



SQC (EQS_{sed}) – Proposal by the Ecotox Centre for: Copper (Cu)

First proposal: 14.07.2016 (last bibliographic research) 15.01.2021 (implementation of the expertise)



Imprint

Publisher Swiss Centre for Applied Ecotoxicology, 1015 Lausanne

Commissioned by

FOEN, Federal Office of the Environment, Water Quality Section, 3003 Bern

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Please note that the suggested EQS and contents of this dossier do not necessarily reflect the opinion of the external reviewer.

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Citation Proposal

Michel Wildi, Carmen Casado-Martinez. 2021. SQC (EQS_{sed}) – Proposal by the Ecotox Centre for: Copper (Cu). Lausanne (CH): Swiss Centre for Applied Ecotoxicology; 44 pp.



Summary

SQC (EQS_{sed}): 9.9 mg/kg d.w.

In the framework of the Module Sediment, which is intended to help cantons in sediment quality assessment, the Ecotox Centre develops proposals for Environmental Quality Criteria for sediment (SQC). SQC are derived applying the methodology described in the EU-Technical Guidance (TGD) for Deriving Environmental Quality Standards (EQS). In order to ensure that the dossiers are internationally comparable, the English terminology of the TGD will be used in the remainder of the dossier. These criteria provide a first screening tool to evaluate sediment chemical quality and the potential risk for the aquatic ecosystem. Based on the scientific literature available at present a SQC for copper (Cu) of 9.9 mg/kg d.w. proposed for standard sediments with 1 % OC.

Zusammenfassung

SQK (EQS_{sed}): 9.9 mg/kg TS

Im Rahmen des Sedimentmoduls, das den Kantonen bei der Bewertung der Sedimentqualität helfen soll, entwickelt das Oekotoxzentrum Vorschläge für Umweltqualitätskriterien für Sedimente (SQK). Diese Kriterien dienen als Methode für ein erstes Screening zur Bewertung der chemischen Sedimentqualität und des potenziellen Risikos für aquatische Ökosysteme. Auf der Basis von Literaturdaten für die Wirkung von Tebuconazol und unter Verwendung der Methode, die in der Technischen Richtlinie der EU zur Ableitung von Umweltqualitätsnormen beschriebenen wird, schlägt das Oekotoxzentrum einen SQK für Kupfer (Cu) von 9.9 mg/kg TS für Standardsedimente mit 1 % OC vor.

Résumé

CQS (EQS_{sed}): 9,9 mg/kg p.s.

Dans le cadre du module Sédiments qui devrait aider les cantons à évaluer la qualité des sédiments, le Centre Ecotox élabore des propositions de critères de qualité environnementale pour les sédiments (CQS). Les CQS sont dérivés en appliquant la méthodologie décrite dans le Guide Technique de l'UE (TGD) pour la Dérivation des Normes de Qualité Environnementale (EQS). Afin que les dossiers soient comparables au niveau international, la terminologie anglaise du TGD est utilisée ci-dessous. Ces critères fournissent un premier outil de dépistage pour évaluer la qualité chimique des sédiments et le risque potentiel pour l'écosystème aquatique. Sur la base des données sur les effets existants dans la littérature un CQS pour le cuivre (Cu) de 9,9 mg/kg p.s. est proposé pour les sédiments standards avec 1 % CO.



Sommario

CQS (EQS_{sed}): 9,9 mg/kg p.s.

Nell'ambito del modulo Sedimenti, che è finalizzato ad aiutare i Cantoni nella valutazione della qualità dei sedimenti, il Centro Ecotox sviluppa proposte per i criteri di qualità ambientale per i sedimenti (CQS). I CQS sono derivati applicando la metodologia descritta nella Guida Tecnica dell'UE (TGD) per la Derivazione degli Standard di Qualità Ambientale (EQS). Per garantire che i dossier siano comparabili a livello internazionale, viene utilizzata la terminologia inglese del TGD. Questi criteri forniscono un primo strumento di screening per valutare la qualità chimica dei sedimenti e il potenziale rischio per l'ecosistema acquatico. Sulla base della letteratura scientifica disponibile allo stato attuale un CQS per il rame (Cu) di 9,9 mg/kg p.s. è proposto per sedimenti standard con 1 % CO.



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1 General Information

Selected information on Cu relevant for sediment is presented in this chapter. Background registration information and risk assessments referred to are:

- VRAR (ECI 2008). European Union Risk Assessment Report Voluntary Risk assessment of Copper, Copper II Sulphate pentahydrate, Copper II Sulphate Pendahydrate, Copper(I)Oxide, Copper(II)Oxide, Dicopper Chloride Trihydroxide, CAS No: 7440-50-8, 7758-98-7, 1317-3-1, 1317–38–0, 1332-65-6, EINECS No: 231–159–6, 231–847–6, 215-270-7, 215–269–1, 215-572-9
- ECHA (2020). Information on Registered Substances: Copper, CAS number: 7440-50-8 https://echa.europa.eu/registration-dossier/-/registered-dossier/15562. Last modified: 17/09/2020.
- EC (2016). Draft renewal assessment report prepared according to the Commission regulation (EU) No 1107/2009. Copper compounds. Available at: https://www.efsa.europa.eu/en/consultations/call/170203
- OECD SIDS (2014). Initial assessment profile Copper and Copper compounds.

1.1. Identity and physico-chemical properties

The EU Voluntary Risk Assessment for Copper and Copper compounds (ECI 2008) includes four CAS numbers¹. The OECD Cooperative Chemicals Assessment Programme (CoCAP) adds one additional CAS number to the Copper Category group, being all of them commonly used Cu substances that are similar from a hazard point of view (OECD 2014)². It is considered that the Cu ion is the reactive functional group driving the toxicity within the category while the counter-ions of the Cu salts (e.g. sulphate, chloride and hydroxide) do not contribute to the environmental (nor systemic human toxicity) of Cu salts³. This dossier therefore describes general properties and characteristics for Cu (CAS 7440-50-8) and measured environmental concentrations and ecotoxicity data refer to total Cu concentrations if not otherwise stated. Nanoform Cu substances are excluded from this assessment because the biological effects of nanoform metals can differ from the ionic forms.

Table 1 summarizes identity and physico-chemical parameters for Cu required for EQS derivation according to the TGD (EC 2018). Where available, experimentally collected data is identified as (exp.) and estimated data as (est.). When not identified, no indication is available in the cited literature.

³ Additional substance from EC (2016): Copper hydroxide [CuH2O2]: CAS N° 20427-59-2

¹ Copper powder and massive [Cu]: CAS N° 7440-50-8 Copper II sulphate pentahydrate [CuSO4.5 H2O]: CAS N° 7758-99-8 Copper(I)oxide [CuO]: CAS N° 1317-38-0 Copper(II)oxide [Cu2O]: CAS N° 1317-39-1

² Additional substance : Dicopper chloride trihydroxide [Cu2Cl(OH)3]: CAS N° 1332-65-6



Table 1 Information required for EQS derivation according to the TGD (EC 2018). Values not used in risk assessment in grey font.

Characteristics	Values	References
Common name	Copper	ECHA (2020)
IUPAC name	Copper	ECHA (2020)
Chemical group	Transitional metal	Kim et al. (2016)
Molecular formula	Си	ECHA (2020)
CAS	Copper powder and massive [Cu]: 7440- 50-8 Copper II sulphate pentahydrate [CuSO4.5 H2O]: 7758-99-8 Copper(I)oxide [CuO]: 1317-38-0 Copper(II)oxide [Cu2O]: 1317-39-1 Dicopper chloride trihydroxide [Cu2Cl(OH)3]: 1332-65-6	ECHA (2020) OECD (2014)
EC Number	231-159-6	ECHA (2020)
SMILES code	[Cu]	ECHA (2020)
Molecular weight [g/mol]	63.546	ECI (2008)
Melting point [°C]	1059 – 1069	ECHA (2020)
Boiling point [°C]	Not determined in view of the high melting point	ECHA (2020)
Vapour pressure [Pa]	Not relevant	ECHA (2020)
Henry's law constant [Pa·m³/mol]	Not relevant	ECHA (2020)
Water solubility [mg/L]	0.130 – 192.03, at 20 - 30 °C and pH 4.03 - 8.98 Insoluble (OECD Guideline 105, water solubility)	ЕСНА (2020)
Dissociation constant (pKa)	Not relevant	ECHA (2020)
Octanol-water partition coefficient (log K _{ow})	Scientifically unjustified	ECHA (2020)
Organic carbon adsorption coefficient (log K _{oc})	Not relevant	ECHA (2020)
Sediment adsorption coefficient (K _p [L/kg])	Sediment: 24 409 (50 th percentile) Suspended matter: 30 246 (50 th percentile)	Heijerick and Van Sprang (2005, 2008) cited in ECHA (2020)
Aqueous hydrolysis DT ₅₀	Scientifically unjustified	ECHA (2020)
Aqueous photolysis DT ₅₀	Scientifically unjustified	ECHA (2020)
Biodegradation in water environment DT ₅₀ [d]	Scientifically unjustified	ECHA (2020)
Biodegradation in sediment DT ₅₀ [d]	Scientifically unjustified	ECHA (2020)
Biodegradation in soil DT ₅₀ [d]	Scientifically unjustified	ECHA (2020)

1.2. Regulatory context and environmental limits

Copper is a high production volume (HPV) chemical included in the OECD Cooperative Chemicals Assessment Programme (CoCAP), which published an initial assessment profile in 2014 (OECD 2014), and is fully registered in the EU as being manufactured and/or imported in the European Economic



Area in 1 000 000 – 10 000 000 tonnes per year (ECHA 2020). The voluntary risk assessment (ECI 2008) concluded that, for the sediment compartment, measured and predicted concentrations exceeded the predicted no effect concentration (PNEC_{sediment} = 87 mg/kg d.w.) in a number of the processing scenarios, pointing to a potential risk for the aquatic environment (water and sediments). At these local sites, the bioavailability corrections, which have been used in the risk assessment (organic carbon –OC- and acid volatile sulfides –AVS- corrections), should be incorporated as an essential part of the risk reduction measurement process for these site.

As an inorganic substance, a PBT and vPvB assessment shall not be conducted. However, as for other essential metals, Cu is regulated by cells/organisms and therefore is considered not to bioaccumulate or biomagnify (ECI 2008, EC 2016, ECHA 2020). Although persistence does not apply either to metals (ECHA 2020), Cu is considered persistent (EC 2016, vol. 1, p. 158) in the context of the present EQS proposal for the sediment compartment.

Table 2 summarizes existing regulations and environmental limits in Switzerland, Europe and elsewhere for Cu.

Europe	
	Not identified as a priority substance in the field of the
EU Priority substance list	Water Framework Directive (Directive 2013/39/EU)
	2 nd priority substance
REACH	Manufactured and/or imported in the European Economic
	Area in 1 000 000 - 10 000 000 tonnes per year
OFCD	High Production Volume substance, included in the
	Cooperative Chemicals Assessment Programme (CoCAP)
Switzerland	
Water protection Ordinance (WPO)	Annex 2: Requirements on Water Quality
(Status as of 1 April 2020)	5 μg/L Cu (total), 2 μg/L Cu (dissolved) (maximum
	concentrations)
	Annex 3: Requirements for the Discharge of Polluted
	Waste Water
	0.5 mg/L Cu (total) for discharge into waters, for discharge
	into public sewers 1 mg/L
Contaminated Sites Ordinance (CSO)	Annex 1: Concentration values for assessing the impact of
(Status as of 1 May 2017)	polluted sites on ground and surface waters (1.5 mg/L)
	Annex 3: Concentration values for the assessment of the
	need for remediation of soil
	1000 mg/L (sites used for agricultural or horticultural
	purposes)
	1000 mg/L (sites in private gardens and allotments,
	children's playgrounds and other facilities where children
	play regularly)
Register relating to Pollutant Release	Annex 2: Threshold value for reporting obligation to air,
Ordinance (PRTRO)	water and soil (100, 50, 50 kg/year respectively)
(Status as at 23 January 2007)	

Table 2 Existing regulation and environmental limits for Cu in Switzerland and Europe⁴. Environmental limits in sediments are included in Table 3.

⁴ Additional environmental limits are available e.g. <u>www.webetox.uba.de</u>



Ordinance on Biocidal Products	Annex 2: List of approved active substances
(OBP) (Status as of 10 March 2020)	
DEFR ordinance on organic farming	Annex 1: Used amount permitted as phytosanitary
(Status as of 1 January 2019)	products
Chemical Risk Reduction Ordinance	Annex 2.6: Pollutant content of organic fertilisers, limit
(ORRChem) (01.01.16)	values
	Organic fertilisers, recycling fertilisers with the exception
	of mineral recycling fertilisers and farm manure: 100 grams
	per tonne of dry matter, 50 g/t dry matter if the proportion
	of pig excrement is more than 50% of dry matter
	Mineral recycling fertilisers 3 000 grams per tonne of
	phosphorus (P)

Table 3 presents sediment quality guidelines for Cu. The $PNEC_{sediment}$ used in the VRAR (ECI 2008) is set at 89 mg/kg d.w. This $PNEC_{sediment}$ was derived through statistical extrapolation (SSD) using effect concentrations normalized to 5% OC from spiked sediments with low AVS and application of an Assessment Factor 1 to the HC5-50.

The most recent EQS_{sed} derived by Sahlin and Ågerstrand (2018) according to the TGD methodology (EC 2018) is set at 36 mg/kg d.w. as added risk approach and normalized to 5% OC, or as 28 mg/kg d.w. as added risk approach independently of the OC content based on the deterministic approach and an assessment factor of 2. If based on the total risk approach, a value of 44 mg/kg d.w. for 5% OC is also proposed based on ECI (2008) and the AF to 2.

Additional threshold values include the threshold used in Norway of 84 mg/kg d.w. (derivation method unknown; Breedveld et al. 2018), the MPA of 37 derived using the Equilibrium Partitioning in the Netherlands (Crommentuijn et al. 2000) and the Interim Sediment Quality Guideline from Canada set at 35.7 mg/kg d.w. based on field and laboratory studies (CCME 1999). The Target Value for suspended matter from the International Commission for the Protection of the Rhine is set at 50 mg/kg d.w. (ICPR 2016), while the EQS value is set at 2.8 μ g/L.

Description	Value	Development method	References
	[mg/kg d.w.]		
PNECsediment	89	Derived using species sensitivity	VRAR (ECI 2008)
		distribution (EC 2018):	
		application of an Assessment	
		Factor 1 to the HC₅, weight of	
		evidence.	
Maximum Permissible	37 (10% OC and	Based on adapted Equilibrium	Crommentuijn et
Addition (MPA)	25% clay	Partitioning Approach.	al. (2000)
		Maximum Permissible	
		Concentration is 73, derived	
		adding the MPA to the	
		background in the Netherlands	
		of 35 mg/kg d.w.	
Target Value	50	For suspended matter, used by	ICPR (2016)
		the International Commission	
		for the Protection of the Rhine.	

Table 3 PNEC/quality standards available from authorities and reported in the literature (additional sediment quality standards based on field data are summarized in Section 7). If not otherwise stated, values derived for sediment.



Description	Value	Development method	References
	[mg/kg d.w.]		
Class II-Class III	84	Based on available information	Breedveld et al.
boundary		from laboratory tests and field	(2018)
		data.	
EQS _{sed}	36 (ARA 5% OC)	Deterministic method and	Sahlin and
	28 (ARA)	application of an Assessment	Ågerstrand (2018)
	44 (TRA 5% OC)	Factor of 2. ARA to be added to	
		relevant background	
		concentration, TRA is total risk	
		approach, based on ECI (2008)	
		and Assessment Factor of 2.	
Interim Sediment	35.7	Based on available information	CCME (1999)
Quality Guideline		from laboratory tests and field	
(ISQG)		data.	
RAC _{sed;ch}	68	Derived using species sensitivity	EC (2016)
		distribution (EC 2018):	
		application of an Assessment	
		Factor 1.	

1.3. Use and emissions

Copper is used for wire, tubing, fitting systems and building construction material. In Switzerland, the mean consumption of Cu between 1995 and 2000 was estimated at 75 000 t/year in finished goods (von Arx 2006, OFEV 2016). Approximately 35 to 40% of this amount was used for construction, 5 to 10% in the automotive industry and 5% for electronics devices (von Arx 2006, OFEV 2016). These figures are in line with the average consumption of 8 kg/cap/year of Cu flows in building, infrastructure and mobile estimated for 2000 by Bader et al. (2011).

According to von Arx (2006), as a chemically active substance Cu is mainly used as a biocide in pesticide products at 65 t/year, wood protection products 55 t/year and antifouling 5 t/year. A non-negligible amount of Cu is used as feed additive for livestock (30 t/year). Cu is also used in pigments for printing and dying paper, textiles and plastics (von Arx 2006). In Switzerland, the release of Cu to the aquatic environment from punctual sources as industries (0.1%) or WWTPs (1.73%) is negligible in comparison with diffuse sources (von Arx 2006). Cu released to the aquatic environment from diffuse sources comes mainly from transport and corrosion of building materials (42.5%, 15 t/year) followed by agriculture (30.5%, 11 t/year) (von Arx 2006), including farmyard manure and treatment of vines (Gubler et al. 2015). The contribution of atmospheric deposition is estimated at 5.2 t/year, accounting for 14.5% of the total amount of Cu to the aquatic environment (von Arx 2006).

1.4. Mode of action and sensitivity of taxonomic groups

Copper is an essential element for living organisms with a defined window of essentiality⁵. According to the substance profile available from ECHA (2020), it is essential for growth and development, neurological function, wound healing and immunocompetence. Toxicity of metals occurs due to blocking of the essential biological functional groups of biomolecules, displacing essential metal ions in biomolecules, and/or modifying the active conformation of biomolecules. Copper can also result in

⁵ According to the VRAR (ECI 2008), the window of essentiality for an essential element is the range between the lowest and highest concentration that allows life.



the production of reactive oxygen radicals leading to tissue damage and molecule dysfunction due to redox cycling within the cell.

Animal carcinogenicity studies with Cu compounds indicate that the target organs in chronic Cu exposure are liver and kidney, but long exposure do not relate to increased severity of disease due to homeostatic control mechanisms that regulate uptake and excretion of Cu in animals on a daily basis (Harrison et al. 1954 and Hebert 1993 cited in ECHA 2020). Although available animal carcinogenicity studies were assessed as of limited quality, based on the existing information, ECHA (2020) concluded that there is no evidence for carcinogenic activity of Cu and Cu compounds considered in the chemical safety report. The overall weight of the evidence from the existing genotoxicity assays suggests that Cu and Cu compounds do not have biologically relevant genotoxic activity and no classification and labelling for mutagenicity is required.

Regarding reproductive and developmental toxicity, chronic studies with rats have shown no effects in reproductive endpoints (growth or fertility) in compliance with the OECD definition of reproductive toxicity while no evidence of test substance-related teratogenicity was observed in chronic studies with pregnant rabbits (ECHA 2020). ECHA (2020) indicates that Cu and Cu compounds are under assessment as endocrine disrupting chemicals.

In freshwater organisms, the target tissue for Cu toxicity is the water-organism interface, with the cell wall and gill-like surfaces being the target biotic ligands and the disturbance of the sodium homeostasis the key indicator of Cu toxicity (Paquin et al. 2002, De Schamphelaere and Janssen 2004, Kamunde et al. 2001 and 2005, cited in ECHA 2020). However, it is still unclear how ionoregulatory disturbance affects organisms and reproduction success in long-term exposures and how acclimation interacts, as fish and invertebrates can effectively regulate newly accumulated Cu depending on species, life stage and duration of exposure (Weber Scannell 2009) but this may come at an energetic cost. Chronic effects of Cu in fish include decrease growth, changes in fish behavior, including olfactory responses, agonistic responses, avoidance and attraction, and changes in swimming ability or swimming speed (Weber Scannell 2009). The mechanisms of metal toxicity in algae are considered different from those in fish and invertebrates. According to the review from Stauber and Davies (2000) as cited in VRAR (ECI 2008), Cu causes changes in membrane potential and permeability or compete with essential metals for binding and uptake. Once in the cytoplasm, Cu can inhibit enzymes (e.g. esterase and Bgalactosidase), cause changes in intracellular pH and affect organelles (e.g. chloroplasts and mitochondria). Structural alterations to thylakoid membranes, inhibition of photosynthesis, disruption in mitochondrial electron transport, disruption of the glutathione metabolism and ultimately inhibition of cell division and therefore algal growth have been reported upon Cu exposure (ECI 2008).

According to available toxicity values reviewed in Weber Scannell (2009), freshwater zooplankton and mussels are more sensitive to acutely toxic effects from Cu than aquatic insects and fish, and no correlation was found between zooplankton and mussels toxicity values and either species or (for mussels) life stage. In chronic exposures, toxicity values for freshwater fish range from 5 to 72 μ g/L, from 3.1 to 12 μ g/L for freshwater mussels and from 0.3 to 0.6 μ g/L for freshwater algae, which appear as the most sensitive taxonomic group (Weber Scannell 2009).

2 Environmental fate

2.1. Speciation and sorption/desorption processes

Copper entering aquatic systems through surface waters is rapidly bound to solid materials (sediments, colloids and suspended particles) in the water phase resulting in very low levels of free Cu²⁺ ion in



solution (EC 2016), therefore adsorption is the most important parameter determining the distribution of Cu in the aquatic compartment. Inorganic particles such as clay minerals and iron (Fe), manganese (Mn) and aluminum (Al) oxides, as well as organic materials, constitute the principal adsorbents for Cu in water and sediment (Landner and Reuther 2005). Both pH and the composition and concentration of solid materials (suspended matter or sediment) determine partitioning coefficients through complexation and speciation processes in both the aqueous and solid phase. There is, however, no clear answer whether the Fe- and Mn-oxides or the organic matter content (OM) of the solid phase are the most important binding sites (ECI 2008). In aerobic environments, the most probable precipitates forms are Cu hydroxide, Cu(OH)₂, malachite, Cu(CO₃)(OH)₂ and azurite, Cu₃(CO₃)₂(OH)₂. In anaerobic, biologically active sediments the solubility of Cu is reduced due to formation of very stable, sulfide-containing Cu(I) and Cu(II) minerals (Landner and Reuther 2005; EC 2016).

The partitioning coefficients retained for risk assessment in the VRAR (ECI 2008), based on the reviews from Heijerick and Van Sprang (2005 and 2008), are the median Kp for suspended matter of 30 246 L/kg and Kp for sediments of 24 409 L/kg.

2.2. Bioavailability

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Copper bioavailability is driven by physicochemical characteristics of the water-sediment system as well as the feeding behavior and exposure route of organisms. Bioavailability of divalent metals such as Cu shows dependency on several measurable parameters such as AVS, Simultaneous Extracted Metals (SEM), Cation Exchange Capacity (CEC), Fe/Mn oxyhydroxides, organic carbon (OC), particle size, hardness and pH (Tarazona et al. 2014). According to the high affinity of divalent metals for AVS in anaerobic sediments, the SEM-AVS approach proposes that metal concentrations in pore waters is determined by the molar relationship between metals and reactive sulfides:

$$\frac{2}{n}Me_{(aq)}^{n+} + FeS(s) < -> Me_{2/n}S(s) + Fe_{(aq)}^{2+}$$
$$\frac{2}{n}Me_{(aq)}^{n+} + MnS(s) < -> Me_{2/n}S(s) + Mn_{(aq)}^{2+}$$

Where $Me^{n_{aq}}$ is the aqueous form of a metal, MnS and FeS(s) are the insoluble Mn and Fe sulfide forms, $Me_{2/n}S$ is the insoluble metal sulfide forms, and $Mn^{2+}_{(aq)}$ and $Fe^{2+}_{(aq)}$ are the soluble Mn and Fe forms.

Where AVS exceed SEM concentrations, all metals will be bound to sulfides leading to decreased concentrations of free metal ions in porewater and bioavailability will decrease accordingly. AVS are expected to be a key parameter accounting for Cu bioavailability according to the greatest affinity of Cu for sulfide compared to other divalent metals such as lead, cadmium, zinc and nickel (ECI 2008). However, it is worth mentioning that the AVS concept is challenged by bioaccumulation in benthic invertebrates occurring where AVS do not exceed SEM, indicating bioavailability of AVS bound metals (De Jonge et al. 2010; Mendez-Fernandez et al. 2014) and the the potential for mobilisation of Cu through ingestion by benthic organisms (SCHER 2009; Sahlin and Ågerstrand 2018).

In aerobic sediments, OC and FeOOH content have the greatest influence on bioavailability by driving exposure of organisms to free ions in porewater and overlying water when this is the major exposure route (Chapman et al. 1998).

In the VRAR (ECI 2008), the SEM-AVS model was not used for sediment PNEC derivation because of analytical artifacts such as AVS content being underestimated due to the high strength of chemical bond of Cu with sulfide (Simpson et al. 1998) and/or SEM overestimated due to Fe(III) catalyzing CuS



oxidation (Dutrizac 1990 cited in ECI 2008) preventing normalization. The VRAR (ECI 2008) proposed OC normalization of spiked sediment toxicity data because normalization against this factor reduced the variability of toxicity tests results for a same species and endpoint when they were obtained from test sediments with different OC content ranging from 0.5 to 10% (ECI 2008).

In line with the VRAR (ECI 2008) and EC (2016), here, normalization of effect data against AVS has not been performed before EQS derivation. Because effect data in which bioavailability is maximized are preferred for EQS derivation, NOEC values from studies in which bioavailability of Cu was limited due to the presence of AVS were considered not relevant for EQS derivation. The threshold value for AVS was set at 0.77 mmol/kg d.w. (ECI 2008; EC 2016). This AVS value corresponds to the 10th percentile of the AVS concentrations derived from a Flemish dataset and is assumed to be representative of oxic conditions (ECI 2008). AVS values in head streams from South Switzerland reported by Burton et al. (2007) ranged from 0.006 to 0.02 mmol/kg d.w. Here we followed the same approach, but it should be noted that the effect data used in EQS derivation may not be representative of worst case scenarios where bioavailability is maximized in Switzerland.

Regarding normalization to OC, the VRAR (ECI 2008) derived sediment PNEC normalized to OC because normalization reduced variability of the effect data. Normalization for OC in test sediments reduces the intra-specific variability for all endpoints of *Chironomus riparius*, *Hyalella azteca* and *Tubifex tubifex* (see section 4.1). However the linear relationship between NOECs and OC is mainly due to one single point with high OC content of approx. 10%. For *Lumbriculus variegatus* and *Gammarus pulex* test sediments presented similar OC content and comparisons were not possible.

Based on this evidence, the approach presented in Figure 1 was applied, which is adapted from the general QS development scheme with five steps to account for bioavailability when deriving QS for metals in water and sediment-specific issues for the development of sediment (Fig. 1; EC 2018). The first step was to derive a QS_{sed,generic} without any consideration of bioavailability. Here the QS_{sed,generic} was derived from effect concentrations from studies with sediments that had a OC content in the 0.5-3% range. Once the QS_{sed,generic} was derived, a QS_{sed,bioav}. was derived using effect concentrations normalized for OC content, including also effect concentrations from spiked sediment toxicity tests with higher OC content because OC correction results in a general decrease in the intra-specific variance in the NOEC values and the proposed bioavailability normalization is able to explain the observed differences in toxicity due to increased OC content.





Figure 1 Approach used for the development of the EQS_{sed} taking into consideration bioavailability (from EC 2018).

2.3. Bioaccumulation and biomagnification

As an essential element, all living organisms require a certain amount of Cu to maintain biological functions. Species' homeostatic capacity allows an organism to actively regulate its tissue concentrations and maintain optimal levels for metabolic requirements under varying external concentrations of the essential element (Luoma and Rainbow 2007). The molecular mechanisms of Cu homeostasis are mediated by P-type ATPases, which can pump Cu across biological membranes in both directions, and Cu chaperones, important for the intracellular Cu homeostasis (Odermatt et al. 1992). These mechanisms are considered universal as the sequences of Cu chaperones are highly conserved between species (Wunderli-Ye et al. 1999). Besides these active regulation mechanisms, some organisms have developed internal regulation mechanisms such as molecular binding and sequestration of Cu in non-bioavailable or detoxified forms as a strategy to cope with Cu excess (Rainbow 1997; Marsden and Rainbow 2004).

According to the regulation of Cu bioaccumulation in living organisms, the traditional Bioconcentration and Bioaccumulation Factors (BCF/BAF) are not appropriate for hazard assessment (see reviews Adams et al. 2003; ECI 2008; EC 2016).

Regarding biomagnification and secondary poisoning, previous studies for the derivation of risk levels for Cu in waters have concluded that food chain transfer of Cu is of equal importance compared to direct exposure (ECI 2008). EC (2016) also concludes that given the absence of indication of bioaccumulation potential as supported by numerous studies secondary poisoning is not considered relevant.

3 Analytics

3.1. Methods for analysis and quantification limit

Copper can be analyzed after extraction by either inductively coupled plasma mass spectrometry (ICP-MS), inductively coupled plasma - optical emission spectrometry (ICP-OES) or atomic absorption spectrometry (AAS). There are several standard methods available that are recommended for metal



analysis including Cu (Table 4). The limits of detection (LOD) range from several μ g/kg d.w. for ICP-MS to few mg/kg d.w. for ICP-OES. Extraction can be performed on a greater amount of sediment to decrease the LOD.

Method	Analysis	Extraction	Reference
NF EN ISO 11885	ICP-OES	Reflux with nitric acid + hydrogen peroxyde	Schiavone and Coquery (2010)
NF EN ISO 15586	GF-AAS	Microwave or autoclave nitric acid or <i>aqua regia</i> mineralization	Schiavone and Coquery (2010)
ASTM D3974-09	AAS	Hot plate reflux with aqua regia	ASTM (2015)
	AAS	Chlorhydric acid extraction at room temperature	
Ifremer method	GF-AAS	Mineralization by heating with nitric	Chiffoleau and
		acid, chlorhydric acid + fluorhydric acid	Truquet (1994)
Triade-Flanders	ICP- AES/AAS	Boiling reflux with aqua regia	TRIADE (2000)
US EPA 3050B	FLAA/ICP- AES	Nitric acid, hydrogen peroxide and HCl digestion by either microwave, hot plate or block digester	US EPA (1996)
US EPA 3050A	GFAA/ICP-	Microwave assisted digestion with	US EPA (2007)
	MS/FLAA/I	concentrated nitric acid	
	CP-AES		
UK EA	ICP-MS	Digestion with boiling aqua regia	EA UK (2006)
	ICP-OES	Digestion with boiling aqua regia	
	CV-AFS	Aqua regia under reflux]
Swiss Soil	ICP-OES	Hot extraction with nitric acid	OSol (2016)
Ordinance (OSol)		2 M	

3.2. Environmental concentrations

The environmental concentrations, here referred to as ambient concentrations, of metals in sediment are dependent on geological and anthropogenic inputs in each watershed. Due to geochemical differences, the natural contribution to metal concentrations in sediments may vary from one region to another. It is therefore necessary to estimate the natural (background) concentrations in sediments in order to develop EQS_{sed} relevant for the region of application. A review of available data relevant for the estimation of the natural background concentration of Cu in sediments from Swiss water bodies is provided in Table 5.

No field campaign or project has been dedicated specifically to develop natural (background) concentrations of Cu in Swiss sediments. Two different types of data relevant for evaluating the natural contribution to Cu concentrations in sediments can be used:

 Natural (background) concentrations measured in (pre-industrial) sediments from lakes, most commonly quantified in lake sediment cores. The concentrations retained as background values are ideally those measured in the layer dated ca. 1850. These concentrations range from 22 mg/kg d.w. in Lake Zurich to 45 mg/kg d.w. in Lake Geneva.



Concentrations that are statistically representative of environmental concentrations not affected by anthropogenic sources of pollution. These include the concentrations in sediments reported for Switzerland in the atlas of the Forum of the European Geological Surveys⁶ (Salminem and Batista 2005), which has been most commonly used as default methodology. The mean concentration for streams (fraction < 150 µm) is set at 15.3 mg/kg d.w. while the average for floodplains (< 2 mm) is 21.7 mg/kg d.w. These concentrations are similar to the 10th percentile of a large database of ambient sediment concentrations measured in Swiss sediments, which is 19 mg/kg d.w. and mostly refer to the fraction < 63 µm.

Ambient concentrations of Cu in sediments from a large database as reviewed in Casado-Martinez et al. (2016) range from 1.3 to 1068 mg/kg d.w., with median concentration of 42 mg/kg d.w. These concentrations most often refer to sediments < 63 μ m and quantification after *aqua regia* extraction. A recent study from the Ecotox Centre quantified Cu concentrations in total sediments (< 2 mm) from Swiss waterbodies ranging from 40 to 80 mg/kg d.w. and concentrations between 208 and 307 mg/kg d.w. at highly impacted sites.

Natural concentrations	Concentrations [mg/kg d.w.]	Comments	Reference
Generic value			
	Bed sediments in streams:	N=10	
	15 (Mean)	Fraction < 150 μm	
FOREGS database	8-29 (min-max)	Aqua regia extraction	<u>http://www.gtk.fi/publ/f</u>
TOREOS database	Bed sediments in floodplains:	N=10	<u>oregsatlas</u>
	22 (Mean)	Fraction < 2 mm	
	12-54 (min-max)	Aqua regia extraction	
Lakes			
Constance	28-34	Dated cores	
Lucerne	30	HNO ₃ /HClO ₄ /HF	Reviewed in Casado- Martinez et al. (2016)
Zurich	22	extraction	
Geneva	45	Dated cores HNO₃/HClO₄/HF extraction	Thevenon et al. (2011)
	30.7	Date core HNO₃ extraction (OSol)	Tosono (2017)
Ambient	Concentrations	Comments	Peference
concentration	[mg/kg d.w.]	comments	Kelerence
Fine sediment (< 63 μm)		
	42 (Median)	N=593	
Ecotox Centre	1.3-1068 (Min-Max)	Mostly fraction < 63	Casado-Martinez et al.
database	13 (5 th PC)	μm and extraction with	(2016)
	19 (10 th PC)	aqua regia	
Total sediment (< 2 mn	ו)		
Low-middle impacted s	ites		
Lienne StLéonard	40-80		
Jona nach Rüti	45	Bed sediment	Ecotox centre (unnubl
Doubs	47-57	Extraction with aqua	data)
Birse Reconvilier	50	regia	
Sihlsee	76		

Table 5 Measured environmental concentrations (MEC) of Cu in sediments from Switzerland.

⁶ <u>http://www.gtk.fi/publ/foregsatlas</u>



Natural concentrations	Concentrations [mg/kg d.w.]	Comments	Reference
High impacted sites			
Klausbach	208	Ded as diment	
Wiese	307	Bed sediment	Ecotox centre (unpublished data)
Birs	288		
Seegraben	208	regiu	

4 Effect data (spiked sediment toxicity tests)

Effect data for benthic organisms up to 2008 were collected from the extensive review performed for the Voluntary Risk Assessment Report for Copper metal (ECI 2008). Studies that were rejected in the VRAR (ECI 2008) for PNEC derivation were not assessed for reliability and relevance and are not included here. Relevance ("C" score in the table below) and reliability ("R" score in the table below) of studies that were accepted by the VRAR (ECI 2008) were further assessed for reliability and relevance according to the more recent CRED criteria (Moermond et al. 2016, Casado-Martinez et al. 2017) based on information reported in ECI (2008) and ECHA (2020).

A complementary bibliographic search was performed in the U.S. Ecotox Data Base (US EPA 2016) as well as a key word search on Scopus for publications from 2008. The search returned 229 references that were checked and assessed for relevance and reliability.

Appendix 1 summarizes effect data as total concentration and OC normalized concentration. Studies were performed on artificial and natural sediments with different concentrations of AVS and OC, also reported in Appendix 1 when available. Studies performed with natural sediments where AVS and OC concentrations in test sediments were not reported (e.g. Milani et al. 2003 cited in ECI 2008) were classified as R4 (not enough information is available) and were not used in EQS derivation.

According to the EU TGD (EC 2018) "What is considered chronic or acute is very much dependent on 1) the species considered and 2) the studied endpoint and reported criterion". According to EFSA, true chronic tests should cover a range of 28-65 d when half-life of a pesticide in sediment is >10 d (EFSA 2015). All chronic data presented here are considered true chronic and therefore are considered relevant for sediment EQS derivation with the exception of the 21 d test with *Hexagenia* sp., which is classified as R4 according to missing information on natural sediment properties.

According to the UE TGD (EC 2018), the concentration in the overlying water during the equilibration period should be measured in semi-static and static sediment toxicity tests and testing should preferably only be initiated when the metal concentration is stable. Here, the studies performed by Ghent University (2004) cited in the VRAR (ECI 2008) were initiated after 7 d of equilibration at exposure conditions but no information on measured concentrations in the different compartments is reported. These data is considered reliable with restrictions and is used for EQS derivation, as the VRAR for Cu (ECI 2008) was based on these data. The study with *Tubifex tubifex* by Vecchi et al. (1999) cited in the VRAR (ECI 2008) was initiated after 14 d of aging and performed under static conditions. Because no information but retained as supportive information. The study with *Potamopyrgus antipodarum* by Pang et al. (2013) was performed under semi-static conditions, aging is not reported and overlying water concentrations were only measured at the end of exposure. This study is considered not reliable and is not used for EQS derivation but retained as supportive information.



Additionally effect data for three estuarine and marine species were provided in Sahlin and Ågerstrand (2018) for the derivation of EQS for marine sediments. Effect data were available for the marine oligochaete *Neanthes arenaceodentata*, the amphipods *Leptocheirus plumosus* and *Melita plumosa*, the copepod *Nitocra spinipes* and the mollusc *Hydrobia ulvae*. Given that the database of effect concentrations for freshwater organisms is extensive and covers these groups of organisms, marine and/or estuarine data have not been further considered in EQS derivation here.

AVS values in test sediments ranged from 0.05 to 56.4 mmol/kg d.w. As discussed in section 2.2 for EQS derivation only test data in which bioavailability is maximized are retained. Following the VRAR (ECI 2008) and EC (2016), only NOEC values generated with sediments that had AVS concentration lower than 0.77 mmol/kg d.w. are retained for EQS derivation. Therefore all effect data derived from studies with sediments with AVS > 0.77 mmol/kg d.w. are classified as C3, not relevant for EQS derivation.

4.1. Graphic representation of effect data

Figure 2 summarizes chronic data in the data set, before and after normalization for OC concentrations in test sediments.

According to the reliable data from chronic tests (Fig. 2), there is no specific group that is significantly more sensitive to Cu. Considering all chronic data in the data set, effect concentrations fall within the same range for the different taxonomic groups, both before and after normalization for OC.

Regarding normalization to OC, normalization for OC in test sediments reduces the intra-specific variability for all endpoints for *C. riparius*, *H. azteca* and *T. tubifex*. For *L. variegatus* and *G. pulex* test sediments presented similar OC content and comparisons were not possible.



Figure 2 Graphical representation of chronic effect data from spiked sediment toxicity tests with Cu for



freshwater organisms before (A) and after OC normalization (B). Empty symbols are data that are not used for EQS derivation according to reliability and relevance assessment.

4.2. Comparison between marine and freshwater species

According to the EU TGD p. 39 (EC 2018), freshwater and saltwater data for metals should be separated a priory and should only be combined when there is no demonstrable difference in sensitivity. As stated above, marine and/or estuarine data have not been further considered in EQS derivation here.

4.3. Overview of reliable and relevant long-term studies

According to the EU TGD (EC (2018) p. 25): "All available data for any taxonomic group or species should be considered, provided the data meet quality requirements for relevance and reliability". Relevant and reliable chronic data is available for 5 species (Table 6).

Table 6 Geometric mean of effect concentrations from relevant and reliable chronic studies from Appendix 1. All values in mg/kg d.w. Most sensitive endpoint is underlined and used for EQS derivation.

Species	Exposure duration [d]	Endpoint	NOEC [not normalized, OC < 3%]*	NOEC [1% OC, all data]
Gammarus pulex	35	Growth	<u>54.6 (N=2)</u>	<u>19.7 (N=2)</u>
	35	Survival	96.0 (N=2)	35.2 (N=2)
Hyalella azteca	28	Growth	<u>38.7 (N=3</u>)	<u>23.2 (N=4)</u>
	28	Survival	140.5 (N=3)	57.0 (N=3)
Chironomus riparius	28	Emergence	<u>59.5 (N=1)</u>	<u>26.0 (N=2)</u>
	28	Growth	67.1 (N=4)	32.2 (N=5)
	28	Survival	83.6 (N=4)	34.3 (N=5)
Lumbriculus variegatus	28	Biomass	<u>86.0 (N=2)</u>	<u>37.9 (N=2)</u>
Tubifex tubifex	28	Growth	<u>39.7 (N=4)</u>	<u>21.7 (N=4)</u>
	28	Survival	89.3 (N=3)	40.5 (N=4)
	28	Reproduction	53.2 (N=4)	27.4 (N=5)

* Effect concentrations based on total Cu concentration from tests with a much higher sediment OC content were not included when deriving the geometric mean.

5 Derivation of QS_{sed}

According to the EU TGD for EQS, sediment toxicity tests, aquatic toxicity tests in conjunction with equilibrium partitioning (EqP) and field/mesocosm studies are used as several lines of evidence to derive QS_{sed} (EC 2018). Thus, in the following, the appropriateness of the deterministic approach (AF-Method), the probabilistic approach (SSD method) and the EqP approach were examined.

5.1. Derivation of QS_{sed, AF} using the Assessment Factor (AF) method

The $QS_{sed, AF}$ is derived using assessment factors (AFs) applied to the lowest credible datum from long-term toxicity tests.

The lowest long-term effect datum available for Cu is the NOEC (geomean, N=3) of 38.7 mg/kg d.w. for the growth of *H. azteca* without OC normalization. This NOEC is used to derive a $QS_{sed,AF,generic}$. The



lowest long-term effect datum available for Cu after OC normalization is the NOEC (geomean, N=2) of 19.7 mg/kg d.w. for 1% OC for the growth of *Gammarus pulex*.

In case of long term tests (NOEC or EC_{10}) being available for three species representing different living and feeding conditions, the EU TGD recommends the application of an assessment factor of 10 on the lowest credible datum (Table 11 in EC (2018)). The exact value of the AF depends on an evaluation of the uncertainties, which are summarized below (a full uncertainty analysis is available at the VRAR (ECI 2008):

- All the endpoints considered are relevant at the population level: mortality, growth, reproduction, and emergence.
- The tests are all considered long-term chronic exposures ranging from 28 to 42 days.
- The age of the test organisms at the start of the test represent the most sensitive life stage, i.e. adult oligochaetes are used for the reproduction test whereas growth was assessed in juveniles/larvae.
- The data base used was extracted from tests performed in a variety of natural/artificial freshwater sediments, covering a considerable part of the wide range of the sediment characteristics that are normally found in European freshwater sediments. The retained ecotoxicological data corresponded to sediments with high bioavailability, i.e. with AVS concentrations between 0.05 0.59 mmol/kg. Organic Carbon content of sediments cover a broad range, ranging from 0.5 3% the data used for deriving the QS_{sed,generic} while up to 10% the QS_{sed,bioav}.
- There is some uncertainty in whether the conditions of sediments of the database retained are protective for the conditions found in Swiss sediments. AVS values in Switzerland come from three wadeable pristine head streams, ranging from 0.006-0.02 mmol/Kg dry weight. These values are lower than the 0.77 mmol/Kg dry weight threshold set in the VRAR (ECI 2008) but effect data did not show any relationship with AVS concentrations in the low AVS range accepted.
- The NOEC data belong to 5 different species relevant for the sediment compartment with different living modes and feeding strategies plus two additional species for which supportive information is available (all in Table 7). Sediment relevant autotrophs and bivalves are missing in this database, although available data indicate that higher plants are less sensitive than pelagic phyto- and zooplankton (ECI 2008) and results from a mesocosm study include these types of organisms (section 7.1).

 Table 7 Characteristics and traits describing the species for which effect data is considered.

Species	Living mode	Feeding mode	Habitat	Exposure phase
Hyalella azteca	Epi-benthic,	Periphyton,	Small rivers,	Mainly overlying
	occasional	algae and	non-	water,
	burrower	detritus at the	autochthonous	resuspended
		sediment-		particles not
		water		excluded
		interface		
Gammarus pulex	Epi-benthic,	Algae and	Small rivers	Mainly overlying
	occasional	detritus	and streams	water,
	burrower			resuspended
				particles not
				excluded



Species	Living mode	Feeding mode	Habitat	Exposure phase
Chironomus	Endo-benthic,	Surface	Eutrophic	Sediment, pore-
riparius	U-shaped	sediment	lakes, ponds	water and
	burrow	ingesting	and streams	overlying water
	irrigated with	deposit-		
	oxygenated	feeder		
	water			
Lumbriculus	Endo-benthic	Sub-surface	Pods, lakes	Sediment, pore-
variegatus		deposit	and marshes	water and
		feeders		overlying water
Tubifex tubifex	Endo-benthic,	Sediment-	Lakes, ponds	Sediment and
	not irrigated I-	ingesting	and rivers	pore-water
	shaped burrow	deposit-		
		feeder		
Hexagenia sp.	Endo-benthic,	Sediment,	Lakes, ponds,	Sediment
	irrigated	detritus and	wetlands,	ingestion and
	burrow	organic	streams and	overlying water
		matter-	rivers.	
		ingestion,		
		filter-feeding		
		of seston		
Potamopyrgus	Epi-benthic,	Grazer,	Rivers, lakes,	Mainly overlying
antipodarum	occasional	periphyton,	streams,	water, sediment
	burrower	diatoms, and	reservoirs,	ingestion nor
		plant and	lagoons,	excluded
		animal	canals, ditches	
		detritus		

An AF of 2 is proposed, as a higher AF would lead to a $QS_{sed,AF}$ lower than the Cu background concentration in Swiss sediments:

$$QS_{sed,AF} = \frac{lowest \ EC10 \ or \ NOEC}{AF}$$
$$QS_{sed,AF,generic} = \frac{38.7 \ \left(\frac{mg}{kg}\right)}{2} = 19.4 \ \left(\frac{mg}{kg}\right)$$
$$QS_{sed,AF,bioav} = \frac{19.7 \ \left(\frac{mg}{kg}\right)}{2} = 9.85 \ \left(\frac{mg}{kg}\right)$$

5.2. Derivation of QS_{sed,SSD} using the species sensitivity distribution (SSD) method

The minimum data requirements recommended for the application of the SSD approach for EQS water derivation is preferably more than 15, but at least 10 NOEC/EC₁₀, from different species covering at least eight taxonomic groups (EC (2018), p. 43). Specific recommendations for the sediment compartment are only available to some extent in the ECHA Proceedings of the Topical Scientific



Workshop Principles for Environmental Risk Assessment of the Sediment Compartment (Tarazona et al. 2014) but further recommendations on the minimum data requirements are not fixed.

The SSD approach has been previously applied for the derivation of sediment PNECs in the VRAR for Copper (ECI 2008) with 6 data from 6 species (trimmed data set of 63 NOECs), using the effect concentrations from *Hexagenia* sp. assessed as R4 here. The data requirements were not met but the VRAR considered more appropriate to derive the PNEC freshwater sediment using the statistical extrapolation methodology (ECI 2008), decision supported by the SCHER (2009) according to existing literature data (weight of evidence) although the SCHER stated that it "should be considered as indicative" and the "statistical and ecological significance of the SSD approach, based on a small number of species, is quite weak".

Recently a Danish draft report for sediment EQS derivation also used the SSD approach to derive a sediment EQS for zinc including effect data for 12 species representing 6 systematic groups where insects were represented by two different orders as Diptera and Ephemeroptera have widely different ecology and feeding strategies, Macrophyta, Oligochaeta, Polychaeta and Amphipoda (Crustacea). Sahlin and Ågerstrand (2018), who derived EQS_{sed} for Cu, considered that the data set was not enough to derive a QS_{sed,SSD}.

The SSD was performed here as additional line of evidence for sediment EQS derivation. Although the same dataset is used for both the deterministic and probabilistic methods (EU TGD p. 44), here the SSD used all effect concentrations for freshwater species in Table 6 plus the geometric mean of NOECs for *Hexagenia* sp. and the NOEC for *P. antipodarum*. The results of the SSD for generic and bioavailable QS derivation are included in Fig. 3 and Appendix 2.



Figure 3 Species sensitivity distribution (SSD) of the chronic effect concentrations of all species for Cu generated with ETX 2.2 before (left) and after OC normalization to 1% OC (right). Number of data points N = 6.

The median fifth percentile (HC₅) was calculated with the software package RIVM ETX 2.2 (van Vlaardingen et al. 2004), resulting in 27.7 mg/kg d.w. (lower and upper 90 % confidence limits 15.8-36.5 mg/kg, standard deviation of the log₁₀ transformed values = 0.15) before OC normalization and 14.3 mg/kg d.w. (lower and upper 90 % confidence limits 6.59-21.0 mg/kg, standard deviation of the log₁₀ transformed values = 0.20) after normalization to 1% OC. The data fitted a lognormal model (Fig. 3), the Anderson-Darling, Cramer von Mises, and Kolmogorov-Smirnov tests for normality are accepted at all significant levels. Note that as the number of data points increases data distribution may fail a goodness of fit test. It is recognized that below 8 entries the Anderson-Darling may not perform well, while the Cramer von Mises and Kolmogorov-Smirnov tests do not perform well when the number of data is less than 20.



According to the EU TGD for EQS, an SSD should be based on the most sensitive groups of species (EC 2018). From the effect data base available for benthic organisms there is no indication of a group that is particularly sensitive to Cu. According to information from water-only tests, primary producers are the most sensitive taxonomic group to Cu.

According to the TGD (EC 2018), an AF to account for residual uncertainty divides the HC₅. An AF=5 is used by default but may be reduced where evidence removes residual uncertainty.

Taking into consideration the effect database used, uncertainty analysis and the different proxies for background concentrations of Cu in Swiss sediments from the available literature, an AF of 2 is proposed:

$$QS_{sed,SSD,generic} = \frac{27.7}{2} = 13.9 \frac{\text{mg}}{\text{kg}} \text{ d.w.}$$

$$QS_{sed,SSD,bioav} = \frac{14.3}{2} = 7.2 \frac{\text{mg}}{\text{kg}} \text{ d.w.}$$

6 Derivation of QS_{sed,EqP} using the Equilibrium Partitioning approach

If no reliable sediment toxicity data are available, the Equilibrium Partitioning (EqP) can be used to estimate the $QS_{sed,EqP}$. This approach, developed for non-ionic substances, is used here for comparison purposes.

6.1. Selection of QS for water

The EqP model has been applied using the limit value set in Switzerland for Cu in freshwater at 2 μ g/L for harmonization among quality guidelines at the national level.

Additionally the most recent EQS for freshwaters of 2.8 μ g/L derived by the ICPR (2016) using the EU TGD (2011) is used, given that the limit value from Switzerland is not derived using the EU TGD approach.

6.2. Selection of partition coefficient

One of the main factors influencing the application of the EqP model is the choice of the partition coefficient. It is stipulated in the ECHA 2017 guideline (p. 143, ECHA (2017)) that "To increase the reliability of PNEC sediment screen derived using the EqP, it is imperative that a conservative but realistic partitioning coefficient (e.g. K_d , K_{oc} , K_{ow}) is chosen. A clear justification must be given for the chosen coefficient and any uncertainty should be described in a transparent way."

The EU TGD prefers measured Kp values for sediment/suspended matter for freshwater, estuarine and marine water bodies respectively (EC 2018). Preference is given to field measurements and not laboratory sorption or toxicity experiments.

The K_p estimated for Cu selected here for deriving a $QS_{sed,EqP}$ is 24 409 L/kg which is the median from literature values in the VRAR and cited in ECHA (2020). Given that the $QS_{sed,EqP}$ is derived for comparison purposes no additional K_p values have been searched.

6.3. Derivation of QS_{sed,EqP}

The derivation of $QS_{sed,EqP}$ is summarized in Table 8, resulting in 48.8 mg/kg d.w. derived from the Swiss limit value of 2 µg/L and 68.4 mg/kg d.w. derived from the EQS for freshwaters of 2.8 µg/L.



An additional AF of 10 should be applied to the resulting $QS_{sed,EqP}$ for substances with log K_{ow} >5. No additional AF is warranted for Cu.

Kp _{sed} [L/kg]	K _{sed-water} [m ³ /m ³]	PNEC _{water} [mg/L]	QS _{sed,EqP} [mg/kg w.w.]	QS _{sed,EqP} [mg/kg d.w.]	Additional AF
24 409	12 205	0.002	18.8	48.8	
24 409	12 205	0.0028	26.3	68.4	

Table 8 Derived QS_{sed,EqP} according to estimated Kp for sediments from Table 1. No additional AF was applied.

7 Determination of QS_{sed} according to mesocosm/field data

7.1. Overview of available mesocosm/microcosm studies

One mesocosm study has reported the effect of Cu on primary producers and OM decomposition in mesocosms studies with spiked sediments at four different Cu concentrations and controls, each with 4 replicates (Gardham et al. 2015). The time -average sediment concentrations were 5.8, 62, 97, 310 and 650 mg/kg d.w. The biological endpoints assessed in Gardham et al. (2015) were the growth of the rooted-submerged macrophyte *Vallisneria spiralis*, chlorophyll a concentrations in the phytoplankton (diatoms, cyanobacteria and green algae), periphyton biomass, and leaf litter and cotton strip assays to assess OM decomposition.

The companion paper by Gardham et al. (2014a) reported changes in benthic and overlying water invertebrate community sampled every month during the initial 6 months, and then at 12 months, with time-average sediment concentrations of 4.6, 71, 99, 410 and 711 mg/kg d.w. in the same mesocosm study as Gardham et al. (2015). Gardham et al. (2014b) described Cu partitioning and distribution among abiotic compartments in the mesocosm experiments.

Significant effects in macrophyte shoot density of *Vallisneria spiralis* were observed at 310 and 650 mg/kg d.w. after 24 weeks and at 97, 310 and 650 mg/kg d.w. A NOEC of 62 mg/kg d.w. is therefore derived from macrophyte shoot density. At 310 and 650 mg/kg d.w. a decrease of grazing pressure due to changes in the abundance of an important grazer, the gastropod *Physa acuta*, was related to an increase in periphyton biomass (Gardham et al. 2015). At the two highest exposure concentrations, there was also a clear difference in the composition of the benthic communities compared with control exposures during the colonization period (Gardham et al. 2014a). Richness, abundance, and structure of the benthic invertebrate assemblages in particular were strongly affected by Cu contamination with benthic Chironominae, a species of Ostracoda, Cladocera, and *P. acuta* being particularly sensitive to Cu. The estimated NOEC for benthic invertebrate assemblages was set at 97 mg/kg d.w. while for macrophyte shoot density it was set at 62 mg/kg d.w. in sediment with approx. 2% OC. This mesocosm NOEC is not used for EQS_{sed} derivation but is used as supportive information for effect concentrations in taxonomic groups not represented in the effect data set (e.g. primary producers, macrophytes).

7.2. Available sediment quality guidelines from field data

There are several sediment quality guidelines based on field data that have been developed over the years (Table 9). The field SQGs based mainly on macrozoobenthos range from 13 to 70 mg/kg d.w. while the TEL_{oligo} derived from field data of oligochaete communities in small and medium streams in Switzerland is slightly below this range. The consensus-based Threshold Effect Concentration (TEC) from MacDonald et al. (2000), widely used for freshwater sediment quality assessments, is set at 31.6 mg/kg d.w. The range of field SQGs also fall within the range of effect concentrations in the data set from spiked sediment toxicity tests. There is also good agreement between field SQGs and the HC₅



obtained from the SSD approach, with the lower estimate of the HC₅ matching the lowest field SQGs. These SQGs include indirectly the background concentration found in the areas where these sediments were collected and are not normalized or include additional bioavailability corrections. Therefore they are considered generic and total SQGs.

SQG	Value [mg/kg d.w.]	Reference	Description
Threshold effect level (TEL)	35.7	Smith et al. (1996)	Sediments are considered to be clean to marginally polluted. No effects on most sediment-dwelling organisms expected below this concentration.
Lileky effect level (LEL)	16	Persaud et al. (1993)	Concentration below which adverse effects are expected to occur only rarely.
Environmental risk limit (ERL)	70	Long and Morgan (1991)	Chemical concentration below which adverse effects would be rarely observed.
Lowest effect level (LEL)	13	de Deckere et al. (2011)	Concentration below which adverse effects on macrozoobentos is rarely observed. A SEL of 85 mg/kg d.w. describes concentration above which macrozoobentos is likely affected.
Threshold effect level (TEL _{oligo})	10.5	Vivien et al. (2020)	Concentration below which oligochaete communities are rarely affected. A PEL _{oligo} of 31 mg/kg d.w. describes concentrations above which oligochaete communities are likely affected.
Threshold effect concentration (TEC)	31.6	MacDonald et al. (2000)	Threshold effect concentration for benthic organisms.

Table 9 Sediment quality guidelines available in the literature based on field data.

8 Toxicity of degradation products

Not relevant for metals.

9 EQS_{sed} proposed to protect benthic species

The different QS values for each derivation method included in the EU TGD (2018) are summarized in Table 10. According to the TGD, the most reliable extrapolation method for each substance should be used (EC 2018). In all cases, data from spiked sediment toxicity tests are preferred over the EqP approach.

A generic $EQS_{sed,generic}$ of 19.4 mg/kg d.w. is proposed, together with a bioavailable $EQS_{sed,bioav}$ of 9.9 mg/kg d.w. for 1% OC that can be used to account for changes in bioavailability due to OC in field sediments. The generic EQS_{sed} is in the low range of relevant concentrations to assess the natural background.



Table 10 QS_{sed} derived according to the three methodologies stipulated in the EU TGD and their corresponding AF, and additional thresholds values for comparison purposes. All concentrations expressed as mg/kg d.w.

	Generic	Bioavailable (1% OC)	AF
QS _{sed,SSD}	13.9	7.2	2
QS _{sed,AF}	19.4	9.9	2
QS _{sed,EqP}	48.8-68.4		
Field SQG	10.5-70		
Background	15-45		
concentrations			
Proposed EQS _{sed}	19.4	9.9	2

9.1. Protection of benthic organisms and uncertainty analysis

The proposed EQS_{sed,generic} is lower or close to existing sediment quality guidelines and thresholds based on field data, thus it should be protective for benthic communities and macrozoobenthos. The TEL_{oligo}, which is derived from total Cu concentrations, is slightly lower than the proposed EQS_{sed,generic} and matches the EQS_{sed,bioav} for 1% OC. Because a) the TEL_{oligo} was derived from field data for small and medium water bodies and limited data is available to determine statistically background concentrations in this type of water bodies, and b) according to Table 6 oligochaetes were not more sensitive that other benthic organisms, it is concluded that there is not enough evidence to state that the proposed EQS_{sed} is not protective for oligochaetes.

The lack of effect concentrations for primary producers is compensated by effect concentrations from a mesocosm study (section 7.1). The proposed $EQS_{sed,generic}$ is also well below the NOEC for primary producers and macrophytes.

The EQS_{sed,generic} was derived with effect concentrations from sediments with AVS that may not be protective for head streams and without further OC normalization. It is recommended to use the EQS_{sed,bioav} to assess Cu concentrations at this type of sites. This would also protect oligochaete communities, which are present in head streams in Switzerland, from adverse effects.

It is possible that the proposed EQS_{sed}, both generic and bioavailable, are below the local/regional background. For implementation, the following tiered approach is proposed (Fig. 4), which is adapted from the approach recommended for metal EQS implementation for waters at EU level when a bioavailability model is available (EC 2019).





Figure 4 Approach used for the development and implementation of EQS_{sed} for Cu (adapted from EC 2019).

10 References

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Appendix I. Effect data

Table A1.1 Sediment effect data for Cu. Data were evaluated for relevance and reliability according to the CRED criteria for sediments (Casado-Martinez et al. 2017) adapted based on Moermond et al. (2016). All concentrations in mg/kg d.w. Data not used in EQS derivation in grey. Underlined data are those included in the dataset for EQS derivation.

Group	Species	Test compou nd	Exposure	Equilibration time	Endpoint	Test duration	Effect concentra tion	Total concent ration	Normalized concentration (1% OC)	Sediment type	Chem. analysis	Note	Validity	References
Crustacean Amphipoda	Gammarus pulex	CuCl ₂ . 2H ₂ 0	Static renewal	7 d	Survival	35 d	NOEC	94.7	36.1	Artificial OECD sediment: AVS 0.05 mmol/kg, pH 7.2-7.8, OC 2.62%	Measu red	No information chemical analysis during equilibration reported	R2/C1	Ghent University, 2004 cited ECI 2008
Crustacean Amphipoda	Gammarus pulex	CuCl₂∙ 2H₂0	Static renewal	8-11 d	Survival	35 d	NOEC	97.4	34.4	Natural sediment: AVS 0.21 mmol/kg, pH 7.2- 7.8, OC 2.83%	Measu red	"	R2/C1	Ghent University, 2004 cited ECI 2008
								<u>96.0</u>	<u>35.2</u>					
Crustacean Amphipoda	Gammarus pulex	CuCl ₂ . 2H ₂ 0	Static renewal	8-11 d	Growth	35 d	NOEC	30.6	10.8	Natural sediment: AVS 0.21 mmol/kg, pH 7.2- 7.8, OC 2.83%	Measu red	No information chemical analysis during equilibration reported	R2/C1	Ghent University, 2004 cited ECI 2008
Crustacean Amphipoda	Gammarus pulex	CuCl ₂ · 2H ₂ 0	Static renewal	7 d	Growth	35 d	NOEC	94.7	36.1	Artificial OECD sediment: AVS 0.05 mmol/kg, pH 7.2-7.8, OC 2.62%	Measu red	u	R2/C1	Ghent University, 2004 cited ECI 2008
								<u>53.8</u>	<u>19.7</u>					
Crustacean Amphipoda	Gammarus pulex	CuCl ₂ . 2H ₂ 0	Static renewal	8-11 d	Survival	35 d	NOEC	1 268	195.7	Natural sediment: AVS 17.5 mmol/kg, pH 7.2- 7.8, OC 6.48%	Measu red	No information chemical analysis during equilibration reported	R2/C3	Ghent University, 2004 cited ECI 2008



Group	Species	Test compou nd	Exposure	Equilibration time	Endpoint	Test duration	Effect concentra tion	Total concent ration	Normalized concentration (1% OC)	Sediment type	Chem. analysis	Note	Validity	References
Crustacean Amphipoda	Gammarus pulex	CuCl ₂ · 2H ₂ 0	Static renewal	8-11 d	Growth	35 d	NOEC	789	113.2	Natural sediment: AVS 17.5 mmol/kg, pH 7.2- 7.8, OC 6.97%	Measu red	u	R2/C3	Ghent University, 2004 cited ECI 2008
Crustacean Amphipoda	Hyalella azteca	CuCl₂∙ 2H₂0	Static renewal	8-11 d	Survival	28 d	NOEC	140	71.4	Natural sediment: AVS 0.10 mmol/kg, pH 7.2- 7.8, OC 1.96%	Measu red	No information chemical analysis during equilibration reported	R2/C1	Ghent University, 2004 cited ECI 2008
Crustacean Amphipoda	Hyalella azteca	CuCl₂∙ 2H₂0	Static renewal	8-11 d	Survival	28 d	NOEC	141	66.5	Natural sediment: AVS 0.28 mmol/kg, pH 7.2- 7.8, OC 2.12%	Measu red	"	R2/C1	Ghent University, 2004 cited ECI 2008
Crustacean Amphipoda	Hyalella azteca	CuCl ₂ . 2H ₂ 0	Static renewal	7 d	Survival	28 d	NOEC	337.6	34.9	Artificial OECD sediment: AVS 0.27 mmol/kg, pH 7.2-7.8, OC 9.66%	Measu red	u	R2/C1	Ghent University, 2004 cited ECI 2008
								<u>140.5</u>	<u>57.0</u>					
Crustacean Amphipoda	Hyalella azteca	CuCl ₂ . 2H ₂ 0	Static renewal	8-11 d	Growth	28 d	NOEC	49.9	25.5	Natural sediment: AVS 0.10 mmol/kg, pH 7.2- 7.8, OC 1.96%	Measu red	No information chemical analysis during equilibration reported	R2/C1	Ghent University, 2004 cited ECI 2008
Crustacean Amphipoda	Hyalella azteca	CuCl ₂ · 2H ₂ 0	Static renewal	7 d	Growth	28 d	NOEC	53.2	20.3	Artificial OECD sediment: AVS 0.05 mmol/kg, pH 7.2-7.8, OC 2.62%	Measu red	u	R2/C1	Ghent University, 2004 cited ECI 2008
Crustacean Amphipoda	Hyalella azteca	CuCl ₂ . 2H ₂ 0	Static renewal	8-11 d	Growth	28 d	NOEC	21.8	10.3	Natural sediment: AVS 0.28 mmol/kg, pH 7.2- 7.8, OC 2.12%	Measu red	u	R2/C1	Ghent University, 2004 cited ECI 2008
Crustacean Amphipoda	Hyalella azteca	CuCl ₂ . 2H ₂ 0	Static renewal	7 d	Growth	28 d	NOEC	538.6	55.8	Artificial OECD sediment: AVS 0.27 mmol/kg, pH 7.2-7.8, OC 9.66%	Measu red	u	R2/C1	Ghent University, 2004 cited ECI 2008
								38.7	23.2					



Group	Species	Test compou nd	Exposure	Equilibration time	Endpoint	Test duration	Effect concentra tion	Total concent ration	Normalized concentration (1% OC)	Sediment type	Chem. analysis	Note	Validity	References
Crustacean Amphipoda	Hyalella azteca	CuCl ₂ . 2H ₂ 0	Static renewal	7 d	Survival	28 d	NOEC	292.5	88.9	Artificial OECD sediment: AVS 4.87 mmol/kg, pH 7.2-7.8, OC 3.29%	Measu red	No information chemical analysis during equilibration reported	R2/C3	Ghent University, 2004 cited ECI 2008
Crustacean Amphipoda	Hyalella azteca	CuCl ₂ · 2H ₂ 0	Static renewal	7 d	Growth	28 d	NOEC	292.	88.9	Artificial OECD sediment: AVS 4.87 mmol/kg, pH 7.2-7.8, OC 3.29%	Measu red	u	R2/C3	Ghent University, 2004 cited ECI 2008
Crustacean Amphipoda	Hyalella azteca	CuCl ₂ · 2H ₂ 0	Static renewal	7 d	Survival	28 d	NOEC	582.6	177.1	Artificial OECD sediment: AVS 12.3 mmol/kg, pH 7.2-7.8, OC 3.29%	Measu red	u	R2/C3	Ghent University, 2004 cited ECI 2008
Crustacean Amphipoda	Hyalella azteca	CuCl ₂ . 2H ₂ 0	Static renewal	7 d	Survival	28 d	NOEC	739.5	76.6	Artificial OECD sediment: AVS 5.30 mmol/kg, pH 7.2-7.8, OC 9.66%	Measu red	u	R2/C3	Ghent University, 2004 cited ECI 2008
Crustacean Amphipoda	Hyalella azteca	CuCl ₂ . 2H ₂ 0	Static renewal	7 d	Growth	28 d	NOEC	492.7	51.0	Artificial OECD sediment: AVS 5.30 mmol/kg, pH 7.2-7.8, OC 9.66%	Measu red	u	R2/C3	Ghent University, 2004 cited ECI 2008
Crustacean Amphipoda	Hyalella azteca	CuCl ₂ . 2H ₂ 0	Static renewal	7 d	Survival	28 d	NOEC	849.5	87.9	Artificial OECD sediment: AVS 8.97 mmol/kg, pH 7.2-7.8, OC 9.66%	Measu red	u	R2/C3	Ghent University, 2004 cited ECI 2008
Crustacean Amphipoda	Hyalella azteca	CuCl ₂ · 2H ₂ 0	Static renewal	7 d	Growth	28 d	NOEC	512.2	53.0	Artificial OECD sediment: AVS 8.97 mmol/kg, pH 7.2-7.8, OC 9.66%	Measu red	u	R2/C3	Ghent University, 2004 cited ECI 2008
Crustacean Amphipoda	Hyalella azteca	CuCl ₂ . 2H ₂ 0	Static renewal	8-11 d	Survival	28 d	NOEC	171	60.4	Natural sediment: AVS 0.18 mmol/kg, pH 7.2- 7.8, OC 2.83%	Measu red	u	R2/C3	Ghent University, 2004 cited ECI 2008
Crustacean Amphipoda	Hyalella azteca	CuCl ₂ · 2H ₂ 0	Static renewal	8-11 d	Survival	28 d	NOEC	3 158	167.1	Natural sediment: AVS 58.6 mmol/kg, pH 7.2- 7.8, OC 18.9%	Measu red	u	R2/C3	Ghent University, 2004 cited ECI 2008
Crustacean Amphipoda	Hyalella azteca	CuCl ₂ . 2H ₂ 0	Static renewal	8-11 d	Growth	28 d	NOEC	1 531	81.0	Natural sediment: AVS 58.6 mmol/kg, pH 7.2- 7.8, OC 18.9%	Measu red	u	R2/C3	Ghent University, 2004 cited ECI 2008



Group	Species	Test compou nd	Exposure	Equilibration time	Endpoint	Test duration	Effect concentra tion	Total concent ration	Normalized concentration (1% OC)	Sediment type	Chem. analysis	Note	Validity	References
Crustacean Amphipoda	Hyalella azteca	CuCl ₂ · 2H ₂ 0	Static renewal	8-11 d	Survival	28 d	NOEC	1 495	230.7	Natural sediment: AVS 18.3 mmol/kg, pH 7.2- 7.8, OC 6.48%	Measu red	u	R2/C3	Ghent University, 2004 cited ECI 2008
Crustacean Amphipoda	Hyalella azteca	CuCl ₂ · 2H ₂ 0	Static renewal	8-11 d	Growth	28 d	NOEC	244.8	37.8	Natural sediment: AVS 18.3 mmol/kg, pH 7.2- 7.8, OC 6.48%	Measu red	и	R2/C3	Ghent University, 2004 cited ECI 2008
Crustacean Amphipoda	Hyalella azteca	CuCl ₂ . 2H ₂ 0	Static	14 d	Survival	28 d	NOEC	59.3	118.6	Natural sediment : OC 0.5%	Measu red	AVS not measured	R4/C1	Milani et al. 2003 cited ECI 2008
Crustacean Amphipoda	Hyalella azteca	CuCl ₂ . 2H ₂ 0	Static	14 d	Growth	28 d	NOEC	59.3	118.6	Natural sediment : OC 0.5%	Measu red	AVS not measured	R4/C1	Milani et al. 2003 cited ECI 2008
Crustacean Amphipoda	Hyalella azteca	CuCl ₂ · 2H ₂ 0	Static	14 d	Survival	28 d	NOEC	66.9	133.8	Natural sediment : OC 0.5%	Measu red	AVS not measured	R4/C1	Milani et al. 2003 cited ECI 2008
Crustacean Amphipoda	Hyalella azteca	CuCl ₂ . 2H ₂ 0	Static	14 d	Growth	28 d	NOEC	66.9	133.8	Natural sediment : OC 0.5%	Measu red	AVS not measured	R4/C1	Milani et al. 2003 cited ECI 2008
Crustacean Amphipoda	Hyalella azteca	CuCl ₂ . 2H ₂ 0	Static	14 d	Survival	28 d	NOEC	155.1	310.2	Natural sediment : OC 0.5%	Measu red	AVS not measured	R4/C1	Milani et al. 2003 cited ECI 2008
Crustacean Amphipoda	Hyalella azteca	CuCl ₂ . 2H ₂ 0	Static	14 d	Growth	28 d	NOEC	52.3	104.6	Natural sediment : OC 0.5%	Measu red	AVS not measured	R4/C1	Milani et al. 2003 cited ECI 2008
Insect Chironomidae	Chironomus riparius	CuCl ₂ . 2H ₂ 0	Static renewal	7 d	Survival	28 d	NOEC	59.5	22.7	Artificial OECD sediment: AVS 0.05 mmol/kg, pH 7.2-7.8, OC 2.62%	Measu red	No information chemical analysis during equilibration reported	R2/C1	Ghent University, 2004 cited ECI 2008
Insect Chironomidae	Chironomus riparius	CuCl₂∙ 2H₂0	Static renewal	8-11 d	Survival	28 d	NOEC	177.1	62.6	Natural Sediment: AVS 0.15 mmol/kg, pH 7.2-7.8, OC 2.83%	Measu red	"	R2/C1	Ghent University, 2004 cited ECI 2008
Insect Chironomidae	Chironomus riparius	CuCl ₂ . 2H ₂ 0	Static renewal	8-11 d	Survival	28 d	NOEC	54.2	25.6	Natural Sediment: AVS 0.28 mmol/kg, pH 7.2-7.8, OC 2.12%	Measu red	u	R2/C1	Ghent University, 2004 cited ECI 2008



Group	Species	Test compou nd	Exposure	Equilibration time	Endpoint	Test duration	Effect concentra tion	Total concent ration	Normalized concentration (1% OC)	Sediment type	Chem. analysis	Note	Validity	References
Insect Chironomidae	Chironomus riparius	CuCl ₂ · 2H ₂ 0	Static renewal	8-11 d	Survival	28 d	NOEC	85.4	43.6	Natural Sediment: AVS 0.10 mmol/kg, pH 7.2-7.8, OC 1.96%	Measu red	"	R2/C1	Ghent University, 2004 cited ECI 2008
Insect Chironomidae	Chironomus riparius	CuCl₂∙ 2H₂0	Static renewal	7 d	Survival	28 d	NOEC	292	29.8	Artificial OECD sediment: AVS 0.30 mmol/kg, pH 7.2-7.8, OC 9.81%	Measu red	u	R2/C1	Ghent University, 2004 cited ECI 2008
								<u>83.6</u>	<u>34.3</u>					
Insect Chironomidae	Chironomus riparius	CuCl ₂ . 2H ₂ 0	Static renewal	7 d	Emergence	28 d	NOEC	<u>59.5</u>	22.7	Artificial OECD sediment: AVS 0.05 mmol/kg, pH 7.2-7.8, OC 2.62%	Measu red	No information chemical analysis during equilibration reported	R2/C1	Ghent University, 2004 cited ECI 2008
Insect Chironomidae	Chironomus riparius	CuCl₂· 2H₂0	Static renewal	7 d	Emergence	28 d	NOEC	292	29.8	Artificial OECD sediment: AVS 0.30 mmol/kg, pH 7.2-7.8, OC 9.81%	Measu red	"	R2/C1	Ghent University, 2004 cited ECI 2008
									<u>26.0</u>					
Insect Chironomidae	Chironomus riparius	CuCl ₂ . 2H ₂ 0	Static renewal	7 d	Growth	28 d	NOEC	89.2	34.0	Artificial OECD sediment: AVS 0.05 mmol/kg, pH 7.2-7.8, OC 2.62%	Measu red	No information chemical analysis during equilibration reported	R2/C1	Ghent University, 2004 cited ECI 2008
Insect Chironomidae	Chironomus riparius	CuCl₂· 2H₂0	Static renewal	8-11 d	Growth	28 d	NOEC	75.4	26.6	Natural Sediment: AVS 0.15 mmol/kg, pH 7.2-7.8, OC 2.83%	Measu red	"	R2/C1	Ghent University, 2004 cited ECI 2008
Insect Chironomidae	Chironomus riparius	CuCl ₂ . 2H ₂ 0	Static renewal	8-11 d	Growth	28 d	NOEC	54.4	26.7	Natural Sediment: AVS 0.28 mmol/kg, pH 7.2-7.8, OC 2.12%	Measu red	u	R2/C1	Ghent University, 2004 cited ECI 2008
Insect Chironomidae	Chironomus riparius	CuCl ₂ . 2H ₂ 0	Static renewal	8-11 d	Growth	28 d	NOEC	55.5	28.3	Natural Sediment: AVS 0.10 mmol/kg, pH 7.2-7.8, OC 1.96%	Measu red	u	R2/C1	Ghent University, 2004 cited ECI 2008



Group	Species	Test compou nd	Exposure	Equilibration time	Endpoint	Test duration	Effect concentra tion	Total concent ration	Normalized concentration (1% OC)	Sediment type	Chem. analysis	Note	Validity	References
Insect Chironomidae	Chironomus riparius	CuCl ₂ . 2H ₂ 0	Static renewal	7 d	Growth	28 d	NOEC	505.9	51.6	Artificial OECD sediment: AVS 0.30 mmol/kg, pH 7.2-7.8, OC 9.81%	Measu red	u	R2/C1	Ghent University, 2004 cited ECI 2008
								<u>67.1</u>	<u>32.2</u>					
Insect Chironomidae	Chironomus riparius	CuCl ₂ . 2H ₂ 0	Static renewal	7 d	Survival	28 d	NOEC	589.3	177.0	Artificial OECD sediment: AVS 4.02 mmol/kg, pH 7.2-7.8, OC 3.33%	Measu red	No information chemical analysis during equilibration reported	R2/C3	Ghent University, 2004 cited ECI 2008
Insect Chironomidae	Chironomus riparius	CuCl ₂ . 2H ₂ 0	Static renewal	7 d	Survival	28 d	NOEC	553.6	166.2	Artificial OECD sediment: AVS 16.21 mmol/kg, pH 7.2-7.8, OC 3.33%	Measu red	и	R2/C3	Ghent University, 2004 cited ECI 2008
Insect Chironomidae	Chironomus riparius	CuCl ₂ . 2H ₂ 0	Static renewal	7 d	Emergence	28 d	NOEC	553.6	166.2	Artificial OECD sediment: AVS 16.21 mmol/kg, pH 7.2-7.8, OC 3.33%	Measu red	u	R2/C3	Ghent University, 2004 cited ECI 2008
Insect Chironomidae	Chironomus riparius	CuCl ₂ · 2H ₂ 0	Static renewal	7 d	Growth	28 d	NOEC	553.6	166.2	Artificial OECD sediment: AVS 16.21 mmol/kg, pH 7.2-7.8, OC 3.33%	Measu red	u	R2/C3	Ghent University, 2004 cited ECI 2008
Insect Chironomidae	Chironomus riparius	CuCl ₂ . 2H ₂ 0	Static renewal	7 d	Survival	28 d	NOEC	934.1	95.2	Artificial OECD sediment: AVS 4.05 mmol/kg, pH 7.2-7.8, OC 9.81%	Measu red	u	R2/C3	Ghent University, 2004 cited ECI 2008
Insect Chironomidae	Chironomus riparius	CuCl ₂ · 2H ₂ 0	Static renewal	7 d	Emergence	28 d	NOEC	934.1	95.2	Artificial OECD sediment: AVS 4.05 mmol/kg, pH 7.2-7.8, OC 9.81%	Measu red	u	R2/C3	Ghent University, 2004 cited ECI 2008
Insect Chironomidae	Chironomus riparius	CuCl ₂ · 2H ₂ 0	Static renewal	7 d	Growth	28 d	NOEC	452.6	46.1	Artificial OECD sediment: AVS 4.05 mmol/kg, pH 7.2-7.8, OC 9.81%	Measu red	u	R2/C3	Ghent University, 2004 cited ECI 2008
Insect Chironomidae	Chironomus riparius	CuCl ₂ · 2H ₂ 0	Static renewal	7 d	Survival	28 d	NOEC	1 417	144.4	Artificial OECD sediment: AVS 12.60	Measu red	u	R2/C3	Ghent University,



Group	Species	Test compou nd	Exposure	Equilibration time	Endpoint	Test duration	Effect concentra tion	Total concent ration	Normalized concentration (1% OC)	Sediment type	Chem. analysis	Note	Validity	References
										mmol/kg, pH 7.2-7.8, OC 9.81%				2004 cited ECI 2008
Insect Chironomidae	Chironomus riparius	CuCl ₂ . 2H ₂ 0	Static renewal	7 d	Emergence	28 d	NOEC	1 417	144.4	Artificial OECD sediment: AVS 12.60 mmol/kg, pH 7.2-7.8, OC 9.81%	Measu red	u	R2/C3	Ghent University, 2004 cited ECI 2008
Insect Chironomidae	Chironomus riparius	CuCl ₂ · 2H ₂ 0	Static renewal	7 d	Growth	28 d	NOEC	1 417	144.4	Artificial OECD sediment: AVS 12.60 mmol/kg, pH 7.2-7.8, OC 9.81%	Measu red	u	R2/C3	Ghent University, 2004 cited ECI 2008
Insect Chironomidae	Chironomus riparius	CuCl ₂ . 2H ₂ 0	Static renewal	7 d	Survival	28 d	NOEC	2 113	326.1	Natural OECD sediment: AVS 15.57 mmol/kg, pH 7.2-7.8, OC 6.48%	Measu red	u	R2/C3	Ghent University, 2004 cited ECI 2008
Insect Chironomidae	Chironomus riparius	CuCl ₂ . 2H ₂ 0	Static renewal	7 d	Emergence	28 d	NOEC	1 320	203.7	Natural OECD sediment: AVS 15.57 mmol/kg, pH 7.2-7.8, OC 6.48%	Measu red	u	R2/C3	Ghent University, 2004 cited ECI 2008
Insect Chironomidae	Chironomus riparius	CuCl ₂ . 2H ₂ 0	Static renewal	7 d	Growth	28 d	NOEC	776.5	119.8	Natural OECD sediment: AVS 15.57 mmol/kg, pH 7.2-7.8, OC 6.48%	Measu red	u	R2/C3	Ghent University, 2004 cited ECI 2008
Insect Ephemeridae	Hexagenia sp.	CuCl ₂ . 2H ₂ 0	Static	14 d	Survival	21 d	NOEC	39.2	78.4	Natural sediment : OC 0.5%	Measu red	AVS not measured	R4/C1	Milani et al. 2003 cited ECI 2008
Insect Ephemeridae	Hexagenia sp.	CuCl ₂ . 2H ₂ 0	Static	14 d	Survival	21 d	NOEC	33.9	67.8	Natural sediment : OC 0.5%	Measu red	AVS not measured	R4/C1	Milani et al. 2003 cited ECI 2008
Insect Ephemeridae	Hexagenia sp.	CuCl ₂ . 2H ₂ 0	Static	14 d	Survival	21 d	NOEC	44.9	89.8	Natural sediment : OC 0.5%	Measu red	AVS not measured	R4/C1	Milani et al. 2003 cited ECI 2008
								39.1	78.2					
Insect Ephemeridae	Hexagenia sp.	CuCl ₂ . 2H ₂ 0	Static	14 d	Growth	21 d	NOEC	23.4	46.8	Natural sediment : OC 0.5%	Measu red	AVS not measured	R4/C1	Milani et al. 2003 cited ECI 2008
Insect Ephemeridae	Hexagenia sp.	CuCl ₂ . 2H ₂ 0	Static	14 d	Growth	21 d	NOEC	29.2	58.4	Natural sediment : OC 0.5%	Measu red	AVS not measured	R4/C1	Milani et al. 2003 cited ECI 2008



Group	Species	Test compou nd	Exposure	Equilibration time	Endpoint	Test duration	Effect concentra tion	Total concent ration	Normalized concentration (1% OC)	Sediment type	Chem. analysis	Note	Validity	References
Insect Ephemeridae	Hexagenia sp.	CuCl ₂ . 2H ₂ 0	Static	14 d	Growth	21 d	NOEC	44.9	89.8	Natural sediment : OC 0.5%	Measu red	AVS not measured	R4/C1	Milani et al. 2003 cited ECI 2008
								31.3	62.6					
Oligochaeta	Lumbriculus variegatus	CuCl ₂ . 2H ₂ 0	Static renewal	7 d	Biomass	28 d	NOEC	80.5	30.7	Artificial OECD sediment: AVS 0.05 mmol/kg, pH 7.2-7.8, OC 2.62%	Measu red	No information chemical analysis during equilibration reported	R2/C1	Ghent University, 2004 cited ECI 2008
Oligochaeta	Lumbriculus variegatus	CuCl₂· 2H₂0	Static renewal	8-11 d	Biomass	28 d	NOEC	91.8	46.8	Natural sediment: AVS 0.10 mmol/kg, pH 7.2-7.8, OC 1.96%	Measu red	u	R2/C1	Ghent University, 2004 cited ECI 2008
								<u>86.0</u>	<u>37.9</u>					
Oligochaeta	Lumbriculus variegatus	CuCl ₂ . 2H ₂ 0	Static renewal	8-11 d	Biomass	28 d	NOEC	416.3	59.7	Natural sediment: AVS 16.5 mmol/kg, pH 7.2-7.8, OC 6.97%	Measu red		R2/C1	Ghent University, 2004 cited ECI 2008
Oligochaeta	Tubifex tubifex	CuCl ₂ . 2H ₂ 0	Static renewal	7 d	Survival	28 d	NOEC	138.5	52.9	Artificial OECD sediment: AVS 0.05 mmol/kg, pH 7.2-7.8, OC 2.62%	Measu red	No information chemical analysis during equilibration reported	R2/C1	Ghent University, 2004 cited ECI 2008
Oligochaeta	Tubifex tubifex	CuCl ₂ . 2H ₂ 0	Static renewal	8-11 d	Survival	28 d	NOEC	54.0	19.1	Natural sediment: AVS 0.27 mmol/kg, pH 7.2-7.8, OC 2.83%	Measu red	u	R2/C1	Ghent University, 2004 cited ECI 2008
Oligochaeta	Tubifex tubifex	CuCl ₂ . 2H ₂ 0	Static renewal	8-11 d	Survival	28 d	NOEC	95.3	45.0	Natural sediment: AVS 0.28 mmol/kg, pH 7.2-7.8, OC 2.12%	Measu red	u	R2/C1	Ghent University, 2004 cited ECI 2008
Oligochaeta	Tubifex tubifex	CuCl ₂ . 2H ₂ 0	Static renewal	7 d	Survival	28 d	NOEC	580.9	59.2	Artificial OECD sediment: AVS 0.59 mmol/kg, pH 7.2-7.8, OC 9.81%	Measu red	u	R2/C1	Ghent University, 2004 cited ECI 2008
								<u>89.3</u>	<u>40.5</u>					



Group	Species	Test compou nd	Exposure	Equilibration time	Endpoint	Test duration	Effect concentra tion	Total concent ration	Normalized concentration (1% OC)	Sediment type	Chem. analysis	Note	Validity	References
Oligochaeta	Tubifex tubifex	CuCl ₂ . 2H ₂ 0	Static renewal	7 d	Reproducti on	28 d	NOEC	79.3	30.3	Artificial OECD sediment: AVS 0.05 mmol/kg, pH 7.2-7.8, OC 2.62%	Measu red	No information chemical analysis during equilibration reported	R2/C1	Ghent University, 2004 cited ECI 2008
Oligochaeta	Tubifex tubifex	CuCl ₂ · 2H ₂ 0	Static renewal	8-11 d	Reproducti on	28 d	NOEC	18.3	6.47	Natural sediment: AVS 0.27 mmol/kg, pH 7.2-7.8, OC 2.83%	Measu red	u	R2/C1	Ghent University, 2004 cited ECI 2008
Oligochaeta	Tubifex tubifex	CuCl₂∙ 2H₂0	Static renewal	8-11 d	Reproducti on	28 d	NOEC	56.1	26.5	Natural sediment: AVS 0.28 mmol/kg, pH 7.2-7.8, OC 2.12%	Measu red	"	R2/C1	Ghent University, 2004 cited ECI 2008
Oligochaeta	Tubifex tubifex	CuCl₂∙ 2H₂0	Static renewal	8-11 d	Reproducti on	28 d	NOEC	98.3	50.2	Natural sediment: AVS 0.10 mmol/kg, pH 7.2-7.8, OC 1.96%	Measu red	"	R2/C1	Ghent University, 2004 cited ECI 2008
Oligochaeta	Tubifex tubifex	CuCl ₂ . 2H ₂ 0	Static renewal	7 d	Reproducti on	28 d	NOEC	580.9	59.2	Artificial OECD sediment: AVS 0.59 mmol/kg, pH 7.2-7.8, OC 9.81%	Measu red	u	R2/C1	Ghent University, 2004 cited ECI 2008
								<u>53.2</u>	<u>27.4</u>					
Oligochaeta	Tubifex tubifex	CuCl ₂ . 2H ₂ 0	Static renewal	8-11 d	Growth	28 d	NOEC	18.3	6.47	Natural sediment: AVS 0.27 mmol/kg, pH 7.2-7.8, OC 2.83%	Measu red	No information chemical analysis during equilibration reported	R2/C1	Ghent University, 2004 cited ECI 2008
Oligochaeta	Tubifex tubifex	CuCl ₂ . 2H ₂ 0	Static renewal	7 d	Growth	28 d	NOEC	79.3	30.3	Artificial OECD sediment: AVS 0.05 mmol/kg, pH 7.2-7.8, OC 2.62%	Measu red	u	R2/C1	Ghent University, 2004 cited ECI 2008
Oligochaeta	Tubifex tubifex	CuCl ₂ . 2H ₂ 0	Static renewal	8-11 d	Growth	28 d	NOEC	32.2	15.2	Natural sediment: AVS 0.28 mmol/kg, pH 7.2-7.8, OC 2.12%	Measu red	u	R2/C1	Ghent University, 2004 cited ECI 2008



Group	Species	Test compou nd	Exposure	Equilibration time	Endpoint	Test duration	Effect concentra tion	Total concent ration	Normalized concentration (1% OC)	Sediment type	Chem. analysis	Note	Validity	References
Oligochaeta	Tubifex tubifex	CuCl ₂ · 2H ₂ 0	Static renewal	8-11 d	Growth	28 d	NOEC	53.0	27.0	Natural sediment: AVS 0.10 mmol/kg, pH 7.2-7.8, OC 1.96%	Measu red	"	R2/C1	Ghent University, 2004 cited ECI 2008
								<u>39.7</u>	<u>21.7</u>					
Oligochaeta	Tubifex tubifex	CuCl ₂ . 2H ₂ 0	Static renewal	7 d	Survival	28 d	NOEC	988.3	296.8	Artificial OECD sediment: AVS 8.04 mmol/kg, pH 7.2-7.8, OC 3.33%	Measu red	No information chemical analysis during equilibration reported	R2/C1	Ghent University, 2004 cited ECI 2008
Oligochaeta	Tubifex tubifex	CuCl ₂ . 2H ₂ 0	Static renewal	7 d	Reproducti on	28 d	NOEC	459.2	137.9	Artificial OECD sediment: AVS 8.04 mmol/kg, pH 7.2-7.8, OC 3.33%	Measu red	u	R2/C1	Ghent University, 2004 cited ECI 2008
Oligochaeta	Tubifex tubifex	CuCl ₂ . 2H ₂ 0	Static renewal	7 d	Growth	28 d	NOEC	163	48.9	Artificial OECD sediment: AVS 8.04 mmol/kg, pH 7.2-7.8, OC 3.33%	Measu red	u	R2/C1	Ghent University, 2004 cited ECI 2008
Oligochaeta	Tubifex tubifex	CuCl ₂ . 2H ₂ 0	Static renewal	7 d	Survival	28 d	NOEC	937	281.4	Artificial OECD sediment: AVS 14.39 mmol/kg, pH 7.2-7.8, OC 3.33%	Measu red	u	R2/C3	Ghent University, 2004 cited ECI 2008
Oligochaeta	Tubifex tubifex	CuCl ₂ · 2H ₂ 0	Static renewal	7 d	Survival	28 d	NOEC	1 267	121.2	Artificial OECD sediment: AVS 5.43 mmol/kg, pH 7.2-7.8, OC 9.81%	Measu red	u	R2/C3	Ghent University, 2004 cited ECI 2008
Oligochaeta	Tubifex tubifex	CuCl ₂ · 2H ₂ 0	Static renewal	7 d	Reproducti on	28 d	NOEC	1 037	105.7	Artificial OECD sediment: AVS 5.43 mmol/kg, pH 7.2-7.8, OC 9.81%	Measu red	u	R2/C3	Ghent University, 2004 cited ECI 2008
Oligochaeta	Tubifex tubifex	CuCl ₂ · 2H ₂ 0	Static renewal	7 d	Growth	28 d	NOEC	1 037	105.7	Artificial OECD sediment: AVS 5.43 mmol/kg, pH 7.2-7.8, OC 9.81%	Measu red	u	R2/C3	Ghent University, 2004 cited ECI 2008
Oligochaeta	Tubifex tubifex	CuCl ₂ . 2H ₂ 0	Static renewal	7 d	Survival	28 d	NOEC	1 357	138.3	Artificial s OECD ediment: AVS 15.15	Measu red	u	R2/C3	Ghent University, 2004 cited ECI 2008



Group	Species	Test compou nd	Exposure	Equilibration time	Endpoint	Test duration	Effect concentra tion	Total concent ration	Normalized concentration (1% OC)	Sediment type	Chem. analysis	Note	Validity	References
										mmol/kg, pH 7.2-7.8, OC 9.81%				
Oligochaeta	Tubifex tubifex	CuCl₂∙ 2H₂0	Static renewal	7 d	Reproducti on	28 d	NOEC	480.9	49.0	Artificial OECD sediment: AVS 15.15 mmol/kg, pH 7.2-7.8, OC 9.81%	Measu red	u	R2/C3	Ghent University, 2004 cited ECI 2008
Oligochaeta	Tubifex tubifex	CuCl ₂ · 2H ₂ 0	Static renewal	7 d	Growth	28 d	NOEC	271.6	27.7	Artificial OECD sediment: AVS 15.15 mmol/kg, pH 7.2-7.8, OC 9.66%	Measu red	u	R2/C3	Ghent University, 2004 cited ECI 2008
Oligochaeta	Tubifex tubifex	CuCl ₂ · 2H ₂ 0	Static renewal	8-11 d	Reproducti on	28 d	NOEC	1 856	74.9	Natural sediment: AVS 56.4 mmol/kg, pH 7.2-7.8, OC 24.8%	Measu red	u	R2/C3	Ghent University, 2004 cited ECI 2008
Oligochaeta	Tubifex tubifex	CuCl ₂ · 2H ₂ 0	Static renewal	8-11 d	Growth	28 d	NOEC	1 856	74.9	Natural sediment: AVS 56.4 mmol/kg, pH 7.2-7.8, OC 24.8%	Measu red	u	R2/C3	Ghent University, 2004 cited ECI 2008
Oligochaeta	Tubifex tubifex	CuSO4	Static	14 d	Survival	28 d	NOEC	67.25	47.7	Artificial sediment: <63µm 69.7%, OC 1.41%	Measu red	No information on chemical analysis of overlying water during equilibration and test	R3/C1	Vecchi et al. 1999 cited in ECI 2008
Oligochaeta	Tubifex tubifex	CuSO4	Static	14 d	Reproducti on	28 d	NOEC	67.25	47.7	Artificial sediment: <63μm 69.7%, OC 1.41%	Measu red	u	R3/C1	Vecchi et al. 1999 cited in ECI 2008
Oligochaeta	Tubifex tubifex	CuSO4	Static	14 d	Survival	28 d	NOEC	385.8	247.3	Natural sediment: <63µm 65.2%, OC 1.56%	Measu red	u	R3/C1	Vecchi et al. 1999 cited in ECI 2008
Oligochaeta	Tubifex tubifex	CuSO4	Static	14 d	Reproducti on	28 d	NOEC	231.7	148.5	Natural sediment: <63µm 65.2%, OC 1.56%	Measu red	u	R3/C1	Vecchi et al. 1999 cited in ECI 2008
Oligochaeta	Tubifex tubifex	CuSO4	Static	14 d	Survival	28 d	NOEC	101.4	98.4	Artificial sediment: <63µm 62.5%, OC 1.03%	Measu red	u	R3/C1	Vecchi et al. 1999 cited in ECI 2008



Group	Species	Test compou nd	Exposure	Equilibration time	Endpoint	Test duration	Effect concentra tion	Total concent ration	Normalized concentration (1% OC)	Sediment type	Chem. analysis	Note	Validity	References
Oligochaeta	Tubifex tubifex	CuSO4	Static	14 d	Reproducti on	28 d	NOEC	62.64	60.8	Artificial sediment: <63μm 62.5%, OC 1.03%	Measu red	u	R3/C1	Vecchi et al. 1999 cited in ECI 2008
Oligochaeta	Tubifex tubifex	CuSO4	Static	14 d	Survival	28 d	NOEC	69.1	67.1	Artificial sediment: <63μm 62.5%, OC 1.03%	Measu red	Without food addition	R3/C1	Vecchi et al. 1999 cited in ECI 2008
Oligochaeta	Tubifex tubifex	CuCl ₂ . 2H ₂ 0	Static	14 d	Survival	28 d	NOEC	237.8	475.6	Natural sediment : OC 0.5%	Measu red	AVS not measured	R4/C1	Milani et al. 2003 cited ECI 2008
Oligochaeta	Tubifex tubifex	CuCl ₂ . 2H ₂ 0	Static	14 d	Survival	28 d	NOEC	246.9	493.8	Natural sediment : OC 0.5%	Measu red	AVS not measured	R4/C1	Milani et al. 2003 cited ECI 2008
Oligochaeta	Tubifex tubifex	CuCl ₂ . 2H ₂ 0	Static	14 d	Survival	28 d	NOEC	270.5	541	Natural sediment : OC 0.5%	Measu red	AVS not measured	R4/C1	Milani et al. 2003 cited ECI 2008
Oligochaeta	Tubifex tubifex	CuCl ₂ . 2H ₂ 0	Static	14 d	Growth	28 d	NOEC	127.8	255.6	Natural sediment : OC 0.5%	Measu red	AVS not measured	R4/C1	Milani et al. 2003 cited ECI 2008
Oligochaeta	Tubifex tubifex	CuCl ₂ . 2H ₂ 0	Static	14 d	Growth	28 d	NOEC	129	258	Natural sediment : OC 0.5%	Measu red	AVS not measured	R4/C1	Milani et al. 2003 cited ECI 2008
Oligochaeta	Tubifex tubifex	CuCl ₂ . 2H ₂ 0	Static	14 d	Growth	28 d	NOEC	270.5	541	Natural sediment : OC 0.5%	Measu red	AVS not measured	R4/C1	Milani et al. 2003 cited ECI 2008
Mollusca	Potamopyrg us antipodarus	CuCl ₂ . 2H ₂ 0	Static renewal	n.r.	Growth	28 d	NOEC	74.7	57.5	Natural sediment: OC 1.3%	Measu red	AVS not measured, OC estimated as OM/1.7. Time-average sediment concentration , overlying water concentration measured at test end; no information on aging and equilibration	R3/C1	Pang et al. 2013



Appendix 2. Goodness-of-fit of toxicity data from ETX SSD results

With no OC normalization:

Anderson-Darling test for normality											
Sign. level	Critical	Normal?									
0.1	0.631	Accepted									
0.05	0.752	Accepted		AD Statistic:	0.289859						
0.025	0.873	Accepted		n:	6						
0.01	1.035	Accepted									
Kolmogorov-Smirnov test for normality											
Sign. level	Critical	Normal?									
0.1	0.819	Accepted									
0.05	0.895	Accepted		KS Statistic:	0.529036						
0.025	0.995	Accepted		n:	6						
0.01	1.035	Accepted									
Cramer von Mis	es test for n	ormality									
Sign. level	Critical	Normal?									
0.1	0.104	Accepted									
0.05	0.126	Accepted		CM Statistic:	0.033343						
0.025	0.148	Accepted		n:	6						
0.01	0.179	Accepted									



With effect data normalized to 1% OC:

Anderson-Darling test for normality											
Sign. level	Critical	Normal?									
0.1	0.631	Accepted									
0.05	0.752	Accepted	AD Statistic:	0.558228							
0.025	0.873	Accepted	n:	6							
0.01	1.035	Accepted									
Kolmogorov-Smirnov test for normality											
Sign. level	Critical	Normal?									
0.1	0.819	Accepted									
0.05	0.895	Accepted	KS Statistic:	0.740282							
0.025	0.995	Accepted	n:	6							
0.01	1.035	Accepted									
Cramer von Mis	es test for n	ormality	· · ·								
Sign. level	Critical	Normal?									
0.1	0.104	Accepted									
0.05	0.126	Accepted	CM Statistic:	0.071473							
0.025	0.148	Accepted	n:	6							
0.01	0.179	Accepted									