2: degradation is not fast, as values are below 0.5 Biowin 3 and 4: primary and ultimate degradations are not fast Biowin 5 and 6: not readily biodegradable based on linear and non-linear models Biowin 7: anaerobic degradation is not fast	Modelling, DS
OECD Guideline 301 C (Ready Biodegradability: Modified MITI) GLP-study Test substance: 2, 2´, 6, 6´- tetra-*tert*-butyl-4,4´-methylene-diphenol (TBMD) Purity: 99.6% Initial test substance concentration: 100 mg/L Concentration of activated sludge: 30 mg/L (adaptation not specified) A blank control, positive control (aniline) and TBMD in water were incubated simultaneously.	Not readily biodegradable. -1, 0 and 0% of the initial dose was degraded within 28 days (triplicates) 4, 2 and 2 % TBMD disappearance based on HPLC (triplicates)	Klimisch 2 Test concentration was far above the water solubility. Activated sludge. Adaptation not specified.	Ministry of International Trade and Industry, Japan (2007)
EU Method C.4-C (Determination of the "Ready" Biodegradability – Carbon Dioxide Evolution Tests) GLP-study Test substance: 2, 2´, 6, 6´- tetra-*tert*-butyl-4,4´-methylene-diphenol (TBMD) Purity: 99.39% Initial test substance concentration: 20 mg/L	Not readily biodegradable. 0-2% degradation after 28 days (CO_2 evolution)	Klimisch 2 Test concentration was far above the water solubility. Activated sludge. Adaptation not specified.	Anonymous (1993) (cited from ECHA dissemination site)

Method	Results	Remarks	Reference
An inoculum blank (inoculated mineral medium only) and positive control (sodium benzoate) were included in the test run. Test temperature: 20 ± 1ºC			
Simulation testing			
OECD 307 (Simulation test in aerobic soil) GLP-study Test substance: 2, 2´, 6, 6´- tetra-*tert*-butyl-4,4´-methylene-diphenol (TBMD) Reported radiochemical purity in study report: 98.8% Reported radiochemical purities of stock solutions via personal communication: Soil 2.2: 95.7% Soil 2.3, 2.4, 5M: 91.2% Four soils tested. Radiolabelled test item with 14-C. Incubated in the dark under aerobic conditions. Temperature at 12 ± 2°C. Test duration 120-139 days. Nominal test item concentration: 5.11 kBq/g soil DW corresp. to 1000 µg test item/kg soil DW	Comparable mineralisation rate in all four soils with a max. of 25.3% applied radioactivity (%AR) at the end of the study. Start of mineralisation >1%AR around day 4-6. Rapid primary degradation/adsorption and formation of degradation products at the start of the study, prior to mineralisation.	Klimisch 2 No reference item and no sterile control were included in the study. Radiochemical purity could not be finally clarified No differentiation of NER into type I, II and III for all data points possible based on the analysis performed. Abiotic degradation of the parent TBMD occurred either before or directly at application.	Anonymous (2021)
OECD 314 B (study based on a OECD draft from 2006) (Simulation Test to Assess the Biodegradability of Chemicals Discharged in Wastewater) GLP-study Test substance: 2, 2´, 6, 6´- tetra-*tert*-butyl-4,4´-methylene-diphenol (TBMD) Radiochemical purity: 99.4% Adapted and non-adapted sludge.	Not rapid degradable. Mineralisation was negligible (max 2.8-4%). Rapid primary degradation with occurrence of degradation products that likely tend to be more persistent. Degradation products were not identified.	Klimisch 2 Test conducted above the water solubility. The OECD 314 TG cannot be used for assessing the degradation in the aquatic environment and is therefore not relevant for classification and labelling.	Anonymous (2007)
Others			
Environment Canada 2004. EPS 1/RM/43 Biological Test Method: Tests for Toxicity of Contaminated Soil to Earthworms Degradation of test item was measured by determination of the recovery of the test item at the end of the test.	Not rapid degradable. At test end (day 63) 87 ± 2.4% of the test item was recovered from the test soil.	Klimisch 2 Mineralisation not measured. No degradation products specified.	Ritchie et al. (2013)

Method	Results	Remarks	Reference
Test substance: 2, 2´, 6, 6´- tetra-*tert*-butyl-4,4´-methylene-diphenol (TBMD) Purity: >98% Nominal test concentration: between 0 to 3000 mg/kg dw Test duration: 14 days (plants); 63 days (invertebrates) Incubation at 20 ± 3°C for 16h of light and 8h of dark (invertebrates). Soil type: sandy soil, amended with peat moss Invertebrate species: Earthworms (*Folsomia candida* and *Eisenia andrei*)			
OECD 317 (Bioaccumulation in Terrestrial Oligochaetes) Degradation data of TBMD were reported based on the uptake phase of the bioaccumulation study. Test substance: 2, 2´, 6, 6´- tetra-*tert*-butyl-4,4´-methylene-diphenol (TBMD) Purity: >98% Nominal test concentration: 10 mg/kg dw (clay loam); 50 mg/kg dw (sandy soil) Duration uptake and depuration phase: 28 days each (clay loam); 21 days each (sandy soil) Invertebrate species: Earthworms (*Eisenia andrei*)	Dissipation half-life: 46 days (clay loam) 11 days (sandy soil)	Klimisch 2 Mineralisation not measuered. No degradation products specified.	Princz et al. (2014)

11.1.1 Ready biodegradability

Estimated data

QSAR calculations were performed with BIOWIN v4.10 QSAR contained within EPI SuiteTM (US-EPA, 2011) for TBMD (ref. to Table 7). Biowin consists of seven models. The substance is predicted to not biodegrade fast using linear (Biowin 1) and non-linear (Biowin 2) biodegradation models, as the values are below 0.5. The calculations are valid as the test substance is in the applicability domain of Biowin 1 and 2. TBMD lies in the applicability domain of the models and is considered valid, as the substance is in the molecular range of the training set and many fragments of the substances are covered by the fragments of the training set. Ultimate biodegradation, the conversion from TBMD to CO_2 (Biowin 3), is predicted to be recalcitrant. Initial steps, primary biodegradation are predicted to occur not fast, in weeks to months (Biowin 4). Biowin 5 and 6 represent MITI testing, TBMD was not considered to be readily biodegradable. Under anaerobic conditions (Biowin 7), the test substance is predicted not to quickly biodegrade. The overall prediction of the ready biodegradability is “no”.

Experimental data

A ready biodegradability study was conducted with TBMD according to OECD 301 C (Ministry of International Trade and Industry Japan, 2007). 100 mg of TBMD was incubated with sludge collected from ten different places in Japan for 28 days at 25°C. The test was conducted far above the water solubility of the test substance. The concentration of the activated sludge was 30 mg/L. During incubation, the oxygen consumption was measured to determine the biodegradability. EC TBMD was also analyzed quantitatively by HPLC after 28 days.

The BOD was 0% after 28 days. The quantitative HPLC analysis showed a disappearance of 4, 2, and 2% for the three replicates after 4 weeks. Under the test conditions TBMD is not readily biodegradable. The study is rated as Klimisch 2, as the test concentration was above the water solubility.

A study performed according to the EU Method C.4-C, also called the modified Sturm method (equalling OECD 301B) resulted in 0% degradation of TBMD after 28 days (Anonymous, 1993). It is noted, that the test concentration of 20 mg/L was far above the expected water solubility. The reference compound sodium benzoate was extensively degraded, with net carbon dioxide evolution of 83-86% $ThCO_2$ at the end of the test. As the threshold for ready biodegradability is not met within 28 days, it can be concluded that the test substance is not readily biodegradable. The study is rated as Klimisch 2, as the test concentration was above the water solubility.

Conclusion on rapid degradability

Both experimental ready biodegradation tests and the QSAR estimates do not demonstrate any biodegradation. Therefore, TBMD is considered as not rapidly degradable.

11.1.2 BOD_5/COD

Not evaluated in this report.

11.1.3 Hydrolysis

No hydrolysis data for TBMD are available. But as the substance does not have any functional groups susceptible to hydrolysis in the environment, hydrolysis is therefore not likely to be a significant degradation process.

11.1.4 Other convincing scientific evidence

The study of Ritchie et al. (2013) is not a biodegradation guideline study as such, as it primarily addresses the effect on plant growth and soil invertebrates *Eisenia andrei* and *Folsomia candida* (survival and reproduction). However, the study also investigated the persistency of TBMD in a sandy soil by measuring the recovery at the beginning and at the end of the study. The sandy soil was chosen to optimize chemical recovery and bioavailability to the invertebrates. The soil was amended with *Sphagnum sp.* peat moss to improve the conditions for reproduction.

TBMD was persistent throughout the tests which ranged from 14 days (plants) to 63 days (earthworms) in duration; 87 ± 2.4% were recovered from the test soil on day 63. Under the test conditions, TBMD is considered as not rapidly degradable. The study is rated as Klimisch 2.

Another publication by Princz et al. (2014) investigated the bioaccumulation of TBMD in terrestrial oligochaetes in a sandy soil and clay loam soil. For the uptake and elimination phase the test duration for each phase was 28 days (clay loam soil) or 21 days (sandy soil). Disappearance of TBMD was used to estimate a degradation rate in the uptake phase and subsequently, derive a half-life in soil using first-order kinetics. It is stated, that no significant loss of TBMD was observed in the clay loam soil, nevertheless, a dissipation half-life value of 46 days was derived. In the sandy soil, a significant disappearance with a

dissipation half-life of 11 days was determined. Mineralisation and transformation products were not measured. The study is rated as Klimisch 2.

11.1.4.1 Field investigations and monitoring data (if relevant for C&L)

Not evaluated for this report.

11.1.4.2 Inherent and enhanced ready biodegradability tests

Not evaluated for this report.

11.1.4.3 Water, water-sediment and soil degradation data (including simulation studies)

An aerobic transformation in soil was investigated in a GLP-study according to TG OECD 307 (Anonymous, 2021). Four natural soils representing different soil properties were field fresh sampled, treated with the ^{14}C-labelled test item and incubated in the dark under aerobic conditions at a temperature of 12 ± 2°C for a minimum of 120 days up to 139 days. Nominal test item concentration was 5.11 kBq/g soil DW, corresponding to 1000 µg test item/kg soil DW. The study is considered valid by the DS. However, some uncertainties occurred with the quality criteria, in particular, for three of the four soils (radiochemical purity of stock solution, stability of test item). DS rates this study as Klimisch 2, in particular for soil 2.2 (ref. to Table 7).

In the OECD TG 307 aerobic soil study, mineralisation was comparable among the four soils and reached a maximum of 25.3% AR after 125 days. Rapid primary degradation was observed at the beginning of the study, however, prior to the start of mineralisation. The presence of multiple degradation products without simultaneous mineralisation already at the start of the study, led to the assumption that abiotic degradation occurred. However, it remains unclear, if the abiotic degradation occurred before application or directly at or after the application. In total, 32 transformation products were detected in the four soils. For 21 transformation products accurate masses and molecular formulas were derived. A molecular structure was proposed only for five transformation products and two of them were confirmed by the use of a reference substance. For three transformation products a CAS number could be allocated by the DS: 4359-97-1, 1620-98-0 and 719-22-2 In the CLP notifications there are self-classifications for CAS 1620-98-0 (EC 216-592-0) as Skin Irrit.2 (H315), Eye Irrit. 2 (H319) and STOT SE 3 (H335). CAS 719-22-2 (EC 211-946-0). is self-classified also as Skin Irrit. 2 (H315), Eye Irrit. 2 (H319) and STOT SE 3 (H335). There are no notifications for CAS 4359-97-1.

CLP Annex I, 4.1.2.9.3 states: "*Primary biodegradation does not normally suffice in the assessment of rapid degradability unless it can be demonstrated that the degradation products do not fulfil the criteria for classification as hazardous to the aquatic environment.*" There is no data on aquatic toxicity available for any detected transformation product. As stated above, there is only self-classification for irritating properties for two transformation products. Many transformation products remain unknown and unidentified. Due to the lack of data for the unknown, unidentified and identified degradation products, the need for classification as hazardous to the aquatic environment of the degradation products cannot be excluded, and therefore, TBMD is considered as not rapid degradable.

An OECD 314 B Simulation Test (Anonymous, 2007) to assess the Biodegradability of chemicals discharged in wastewater is available. According to CLP Annex I "*Results from tests simulating the conditions in a sewage treatment plant (STP) […] cannot be used for assessing the degradation in the aquatic environment"*. Therefore, the OECD 314 B study is not relevant for classification and labelling.

Conclusion of biodegradability in soil

In the OECD TG 307 aerobic soil study (Anonymous, 2021) mineralisation was comparable among the four soils and reached a maximum of 25.3% applied radioactivity (%AR) after 125 days. Several unknown and unidentified degradation products occurred prior or shortly after application of radioactive labelled TBMD. Mineralisation started afterwards. No aquatic toxicity data is available for any degradation product, including

for those three degradation products where a CAS number was allocated (CAS 4359-97-1, CAS 1620-98-0 and CAS 719-22-2). Based on this study, TBMD is considered as not rapid degradable.

11.1.4.4 Photochemical degradation

Not evaluated in this report.

11.2 Environmental transformation of metals or inorganic metals compounds

Not relevant for this dossier.

11.3 Environmental fate and other relevant information

Table 8: Summary of relevant information on adsorption coefficient

Method	Results	Remarks	Reference
EU Method C.19 (Estimation of the Adsorption Coefficient (KOC) on Soil and Sewage Sludge Using High Performance Liquid Chromatography (HPLC)) HPLC estimation method GLP-study Test substance: 2, 2´, 6, 6´- tetra-*tert*-butyl-4,4´-methylene-diphenol (TBMD) Purity: 98.48%	Adsorption coefficient: K_{OC}: >427 000 at 30°C log K_{OC}: >5.63 at 30°C	Klimisch 1 Original study not available.	Anonymous (2012c) (cited from ECHA dissemination site)
QSAR model KOCWIN v2.00 (EPI SuiteTM)	Adsorption coefficient: K_{OC}: 36440000 (MCI method) log K_{OC}: 7.562 (MCI method) K_{OC}: 1704000 (Kow method) log K_{OC}: 6.231 (Kow method)	Klimisch 2	Anonymous (2012d)

K_{OC} was measured in an HPLC screening test. The method used was EU Method C.19 (Estimation of the Adsorption Coefficient (K_{OC}) on Soil and Sewage Sludge Using High Performance Liquid Chromatography (HPLC)). The log K_{OC} was >5.6 and the K_{OC} >427 000, indicating that the substance is likely to adsorb to soil and sediment (Anonymous, 2012c).

Log K_{OC} values have been estimated with EPISuiteTM (K_{OC}WIN v2.00) and yielded a log K_{OC} of 7.562 (MCI method) and 6.231 (K_{OW} method) (Anonymous, 2012d). Both estimated values give the indication, that TBMD is likely to adsorb to soil and sediment.

Conclusion on adsorption

TBMD is considered as not mobile.

11.4 Bioaccumulation

Table 9: Summary of relevant information on bioaccumulation

Method	Results	Remarks	Reference
QSAR model KOWWIN v1.68 (EPI SuiteTM)	log Kow = 8.99	Indication for bioaccumulation	Modelling, DS
OECD TG 305 Bioconcentration: Flow-through fish test GLP-study Test substance: 2, 2´, 6, 6´- tetra-*tert*-butyl-4,4´-methylene-diphenol (TBMD) Purity: 99.4% Uptake phase: 35 days Depuration phase: 60 days Fish species: Rainbow trout (*Oncorhynchus mykiss)*	BCF > 500 Result stated by registrants: BCF_{SS} (whole body): 600 L/kg	Klimisch 1 Significant fish growth occurred during the study period. Several uncertainties were observed in the data evaluation and interpretation carried out by the study authors.	Anonymous (2010)
OECD TG 305 Bioconcentration: Flow-through fish test GLP-study Test substance: 2, 2´, 6, 6´- tetra-*tert*-butyl-4,4´-methylene-diphenol (TBMD) Purity: 99.6% Two nominal test concentrations: 0.1 and 0.01 µg/L. Exposure time: 60 – 67 days Depuration time: 54 days Fish species: Common carp (*Cyprinus carpio)*	BCF > 500 BCF_{SS}: 4600 L/kg (at 0.1 µg TBMD/L) BCF_{SS}: 9200 L/kg (at 0.01 µg TBMD/L)	Klimisch 2	Kurume (2006) (in Japanese; partial English translation available)
OECD TG 305: dietary study GLP-study Test substances: 2, 2´, 6, 6´- tetra-*tert*-butyl-4,4´-methylene-diphenol (TBMD); Purity: 99.1% & Hexachlorbenzol (HCB); Purity: 99.7%	Re-calculated growth and lipid corrected kinetic dietary biomagnification factor: BMF_{KgL} (TBMD) = 0.95 BMF_{KgL} (HCB) = 0.55 Derived BCF values: 11.037 – 14.004 L/kg	Klimisch 2 The study was re-evaluated by the DS.	Anonymous (2009) (in Japanese; partial English translation available)

Method	Results	Remarks	Reference
Total uptake duration: ca. 10 days Total depuration duration: 37 days Fish species: Common carp (*Cyprinus carpio*) Feeding: 2x / day (30 min) Amount of food: 3% of body weight/day			

11.4.1 Estimated bioaccumulation

According to the CLP Regulation (EC) No 1272/2008 a log $K_{OW} \geq 4$ is used to indicate a risk for bioaccumulation (ECHA, 2017a). Based on the estimated log K_{OW} of 8.99 (EPISuiteTM, KOWWIN v1.68), a BCF value of 845 L/kg ww was calculated (EPISuiteTM, BCFBAF v3.01) (Modelling, DS).

11.4.2 Measured partition coefficient and bioaccumulation test data

Partition coefficient

Several different log K_{OW} values were reported, all of them being ≥ 4. An OECD TG 117 GLP-study from 2005 using reverse phase HPLC analysis yielded a log K_{OW} of >6.4 (Anonymous, 2005). An older OECD TG 117, HPLC method study derived a log K_{OW} of 7.4 (Anonymous, 1988).

According to registrants, "*extrapolation of the HPLC curve [from the 2005 study] indicates that it is likely that the calculated value [8.99] is correct*" (ECHA dissemination site, accessed: 2022-12). An observed high affinity of the test substance for the stationary phase of the column and the requirement to use 100% solvent for the elution of the sample are stated to be additional indications that the estimated log K_{OW} is more reliable (REACH registration, IUCLID data, 2022). The applicability domain of the OECD TG 117 HPLC method is usually in the range of log K_{OW} 0-6, although an expansion of the method to log K_{OW} 10 is possible in exceptional cases (OECD, 2022). Nevertheless, for extreme log K_{OW}-values (<0, >6) the technical feasibility of the HPLC method is limited. It is concluded that the log K_{OW} of 8.99 based on QSAR estimation is considered most reliable.

Measured bioaccumulation test data

According to the CLP Regulation (EC) No 1272/2008 a "*BCF in fish of ≥ 500 is indicative of the potential to bioconcentrate for classification purposes*" (ECHA, 2017a). As described below, this criteria was exceeded by several experimental studies.

A bioconcentration study on fish (OECD 305), requested by the EU PBT Working Group, was carried out with rainbow trout (*Oncorhynchus mykiss*) according to the OECD TG 305 with 14C-labelled substance (radiochemical purity 99.4%) (Anonymous, 2010). Due to analytical limitations (low water solubility), only one test concentration at 0.025 µg/L was used. Steady-state BCF value based on parent TBMD concentrations was 600 L/kg in whole fish tissue. Kinetic BCF value based on total radioactivity was 1265 L/kg in whole fish tissue. Therefore, the CLP criteria is fulfilled. The registrants consider the steady-state estimates of the BCF to be the most accurate of the available BCF estimates from this study.

The DS rates the study as Klimisch 1. Nevertheless, several uncertainties were observed in the data evaluation and interpretation carried out by the study authors. The modelled depuration curve does not fit well with the data itself. Furthermore, there was significant fish growth (from 5.88 g to around 16 g) during the study period. DS evaluated, that the growth rate constant is close to the overall depuration rate constant,

indicating that growth dilution is the main 'depuration' process. Therefore, the kinetic BCF corrected for growth is preferred over the steady-state BCF. The ECHA Guidance - Chapter R.11: PBT/vPvB assessment specifically states that growth dilution and lipid normalisation to 5% should be applied in the B assessment (ECHA, 2017b). Consequently, the BCF value used in the PBT assessment will be considerably higher than the above stated 600 L/kg which is sufficient for CLP classification and labelling purposes.

A bioconcentration test of TBMD was conducted in accordance with the following test methods: OECD TG 305 Bioconcentration - Flow-through fish test (Kurume, 2006). Test description and results are available, however, detailed raw data are not available. DS rates the study as Klimisch 2. The bioconcentration test (Kurume, 2006) consisted of a 67-day (at 0.1 µg/L tests concentration) and 60-day (at 0.01 µg/L test concentration) uptake phase and a 54-day depuration phase. Two nominal concentrations of 0.1 and 0.01 µg/L of TBMD were used to expose the test organisms. Measured test substance concentration ranged between 0.06 to 0.109 µg/L (for the nominal concentration at 0.1 µg/L) and between 0.0059 to 0.0097 µg/L (for the nominal concentration at 0.01 µg/L). The water used in the study had a pH of 7.8. At the start of the exposure the dissolved oxygen was 8.1 mg/L and the temperature was 24.5 °C. For each test concentration 10 fish (common carp *Cyprinus carpio*) were used, with a length ranging between 7 and 11.5 cm. The age of the fish was 1 year and the lipid content of fish was 3.19% at the start and 4.17% at the end of the test.

The concentration at the steady state was within the ± 20% of the measured value. The steady-state bioconcentration factor (BCFss) values were determined from the mean tissue concentrations at apparent steady-state divided by the average water concentration and revealed at 0.1 µg TBMD/L a BCFss value of 4600 L/kg and at 0.01 µg TBMD/L a BCFss value of 9200 L/kg. Corrected for the lipid content the BCFss was 9000 for the higher concentration. Kinetic BCF values cannot be calculated because data on carp length are not reported.

A supporting dietary study on fish was conducted with Hexachlorbenzol (HCB) and TBMD (Anonymous, 2009). HCB and TBMD were dissolved in pollock cod-liver oil and then mixed well with formula feed for rearing carp fry (> 3% lipid content and > 43% protein content). The analysis of TBMD in the diet gave a recovery rate of 102%. The nominal concentration of the test substance in the diet was 100 µg/g and the average measured concentration was 95.2 µg/g. HCB and TBMD did not dissolve in water when fed to fish. The intestines were analyzed on day 1 and 14 (0 and 13) of the depuration phase. The recovery rate in fish was 76.9% for TBMD.

The derived endpoint was a dietary biomagnification factor ($BMF_{dietary}$) for TBMD and HCB. As several deficiencies were noticed in the evaluation carried out by the study authors, the study was re-evaluated by the DS. Deficiencies included, for instance, the usage of an incorrect formula for the calculation of the assimilation efficiency, an incorrect determination of the start of the depuration phase and no correction of the depuration rate constant for growth dilution. **DS rates the study as Klimisch 2.**

The growth corrected depuration rate constant $k_{2g} = k_2 - k_g = 0.0495 - 0.0231$ is 0.0264 d^{-1}. The recalculated assimulation efficiency following equation A7.1 from OECD 305 and a fish growth adjusted feeding rate ($I_G = (0.03 \times 3.27)/4.37 = 0.022$ g× $g^{-1} \times day^{-1}$) yields $\alpha_G \cong 0{,}29$. The growth and lipid corrected $BMF_{KgL} = [(I_G \times \alpha_G)/k_{2g}]/L_{average} = [(0.022 * 0.29)/0.0264]/0.347$ is 0.95. At the same time, the recalculated BMF_{KgL} for HCB is **0.55**. According to the ECHA Guidance R.7c BCF values can be derived from dietary data, by calculating the uptake rate constant following Sijm et al. 1995 ($k_U = (520 \pm 40) \times W^{-0.32 \pm 0.03}$) (ECHA, 2017c). This yields a range of BCF between 11.037 – 14.004 L/kg.

Conclusion on bioaccumulation

All reported log K_{OW} values were ≥ 4, indicating a potential for bioaccumulation. There are two available BCF studies (Anonymous, 2010; Kurume, 2006) that both derive BCF values > 500 L/kg. A supporting dietary study (Anonymous, 2009) yielded a BMF value of 0.95, which is higher than the BMF for hexachlorbenzol analysed within the same study. It is concluded, that the substance TBMD has a potential to bioaccumulate in aquatic environments.

11.5 Acute aquatic hazard

Table 10: Summary of relevant information on acute aquatic toxicity

Method	Species	Test material	Results[1]	Remarks	Reference
Fish					
Industrial waste water test methods, acute toxicity test in fish (equivalent or similar to OECD TG 203 (Fish, Acute Toxicity Test)) Test performed according to GLP Test concentration: 0.15 mg/L (nominal); no analytical verification of test concentration Semi-static conditions Observed endpoint: mortality Test temperature: At start 24.5 °C Before changing water 25-25.1 °C Dissolved oxygen concentration: At start: 8.1 mg/L Before changing water 6.2-6.4 mg7L pH: At start 7.8 Before changing water 7.7-7.8	*Oryzias latipes* (rice-fish)	TBMD Purity ≥ 99.6 %	LC_{50} (96 h): > water solubility limit of 0.032 µg/L (LC_{50} (96 h) according to study authors: > 0.15 mg/L, nominal)	Klimisch 2 (reliable with restrictions) Toxicity screen performed for BCF study Test concentration > water solubility Analytical measurement of test concentration not performed The study is considered reliable with restrictions that up to water solubility no toxicity was observed	Kurume (2006) (in Japanese; partial English translation available)
U.S. Environmental Protection Agency (1975). Methods for acute toxicity testing with fish, macro-invertebrates and amphibians. EPA-660/3-75-009. Semi-static conditions Test concentration:10, 30, 100, 300 and 1000	*Salmo gairdneri* (rainbow trout)	TBMD (Ionox 220) Purity ≥ 99.04 %	LC_{50} (96 h) > water solubility of 0.032 µg/L (LC_{50} (96 h) according to study authors: 820 mg/L, nominal)	Klimisch 2 (reliable with restrictions) TBMD visibly not soluble at concentrations > 10mg/L All test concentrations > water solubiity No analytical measurement of test	Anonymous (1988)

Method	Species	Test material	Results[1]	Remarks	Reference
mg/L (nominal); no analytical verification of test concentration Observed endpoint: mortality Test temperature at highest concentration: 13-17°C Concentration of dissolved oxygen at highest concentration: 9.8 – 10.4 mg/L pH concentration: 7.7 – 8.3				concentration, but the study is considered reliable with restrictions for the fact that up to water solubility no toxicity was observed	
Invertebrates					
OECD 202 (1984): 48h Acute Immobilisation Test Test performed according to GLP Static conditions Test concentration: 0.10 mg/L (nominal); no analytical verification of test concentration Observed endpoint: Immobilisation Test temperature: 21.0°C Dissolved oxygen concentration: 8.0 – 8.2 mg/L pH: 8.0	*Daphnia magna*	TBMD (H4702) Purity of the test material not reported	EC_{50} (48 h): > water solubility of 0.032 µg/L (LC_{50} (96 h) according to study authors: > 0.10 mg/L, nominal)	Klimisch 2 (reliable with restrictions) The used test concentration was above water solubility, but the study is considered reliable with restrictions for the fact that up to water solubility no toxicity was observed	Anonymous (2001)
U.S. Environmental Protection Agency (1975). Methods for acute toxicity testing with fish, macro-invertebrates and amphibians. EPA-660/3-75-009. Static-conditions	*Daphnia magna*	TBMD (Ionox 220) Purity ≥ 99.04 %	EC_{50} (48h): > water solubility of 0.032 µg/L (LC_{50} (96 h) according to study authors: > 1000 mg/L, nominal)	Klimisch 2 (reliable with restrictions) TBMD was visibly not soluble at concentrations > 10 mg/L All test concentrations were above water solubility, but the study is considered reliable with	Anonymous (1988)

Method	Species	Test material	Results[1]	Remarks	Reference
Test concentration: 500 and 1000 mg/L (nominal); no analytical verification of test concentration Observed endpoint: Immobilisation Test temperature: 18-22°C Concentration of dissolved oxygen at highest concentration: 9.0 – 9.2 mg/L pH at highest concentration: 7.8 – 8.0				restrictions for the fact that up to water solubility no toxicity was observed	
Algae					
OECD Guideline 201 (2006): Freshwater Alga and Cyanobacteria, Growth Inhibition Test Test performed according to GLP Observed endpoint: growth rate, yield, cell density Test concentrations: 1.99; 3.99; 7.98; 16.0; 31.9 ng/L (nominal); analytical verification of test concentration Test temperature: 24.3-24.9 °C pH: 7.1-8.5	*Raphidocelis subcapitata* (formerly known as *Pseudokirchneriella subcapitata*)	U-^{14}C-labelled TBMD Purity ≥ 99.8 %	EC_{50} (96 h): > 12.53 ng/L test mat. (mean measured based on geometric mean) based on: cell density E_rC_{50} (96 h): > 12.53 ng/L test mat. (mean measured based on geometric mean) based on: growth rate E_yC_{50} (96 h): > 12.53 ng/L test mat. (mean measured based on geometric mean) based on: yield	Klimisch 2 (reliable with restrictions) Test concentrations measured was very low at the end of the test, implying a loss of the substance. No explanation for the loss was provided by the study authors.	Anonymous (2012a)
Miller, W. E. and	*Scenedesmus*	TBMD	EC_{50} (96 h):	Klimisch 2 (reliable with	Anonymous

Method	Species	Test material	Results[1]	Remarks	Reference
Green, J . C. (1978) . The Selenastrum capricornutum (Prinz) algal bottle test. EPA-600/ 9-78-018 Observed endpoint: Chlorophyll a concentration Test concentration: 1 – 1000 mg/L; no analytical verification of test concentration Test temperature: 22.0-27.6 °C pH at highest concentration : 7.7-7.8	*capricornutum*	(Ionox 220) Purity ≥ 99.04 %	> water solubility of 0.032 µg/L (LC50 (96 h) according to study authors: > 1000 mg/L, (nominal)	restrictions) TBMD visibly not soluble at concentrations > 10 mg/L All test concentrations were above water solubility, but the study is considered reliable with restrictions for the fact that up to water solubility no toxicity was observed	(1988)

[1] Indicate if the results are based on the measured or on the nominal concentration

11.5.1 Acute (short-term) toxicity to fish

A toxicity screen was performed for a BCF study with TBMD and the test organism *Oryzias latipes* according to a method which is equivalent or similar to OECD Guideline 203 (Kurume, 2006). Ten fish were exposed to a nominal test concentration of 0.15 mg/L of TBMD over a period of 96 hours. Prior to the test start the fish were examined visually and any abnormal animals were removed. Further, the test organisms were immersed in a drug bath (oxytetracycline hydrochloride, sodium chloride, formalin) to prevent disease and to eradicate parasites and acclimatized in running water. An analytical measurement of the test concentrations was not performed.

Results: No mortality was observed up to 0.15 mg/L (nominal), which is above the water solubility. Sub-lethal effects were not evaluated. The study was rated as reliable with restrictions.

The acute toxicity of TBMD to the organism *Salmo gairdneri* in a semi-static test was determined (Anonymous, 1988). The study was performed according to U.S. Environmental Protection Agency, methods for acute toxicity testing with fish, macro-invertebrates and amphibians. Ten fish were exposed per test concentration with a daily re-newel of the test solution. The study duration was 96 hours. The nominal test concentrations were 10, 30, 100, 300 and 1000 mg/L. The fish length ranged from 3.0 to 4.3 cm, and the weight ranged from 0.39 to 1.07 g. Prior to the test the organisms were acclimated to the test conditions for more than 10 days. Aeration occurred during the test. There was no analytical monitoring performed and the test substance was visibly not soluble at concentrations > 10 mg/L. Every 24 hours the fish were observed and the number of fish exhibiting toxic symptoms were recorded. Dechlorinated mains water was included as negative control.

Results: The study resulted in a 96-hour-LC_{50} of 820 mg/L (nominal) for *Salmo gairdneri* which is above the water solubility. The study is assessed as reliable with restrictions.

Conclusion

According to the available experimental data, there is no indication that TBMD comprises acute toxicity towards fish up to its solubility limit of 0.032 µg/L.

11.5.2 Acute (short-term) toxicity to aquatic invertebrates

A short term toxicity test with the test organism *Daphnia magna* was conducted according to OECD Guidline 202 under GLP conditions (Anonymous, 2001). Following a range finding study, forty daphnids (4 replicates with 10 animals each) were exposed to a nominal test concentration of 0.10 mg/L of TBMD. The test material was prepared using a preliminary solution in dimethylformamide. Immobilisation or adverse effects were recorded after 24 and 48 hours. A negative control (reconstituted water) and solvent control (dimethylformamide) was included. The actual concentration, homogenicity and stability of the test material in the test solutions was not determined for the purpose of this study. The test solutions were not renewed during the exposure period.

Results: No immobilisation was observed in the treatment goups and control groups after 48 hours neither in the range finding test nor in the definity study. The 48h-EC_{50} value is > 0.10 mg/L (nominal) for *Daphnia magna*. The test media showed to be clear colourless solutions throughout the study duration. The study is rated as reliable with restrictions.

In another 48-hour static acute toxicity test with the test organism *Daphnia magna* (Anonymous, 1988) ten daphnides were exposed per test concentration. The study was conducted according to U.S. Environmental Protection Agency (1975), methods for acute toxicity testing with fish, macro-invertebrates and amphibians (EPA-660/3-75-009). The nominal test concentrations were 500 and 1000 mg/L. After 24 and 48 hours the number of immobilised daphnids was counted. There was no analytical monitoring performed, and the test substance was visibly not soluble at both test concentrations used. A negative control with reconstituted fresh water was included.

Results: At the end of the test one daphnid was immobile at the highest test concentration after 24 and 48 hours. The 48h-EC_{50} is > 1000 mg/L (nominal) for *Daphnia magna* which is above the water solubility. The study is rated as reliable with restrictions.

Conclusion

According to the available experimental data, there is no indication that TBMD comprises acute toxicity towards aquatic invertebrates up to its solubility limit of 0.032 µg/L.

11.5.3 Acute (short-term) toxicity to algae or other aquatic plants

A toxicity test with TBMD and the freshwater alga *Raphidocelis subcapitata* according to OECD TG 201 is available (Anonymous, 2012a). *Raphidocelis subcapitata* was exposed to five test concentrations of U-^{14}C-labelled TBMD (nominal: 1.99; 3.99; 7.98; 16.0; 31.9 ng/L) for a study duration of 96 hours. Measured concentrations were determined at the beginning (1.5, 3.35, 6.84, 12.9 and 26.5 ng/L) and at the end of the test, after 96 hours (0.47, 0.67, 1.29, 2.49, and 5.92 ng/L). At day 0 measured concentrations ranged from 75 to 82.8% of the nominal concentration, but at the end of the test only 15.6 to 23.7% of the test substances were observed. The reason for the loss was not discussed within the report. The geometric mean concentrations during exposure were 0.84, 1.5, 2.97, 5.67 and 12.53 ng/L. A negative control (culture medium) and a solvent control [0.1 mL/L HPLC-grade dimethylformamide (DMF)] were included for 96 hours. The concentration of DMF was the same in all treatment groups. Effects were evaluated based on cell density, yield and growth rate. Inoculum used had 10,000 cells/mL. Flocculation and aggregation of cells were not observed.

Results: The negative and solvent controls were compared. No statistically significant differences were seen between the negative and solvent control data (students t-test (α = 0.05)). The following table gives an overview about the mean cell density and the respective inhibition in the different treatment groups compared to the negative control.

Table 11: Overview on the mean cell density and the percent of inhibition in the treatment groups (Anonymous, 2012a).

Geometric mean	24 hours	48 hours	72 hours	96 hours

concentrations (ng/L)	Mean cell density (cells(mL))	Inhibition (%)	Mean cell density (cells(mL))	Inhibition (%)	Mean cell density (cells(mL))	Inhibition (%)	Mean cell density (cells(mL))	Inhibition (%)
Negative control	56333	-	493333	-	1885000	-	3816667	-
Solvent control	71333	-	486667	-	1846667	-	4093333	-
0.84	65000	-15	363333	26	1610000	15	4713333	-23
1.5	58000	-3	343333	30	1736667	8	4646667	-22
2.97	63667	-13	283333	43	960000	49*	3373333	12
5.67	100667	-79	616667	-25	2053333	-9	4180000	-10
12.53	87667	-56	593333	-20	1940000	-3	4006667	-5

*Statistically significant reduction from the negative control mean using Dunnett's one-tailed t-test ($p \leq 0.05$)

After 72 hours there was a statistically significant reduction of the mean cell density in the 2.97 ng/L treatment group when compared to the negative control (Dunnett's one-tailed t-test ($p \leq 0.05$)). After study duration of 96 hours there was no statistically significant reduction of the mean cell density when comparing the treatment groups to the negative control (Dunnett's test; $p > 0.05$).

The following Table 12 shows the mean growth rate per hour and the respective inhibiton in the different treatment groups compared to the negative control.

Table 12: Overview on the mean growth rate per hour and the percent of inhibition in the treatment groups (Anonymous, 2012a).

Geometric mean concentrations (ng/L)	0-24 hours		0-48 hours		0-72 hours		0-96 hours	
	Mean growth rate (hours)	Inhibition (%)	Mean growth rate (hours)	Inhibition (%)	Mean growth rate (hours)	Inhibition (%)	Mean growth rate (hours)	Inhibition (%)
Negative control	0.0705	-	0.0803	-	0.0724	-	0.0618	-
Solvent control	0.0791	-	0.0799	-	0.0722	-	0.0626	-
0.84	0.0770	-9	0.0747	7	0.0696	4	0.0639	-3
1.5	0.0729	-3	0.0735	9	0.0714	1	0.0640	-4
2.97	0.0748	-6	0.0696	13	0.0626	13*	0.0606	2
5.67	0.0961	-36	0.0854	-6	0.0740	-2	0.0628	-2
12.53	0.0903	-28	0.0850	-6	0.0730	-1	0.0624	-1

*Statistically significant reduction from the negative control mean using Dunnett's one-tailed t-test ($p \leq 0.05$)

After 72 hours the 2.97 ng/L treatment group revealed a statistically significant reduction of the mean growth rate when comparing to the negative control (Dunnett's one-tailed t-test ($p \leq 0.05$)). At study end no statistically significant reduction of the mean growth rate was seen when comparing the treatment groups to the negative control (Dunnett's test; $p > 0.05$).

In Table 13 the mean yield and the percent of inhibition is shown.

Table 13: Overview of the mean yield and the precent of inhibition in the treatment groups (Anonymous, 2012a).

Geometric mean concentrations (ng/L)	72 hours		96 hours	
	Mean cell density (cells(mL))	Inhibition (%)	Mean cell density (cells(mL))	Inhibition (%)
Negative control	1875000	-	3806667	-

Solvent control	1836667	-	4083333	-
0.84	1600000	15	4703333	-24
1.5	1726667	8	4636667	-22
2.97	950000	49*	3363333	12
5.67	2043333	-9	4170000	-10
12.53	1930000	-3	3996667	-5

*Statistically significant reduction from the negative control mean using Dunnett's one-tailed t-test ($p \leq 0.05$)

After 72 hours there was a statistically significant reduction of the mean yield in the 2.97 ng/L treatment group compared to the negative control (Dunnett's one-tailed t-test ($p \leq 0.05$)). At test end no statistically significant reduction of the mean yield was revealed (Dunnett's test; $p > 0.05$). As there was no statistically significant reduction in cell density, yield and growth rate in the treatment groups compared to the control (Dunnett's test; $p > 0.05$) after 96 hours the 96-hour EC_{50} for cell density, E_rC_{50} and E_yC_{50} values were all > 12.53 ng/L (based on geometric mean measured concentration). As the statistically significant reduction in cell density, growth rate and yield was only seen in the 2.97 ng/L treatment group after 72 hours this reduction was not considered to be concentration-responsive and treatment related. The study is rated as reliable with restrictions.

The acute toxicity of TBMD to the freshwater alga *Raphidocelis subcapitata* was investigated in a 4 day growth test (Anonymous, 1988). The study was performed according to the *Selenastrum capricornutum* (Prinz) algal bottle test (EPA-600/ 9-78-018). The nominal test concentrations ranged from 1 to 1000 mg/L of TBMD. Acetone was used as solvent (adjusted to 0.1 ml/L in each flask including controls). Each flask was inoculated with an initial concentration of 500 cells/mL of the test organism. Test flasks containing only culture medium served as negative control. The flasks were incubated under constant illumination for 4 days. After the incubation time the algal biomass was estimated by determination of the concentration of chlorophyll a. No analytical monitoring was performed and the test substance was visibly not soluble at concentrations > 10 mg/L.

Results: After 96 hours the chlorophyll a concentration in the treatment groups was higher than the mean concentration in the controls. The 96h-EC_{50} was > 1000 mg/L (nominal), the highest concentration tested which is above the water solubility. The study is assessed as reliable with restrictions.

Conclusion

According to the available experimental data, there is no indication that TBMD comprises acute toxicity towards algae up to its solubility limit of 0.032 µg/L.

11.5.4 Acute (short-term) toxicity to other aquatic organisms

No data available.

11.6 Long-term aquatic hazard

Table 14: Summary of relevant information on chronic aquatic toxicity

Method	Species	Test material	Results[1]	Remarks	Reference
Fish					
QSAR estimation with ECOSAR v.1.11 using class "Phenols, Poly" and "Neutral Organic SAR" Input parameter:	Fish	TBMD	ChV: ≥ 0.0003 mg/L based on: estimate QSAR ECOSAR class " Phenols, Poly" ChV: ≥ 0.0000383 mg/L based on: estimate QSAR	Klimisch 3 (not reliable) Used logKow of 8.99 is above the endpoint specific logKow cut-off values for both classes.	Anonymous (2013)

Method	Species	Test material	Results[1]	Remarks	Reference
Log Kow: 8.993 (EPISuite Kowwin v1.68 Estimate) Water solubility: 0.0000319 mg/L (user entered)			ECOSAR class "Neutral Organic SAR"		
Invertrebrates					
OECD 211 (2012): *Daphnia magna* Reproduction Test Duration: 21 d Flow-through system Test performed according to GLP Nominal test concentrations (ng/L): 1.6, 3.1, 6.3, 13, 25, 50; analytical verification of test concentration Radiochemical concentration of the test substance: 968 µCi/mL Test temperature: 20.0 ± 1.0 °C Dissolved oxygen concentration: 7.0 – 8.3 mg/L (≥ 62% of saturation) pH: 8.0 – 8.2	*Daphnia magna*	^{14}C-labelled TBMD (AN-2) Purity of the test material not reported	21d-NOEC for growth: 0.0000014 mg/L (1.4 ng/L, mean measured concentration)	Klimisch 2 (reliable with restrictions) Key study Mortality in the negative control is higher than recommended by the OCED TG, probably due to a handling mistake. The amount of feeding exceeds the recommended amount. Missing output expressed as total number of living offspring produced by parent. This calculation was done by DS.	Anonymous (2012b)
Algae					
OECD Guideline 201 (2006): Freshwater Alga and Cyanobacteria, Growth Inhibition Test Test performed according to GLP Observed endpoint: growth rate, yield,	*Raphidocelis subcapitata* (formerly known as *Pseudokirchneriella subcapitata*)	U-^{14}C-labelled TBMD Purity ≥ 99.8 %	NOEC (96 h): ≥ 12.53 ng/L test mat. (mean measured based on geometric mean) based on: cell density NOEC (96 h): ≥ 12.53 ng/L test mat. (mean measured based on geometric	Klimisch 2 (reliable with restriction)	Anonymous (2012a)

Method	Species	Test material	Results[1]	Remarks	Reference
cell density Test concentrations: 1.99; 3.99; 7.98; 16.0; 31.9 ng/L (nominal); analytical verification of test concentration Test temperature: 24.3-24.9 °C pH: 7.1-8.5			mean) based on: growth rate NOEC (96 h): ≥ 12.53 ng/L test mat. (mean measured based on geometric mean) based on: yield		
Miller, W. E. and Green, J . C. (1978). The Selenastrum capricornutum (Prinz) algal bottle test. EPA-600/ 9-78-018 Observed endpoint: Chlorophyll a concentration Test concentration: 1 – 1000 mg/L; no analytical verification of test concentration Used Test concentration > water solubility Test temperature: 22.0-27.6 °C pH at highest concentration: 7.7-7.8	*Raphidocelis subcapitata* (formerly known as *Selenastrum capricornutum*)	TBMD (Ionox 220) Purity ≥ 99.04 %	NOEC (96 h): ≥ 1000 mg/L test mat. (nominal)	Klimisch 2 (reliable with restrictions) TBMD visible not soluble at concentrations > 10 mg/L All test concentrations > water solubility	Anonymous (1988)

11.6.1 Chronic toxicity to fish

There are no experimental long-term toxicity data available for either marine or freshwater fish for TBMD. In the registration dossier results from QSAR calculations were available (Anonymous, 2013). The model revealed a ChV of ≥ 0.0003 mg/L for the class of "Phenols, Poly" and a ChV of ≥ 0.0000383 mg/L for the class "Neutral Organic SAR". The model predicts therefore a toxicity level above the solubility limit. As the used logKow of 8.99 is above the endpoint specific logKow cut-off values (8.0 for both classes) no effects at saturation are expected for those endpoints. Therefore, the results are not considered to be useable for classification.

11.6.2 Chronic toxicity to aquatic invertebrates

A GLP-study according to OECD Guideline 211 was performed (Anonymous, 2012b). *Daphnia magna* was exposed to six test concentrations of ^{14}C-labelled TBMD (mean measured: 1.4; 2,4; 5,5; 12; 21; and 43 ng/L) under flow-through test conditions for 21 days and the effects on survival, growth (length and dry weight) and reproduction were determined for first generation daphnids. In total 20 daphnids (two replicates with 2 compartments containing 5 daphnids) were tested in each treatment and control group. A negative control (dilution water) and a solvent control [0.025 mL/L HPLC-grade dimethylformamide (DMF)] were included. The concentration of DMF was the same in all treatment groups. Two replicates were available for each test concentration and control. The limit of quantification was 0.873 ng/L. Daphnids were fed 2-3 times per day up to and including day 7 and then were fed four times per day until the last day of the study. The daphnids were fed a mixture of yeast, cereal grass media, trout chow and a suspension of the freshwater alga *Raphidocelis subcapitata*. Observations of each first-generation daphnid were made daily during the test. The presence of eggs in the brood pouch, aborted eggs, males or ephippia also were recorded daily. The body length and dry weight of each surviving first generation daphnid were measured at the end of the test.

Results:

Samples for analytical measurement were taken on day 0, 7, 14 and 21. As the analytical result of the sample at 1.6 ng/L treatment group was 177% of recovery on day 21 an additional sample was collected to confirm the test concentration on day 22. Therefore, the result of the sample for the 1.6 ng/L treatment group collected on day 21 was excluded from the calculation of mean measured test concentrations.

Table 15: Mean measured concentrations over 21 days of TBMD and percent of nominal (Anonymous, 2012b).

Nominal concentration (ng/L)	Mean measured concentration (ng/L)	Mean percent of nominal (%)
1.6	1.4	88
3.1	2.4	77
6.3	5.5	87
13	12	92
25	21	84
50	43	86

No significant differences between the control groups were found for any parameter tested ($p > 0.05$). Therefore, the control data for all parameters were pooled for comparison with the treatment groups. The results for adut survival and mean number of neonates per reproductive day are presented in Table 16.

Table 16: Summary table of survival and reproduction (Anonymous, 2012b).

Mean measured concentration (ng/L)	Adult survival (%)	Mean no. neonates / reproductive day ± standard deviation
Pooled Control	85	8.4 ± 1.0
Negative Control	75	8.1 ± 0.53
Solvent Control	95	8.7 ± 1.3
1.4	90	8.9 ± 1.2
2.4	90	6.7 ± 1.1
5.5	100	8.1 ± 0.74

12	95	6.9 ± 1.1
21	90	7.7 ± 1.7
43	90	8.3 ± 1.6

The % adult survival in the treatment groups ranged from 90 to 100%. In the solvent control 95% adults survived and in the negative control only 75% (probably due to an handling error, please find details below under the point "Mortality in negative control"). There was no statistically significant difference in survival between the negative and solvent control groups, therefore the pooled control was used for comparison with the treatment groups. The survival in the pooled control was 85%. There was no statistically significant difference between the treatment groups and the pooled control (Fisher's Exact test, $p > 0.05$).

The mean number of neonates per reproductive day ranged from 6.7 (2.4 ng/L) to 8.9 (1.4 ng/L) in the treatment groups. In the negative control and solvent control the number was 8.1 and 8.7 respectively. As there was no statistically significant difference in reproduction between the negative and solvent control the pooled control was used for comparison with the treatment groups. The mean number of neonates / reproductive day in the pooled control was 8.4. No statistical significant difference was revealed between the pooled control and the treatment groups (Dunnet's test, $p > 0.05$).

No males or ephippia were produced during the study duration. The first day of brood in the control groups and treatment groups was on day 7, 8 or 9. Aborted eggs or shed eggs were not present in any of the control or treatment groups. The following table gives an overview on the observations of pale daphnids at different test concentrations.

Table 17: Summary table of pale daphnids (Anonymous, 2012b).

Mean measured conc. (ng/L)	Total individuals (n)	Pale individuals (n)	Pale individuals (%)
Negative Control	15	0	0
Solvent control	20	1*	5*
1.4 ng/L	20	3	15
2.4 ng/L	20	2	10
5.5 ng/L	20	0	0
12 ng/L	20	2	10
21 ng/L	20	5	25
43 ng/L	20	7	35

* Handling mistake: trapped on the screen or to the silicone sealant in the test compartment

At the end of the test approximately 35% of surviving first-generation daphnids in the 43 ng/L treatment group appeared to be pale in comparison with the control organisms (Figure 1). In every case immobility of the daphnids occurred when discoloration was observed before. Therefore discolouration seems to indicate an adverse effect on the organisms. A prolongation of the testing period may have revealed a higher mortality in the two highest treatment groups as the most pale individuals occurred at these concentrations.

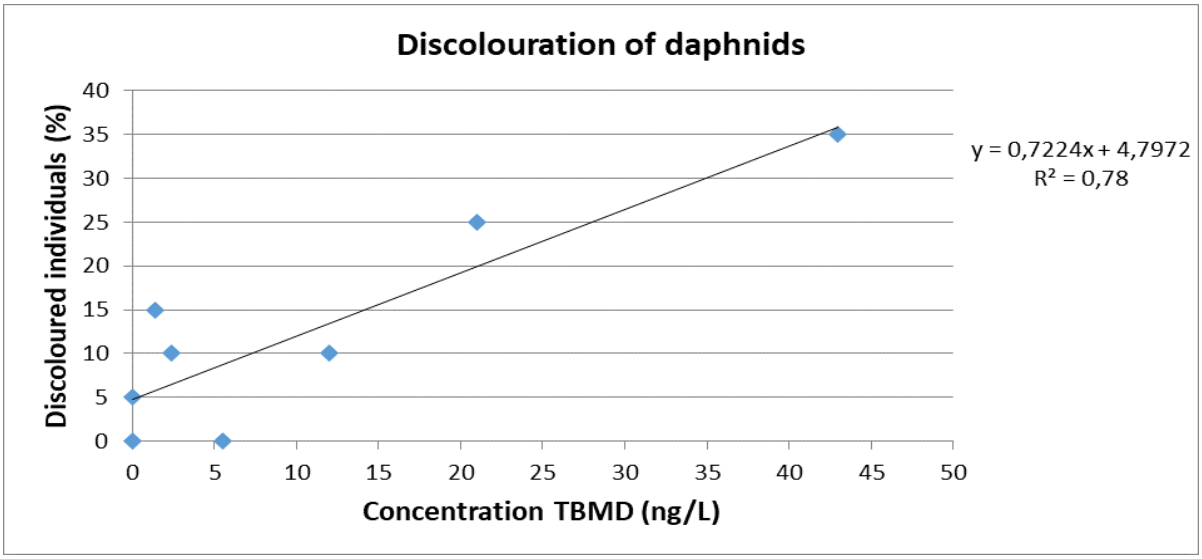


Figure 1: Discolouration of Daphnids at different test concentrations

Table 18: Summary table growth (length in mm) of first generation *Daphnia magna* (Anonymous, 2012b).

Mean measured concentration (ng/L)	Mean body length (mm)	Standard deviation	Individuals (n)	Reduction (%)
Pooled control	4.55	0.12	34	-
Negative control	4.57	0.14	15	-
Solvent control	4.54	0.10	19	-
1.4	4.52	0.14	18	0.66
2.4	4.32	0.13	18	5.05 (*); *
5.5	4.41	0.12	20	3.07 (*); *
12	4.37	0.14	19	3.95 (*); *
21	4.43	0.14	18	2.63 (*); *
43	4.42	0.16	18	2.85 *

(*)Statistically significant effect (Dunnett's one-tailed test; $p \leq 0.05$) performed by study authors
*Statistically significant effect (Mann Whitney Test; $p \leq 0.05$) performed by DS

There was no statistical significant difference in the growth parameters between the negative and solvent control. Therefore, the data were pooled and compared to the treatment groups. Treatment groups were smaller in length compared to the controls. The mean length in the treatment groups ranged from 4.32 to 4.52 mm and the length of the controls revealed a mean length of 4.57 mm in the negative control, 4.54 mm in the solvent control and 4.55 mm for the pooled controls. Dunnett's test indicated a statistical significant difference in the mean body length in the 2.4, 5.5, 12 and 21 ng/L treatment group when compared to the pooled control ($p \leq 0.05$). It was mentioned by the authors that the reductions in mean body length in the 2.4, 5.5, 12 and 21 ng/L treatment group did not follow a concentration response pattern and the differences were not considered to be biologically relevant. Further it was noted that the mean length of the daphnids in the 2.4, 5.5, 12 and 21 ng/L treatment group were within the ranges of the mean length of daphnids in the historical control data (3.6 to 6.3 mm).
The descriptive statistic for the length of the first generation daphnids was also performed by DS. Data from the solvent control and the combined controls and the 2.4 ng/L treatment group are not normally distributed (Shapiro-Wilk test). As the data are not normaly distributed a Mann Whitney Test was considered to be necessary and performed accordingly. In daphnids exposed to a mean measured concentration of 1.4 ng/L no

statistically significant reduction in body length of 0.66 % was observed. For daphnids exposed to a mean measured concentration from 2.4 ng/L onwards a statistically significant reduction in body length in the range of 2.63 to 5.05 % was observed. Based on the reduced length, the **$LOEC_{growth,\ lenght}$ is 0.0000024 mg/L (2.4 ng/L)**, and the **$NOEC_{growth,\ length}$ is 0.0000014 mg/L (1.4 ng/L)**. Both values are based on mean measured concentrations.

Table 19: Summary table growth (dry weight in mg) of first generation *Daphnia magna* (Anonymous, 2012b).

Mean measured concentration (ng/L)	Mean body weight (mg)	Standard deviation	Individuals (n)	Reduction (%)
Pooled control	0.67	0.12	34	-
Negative control	0.63	0.15	15	-
Solvent control	0.69	0.09	19	-
1.4	0.75	0.13	18	-11.94
2.4	0.60	0.10	18	10.44
5.5	0.74	0.11	20	-10.44
12	0.76	0.12	19	-13.43
21	0.63	0.10	18	5.97
43	0.60	0.10	18	10.44

As there was no statistically significant difference between the control groups for the body weight, the solvent and negative control were pooled for comparison with the treatment groups. In the 1.4, 5.5 and 12 ng/L treatment groups the mean body weight was higher when compared to the pooled control. In the 2.4, 21 and 43 ng/L treatment groups there was a reduction in the mean body weight when compared to the pooled control. The study revealed no statistically significant difference between the pooled control and the treatment groups (using Dunnet's test, $p > 0.05$).

Deviations compared to the OECD TG 211

- **Mortality in negative control**

The number of surviving parents in the untreated controls is a validity criterion according to OECD TG 211. In OECD TG 211 it stated that "*For a test to be valid the mortality of the parent animals does not exceed 20% at the end of the test" and "The same validity criterion (20%) can be used for accidental and inadvertent parental mortality for the control as well as for each of the test concentration*". 4 replicates of the negative control each containing 5 daphnids were used; 3 replicates worked well, while one replicate was not usable most probably due to improper handling. The study is considered as valid by the DS, as three negative replicates and four replicates with the solvent control were available for assessment and this inadvertent mortality is not considered to change the outcome of the study.

- **Amount of feeding**

Daphnids were fed 2 to 3 times per day through day 7 and then 4 times per day until the end of the study. The amount of fed was appr. 0.7 mg C/daphnids/day. This amount exceeds the recommended amount of 0.1 to 0.2 mg C/daphnids/day. The authors fed more to obtain acceptable reproduction rates.

- **Missing output expressed as total number of living offspring produced by parent**

In this study organisms were held in groups of 5 individuals per chamber (in total 20 / treatment group). In that case it is not possible to exclude any offspring from the statistical analysis if accidental/inadvertent parental mortality occurs, when reproduction has begun. Therefore, the output shall be expressed according to OECD TG 211 as „total number of living offspring produced per parent". Output expressed as total

number of living offspring produced by parent was not indicated by authors, but calculated by DS (see table below).

Table 20: Summary table of output as total number living offspring produced per parent (calculated by DS)

Mean measured concentration (ng/L)	Replicates	Sum of living offspring (n)	Sum parents alive (n)	Number of living offspring per parent (n)	Mean numbers of living offspring per parent per control or treatment group (±Standard deviation)
Negative control	A1	589	5	118	121.3 (± 7.7)
	A2	-	-	-	
	B1	580	5	116	
	B2	651	5	130	
Solvent control	A1	615	4	154	130.0 (± 19.6)
	A2	627	5	125	
	B1	671	5	134	
	B2	533	5	107	
1.4	A1	693	5	139	124.3 (± 12.8)
	A2	554	5	111	
	B1	656	5	131	
	B2	583	5	117	
2.4	A1	515	5	103	94.6 (± 13.2)
	A2	393	5	79	
	B1	538	5	108	
	B2	446	5	89	
5.5	A1	653	5	131	122.0 (± 11.1)
	A2	574	5	115	
	B1	661	5	132	
	B2	551	5	110	
12	A1	635	5	127	102.5 (± 16.3)
	A2	468	5	94	
	B1	476	5	95	
	B2	471	5	94	
21	A1	571	5	114	109.3 (± 23.2)

	A2	680	5	136	
	B1	536	5	107	
	B2	399	5	80	
43	A1	482	5	96	123.5 (± 25.1)
	A2	639	5	128	
	B1	570	5	114	
	B2	623	5	156	

The mean number of living off-spring produced per parent animal surviving at the end of the test is ≥ 60 in the controls and the coefficient of variation around the mean number of living offspring produced per parent animal in the controls is ≤ 25%. The validity criteria according to the OCED TG are fulfilled.

Conclusion

The mortality in the negative control is higher than recommended by the OECD TG. But the mortality occurred only in 1 out of 4 replicates, most probably due to a handling mistake. The mean number of living off-spring produced per parent animal surviving at the end of the test is ≥ 60 in the controls and the coefficient of variation around the mean number of living offspring produced per parent animal in the controls is ≤ 25%. Separate from the above described mortality issue all other validity criteria according to the OCED TG are fulfilled. The study is considered as valid by the DS, as three negative replicates and four replicates wih the solvent control were available for assessment and this inadvertent mortality is not considered to change the outcome of the study. The study is rated as Klimisch 2 by the DS.

Based on the reduced length or individual daphnids, a **NOECgrowth, length of 0.0000014 mg/L (1.4 ng/L)** could be derived which is based on mean measured concentrations. The study is valid and the NOEC can be used for classification purposes.

Please note that according to the study authors and the registrants no statistically significant treatment-related effects on survival, reproduction and growth (length and weight) at test concentrations ≤ 0.000043 mg/L (43 ng/L) was observed, resulting in a NOEC based on reproduction, survival and growth of 0.000043 mg/L (43 ng/L) which is the highest test concentration. For effects on growth, the study authors used the Dunnett's one-tailed test for comparison of the treatment and control groups. The study authors noted a statistical significant reduction in mean total length in the 2.4, 5.5, 12 and 21 ng/L treatment group when compared to the pooled control. The differences in the mean total length were considered not to follow a dose response pattern and therefore not to be biologically meaningful by the study authors. Further the study authors mentioned that the mean length of daphnids in the 2.4, 5.5, 12 and 21 ng/L treatment group were within the ranges of the mean length observed in historical control data (3.6 to 6.3 mm).

The descriptive statistic for the length of first generation daphnids was also performed by the DS. A Dunnetts's test can be performed when the data are normal and homogenous. It was exhibited that data from the solvent control and the combined controls and the 2.4 ng/L treatment group are not normally distributed using the Shapiro-Wilk test. As the data are not normally distributed a Mann Whitney test was considered to be more necessary by the DS and was performed accordingly. A $NOEC_{growth,\ length}$ of 0.0000014 mg/L (1.4 ng/L) was derived as for daphnids exposed to a mean measured concentration from 2.4 ng/L onwards a statistically significant reduction in body length was observed (see Table 18).

11.6.3 Chronic toxicity to algae or other aquatic plants

In the study by Anonymous (2012a) no toxicity was observed after the test duration of 96 hours leading to a NOEC of ≥ 12.53 ng/L (based on geometric mean measured concentrations) for the organism *Raphidocelis subcapitata* based on cell density, growth rate and yield.

In the second available study by Anonymous (1988) no toxicity was observed after 96 hours. The 96h-NOEC was ≥ 1000 mg/L based on nominal test concentrations for the organism *Raphidocelis subcapitata*.

For detailed test description of both studies see section 11.5.3.

11.6.4 Chronic toxicity to other aquatic organisms

No data available.

11.7 Comparison with the CLP criteria

11.7.1 Acute aquatic hazard

As there are acute data available on fish, invertebrates and algae, there is a need to assess the criteria given in Table 4.1.0(a) of the CLP Regulation. The classification would, subsequently, be according to the most stringent outcome.

Acute (short-term) aquatic hazard classification categories for hazardous to the aquatic environment (Table 4.1.0(a) of the CLP Regulation):

Category Acute 1: (Note 1)	
96 hr LC 50 (for fish)	≤ 1 mg/l and/or
48 hr EC 50 (for crustacea)	≤ 1 mg/l and/or
72 or 96 hr ErC 50 (for algae or other aquatic plants)	≤ 1 mg/l. (Note 2)

Note 1:When classifying substances as Acute Category 1 and/or Chronic Category 1 it is necessary at the same time to indicate the appropriate M-factor(s) (see Table 4.1.3).

Note 2:Classification shall be based on the ErC_{50} [= EC_{50} (growth rate)]. In circumstances where the basis of the EC_{50} is not specified or no ErC_{50} is recorded, classification shall be based on the lowest EC50 available.

The available experimental acute data on fish, invertebrates and algae indicate that TBMD is not toxic up to its solubility limit of 0.032 µg/l. Assessing the criteria of Table 4.1.0(a), a classification as Acute Category 1 for TBMD is not warranted.

11.7.2 Long-term aquatic hazard (including bioaccumulation potential and degradation)

Bioaccumulation potential

The estimated log K_{OW} is 8.99 (EPISuiteTM - KOWWIN v1.68). The BCF key study and the supporting BCF study derived a BCF value > 500 L/kg. In a supporting OECD 305 dietary study BMF_{KgL} values were derived for TBMD and Hexachlorbenzol (HCB). The BMF_{KgL} was higher for TBMD than for HCB (TBMD: BMF_{KgL} = 0.95; HCB: BMF_{KgL} = 0.55).

As the estimated log K_{OW} is > 4 and the experimental determined BCF values for fish are > 500 L/kg, TBMD has a potential to bioaccumulate in aquatic environments.

Rapid degradability

According to 4.1.2.9.5. of Annex I of Regulation (EC) No 1272/2008 (CLP Regulation) substances are considered rapidly degradable in the environment if one of the following criteria holds true:

(a) if, in 28-day ready biodegradation studies, at least the following levels of degradation are achieved:

(i) tests based on dissolved organic carbon: 70 %;

(ii) tests based on oxygen depletion or carbon dioxide generation: 60 % of theoretical maximum.

or (b) if, in those cases where only BOD and COD data are available, when the ratio of BOD 5 /COD is ≥ 0,5; or

or (c) if other convincing scientific evidence is available to demonstrate that the substance can be degraded (biotically and/or abiotically) in the aquatic environment to a level > 70 % within a 28-day period

(ECHA, 2017a)

TBMD is not readily biodegradable based on a 28-day test for ready biodegradability (OECD 301B), as the BOD was 0% after 28 days and the quantitative HPLC analysis showed disappearance of 4, 2, and 2 % after 4 weeks.

A study performed according to the EU Method C.4-C, equalling OECD 301B, resulted in 0% degradation of TBMD after 28 days. As the threshold for ready biodegradability is not met within 28 days, it can be concluded that TBMD is not readily biodegradable.

Regulation (EC) No 1272/2008, Annex I 4.1.2.9.3. allows the usage of degradation data that are available (degradation half-lives) and "*these can be used in defining rapid degradation provided that ultimate biodegradation of the substance, i.e. full mineralisation, is achieved. Primary biodegradation does not normally suffice in the assessment of rapid degradability unless it can be demonstrated that the degradation products do not fulfil the criteria for classification as hazardous to the aquatic environment*" (ECHA, 2017a).

In the OECD TG 307 aerobic soil study (Anonymous, 2021) mineralisation was comparable among the four soils and reached a maximum of 25.3% applied radioactivity (%AR) after 125 days. Several unknown and unidentified degradation products occurred prior or shortly after application of radioactive labelled TBMD. Mineralisation started afterwards. Aquatic toxicity data is not available for any degradation product, including for those three degradation products where a CAS number was allocated by DS (CAS 4359-97-1, CAS 1620-98-0 and CAS 719-22-2). Due to the lack of data for the unknown, unidentified and identified degradation products, the need for classification as hazardous to the aquatic environment of the degradation products cannot be excluded, and therefore, TBMD is considered as not rapid degradable.

Based on all information available, TBMD can be considered as not rapidly degradable.

Long-term aquatic hazard

As there are long-term (chronic) data available on invertebrates and algae, and the substance is not rapidly degradable there is a need to assess the criteria given in Table 4.1.0(b)(i) of the CLP Regulation. The classification would, subsequently, be according to the most stringent outcome.

(i) Non-rapidly degradable substances (Note 3) for which there are adequate chronic toxicity data available (Table 4.1.0(b)(i) of the CLP Regulation)

Category Chronic 1: (Note 1)	
Chronic NOEC or EC x (for fish)	≤ 0,1 mg/l and/or
Chronic NOEC or EC x (for crustacea)	≤ 0,1 mg/l and/or
Chronic NOEC or EC x (for algae or other aquatic plants)	≤ 0,1 mg/l.
Category Chronic 2:	
Chronic NOEC or EC x (for fish)	> 0,1 to ≤ 1 mg/l and/or
Chronic NOEC or EC x (for crustacea)	> 0,1 to ≤ 1 mg/l and/or
Chronic NOEC or EC x (for algae or other aquatic plants)	> 0,1 to ≤ 1 mg/l.

Note 1:When classifying substances as Acute Category 1 and/or Chronic Category 1 it is necessary at the same time to indicate the appropriate M-factor(s) (see Table 4.1.3).

Note 3: When no useful data on degradability are available, either experimentally determined or estimated data, the substance should be regarded as not rapidly degradable

Assessing the criteria of Table 4.1.0(b)(i) of the CLP Regulation, leads to classification as Aquatic Chronic 1, based on the lowest chronic 21d-NOEC of 0.0000014 mg/L (1.4 ng/L), which was derived for growth (effects on length) in daphnids.

According to Table 4.1.3 of the CLP Regulation a chronic M-factor of 10000 is warranted.

As there are only acute experimental data available on fish and the QSAR calculations on chronic fish toxicity are not deemed usable, a comparison with Table 4.1.0 (iii) of the CLP Regulation is performed for this trophic level.

(iii) Substances for which adequate chronic toxicity data are not available (Table 4.1.0 (iii) of the CLP Regulation)

Category Chronic 1: (Note 1)	
96 hr LC_{50} (for fish)	≤ 1 mg/l and/or
Category Chronic 2:	
96 hr LC_{50} (for fish)	> 1 to ≤ 10 mg/l and/or
Category Chronic 3:	
96 hr LC_{50} (for fish)	> 10 to ≤ 100 mg/l and/or

Note 1:When classifying substances as Acute Category 1 and/or Chronic Category 1 it is necessary at the same time to indicate the appropriate M-factor(s) (see Table 4.1.3).

The available experimental acute data on fish indicate that TBMD is not toxic up to its solubility limit of 0.032 µg/l. Assessing the criteria of Table 4.1.0 (iii) a classification as Chronic Category 1, 2, or 3 for TBMD is not warranted, while a classification is needed as Aquatic Chronic 1 with a chronic M-factor of 10000 based on chronic data for invertebrates.

11.8 CONCLUSION ON CLASSIFICATION AND LABELLING FOR ENVIRONMENTAL HAZARDS

Based on available long-term data on invertebrates it is proposed to classify TBMD as Aquatic Chronic 1; H410 (Very toxic to aquatic life with long lasting effects) with a chronic M-factor = 10000.

12 EVALUATION OF ADDITIONAL HAZARDS

12.1 Hazardous to the ozone layer

Not evaluated in this report.

13 ADDITIONAL LABELLING

Not relevant.

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