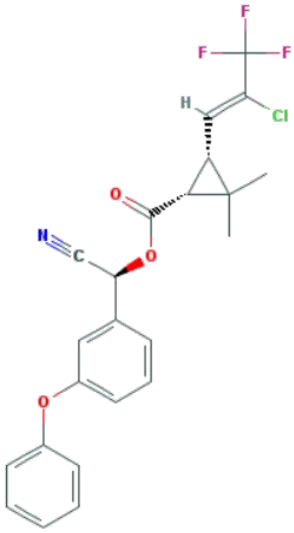
Miljø- og
Fødevareministeriet
Miljøstyrelsen

Fastsættelse af kvalitetskriterier for vandmiljøet

Gamma-Cyhalothrin
γ-Cyhalothrin

CAS nr. 76703-62-3



Vandkvalitetskriterium	$VKK_{ferskvand}$	0,22 ng/l
Vandkvalitetskriterium	$VKK_{saltvand}$	0,022 ng/l
Korttidsvandkvalitetskriterium	$KVKK_{ferskvand}$	0,22 ng/l
Korttidsvandkvalitetskriterium	$KVKK_{saltvand}$	0,022 ng/l
Sedimentkvalitetskriterium	$SKK_{ferskvand}$	28 ng/kg OC tørvægt
Sedimentkvalitetskriterium	$SKK_{saltvand}$	28 ng/kg OC tørvægt
Biota-kvalitetskriterie, sekundær forgiftning	$BKK_{sek.forgiftn.}$	15,9 mg/kg lipid vådvægt
Biota-kvalitetskriterie, sundhed	$BKK_{sundhed}$	0,6 mg/kg lipid vådvægt

Indhold

Forord

Et kvalitetskriterium for vandmiljøet er det højeste koncentrationsniveau, ved hvilket der skønnes ikke at forekomme uacceptable negative effekter på vandøkosystemer.

Miljøstyrelsen (MST) udarbejder kvalitetskriterier for kemikalier i vandsøjlen (vandkvalitetskriterium), i sediment og i dyr og planter (biota).

Miljøstyrelsen bruger kvalitetskriterierne som det faglige grundlag til at kunne fastsætte miljøkvalitetskrav, hvorved der forstås den endelige koncentration af et bestemt forurenende stof i vand, sediment eller biota, som ikke må overskrides af hensyn til beskyttelsen af miljøet og menneskers sundhed.

Metodikken, der anvendes til udarbejdelse af miljøkvalitetskriterier, er harmoniseret i EU og baserer sig på vandrammedirektivet (EU 2000), EU's revideret vejledning til fastsættelse af kvalitetskriterier i vandmiljøet også kendt som Technical Guidance Document No. 27 eller TGD#27 (EU 2018) og Miljøstyrelsens vejledning til fastsættelse af vandkvalitetskriterier (Miljøstyrelsen 2004). Metodikken er endvidere i overensstemmelse med EU's vejledning til risikovurdering under REACH forordningen (EU 2008).

Den sidste litteratursøgning er foretaget den 1.12.2018.

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English Summary and conclusions

The insecticide gamma-cyhalothrin is one of four isomers originating from cyhalothrin, a pyrethroid type plant protection product. Gamma cyhalothrin is considered the most toxic of the four isomers. As an insecticide, gamma-cyhalothrin is very toxic to invertebrates, but also relatively toxic to e.g. fish. The availability of ecotoxicological data is relatively high. It has therefore been possible to establish annual averages based environmental quality standards (AA-EQS) for all matrices, including the maximum acceptable concentration (MAC) in surface water.

Gamma-cyhalothrin has a log K_{ow} of 4.96 and is moderately persistent in soil with DT90, ranging from 86 to more than 100 days. The chemical-physical properties result in a relatively high bioconcentration in fish, with an estimated average BCF of 4982 based upon studies with lambda-cyhalothrin.

The following annual averages EQS have been derived for gamma cyhalothrin:

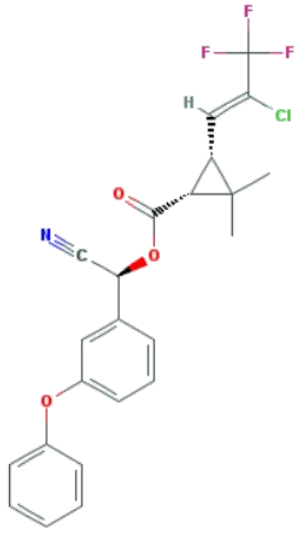
$AA\text{-}EQS_{freshwater}$ =	0.22 ng L^{-1}
$AA\text{-}EQS_{saltwater}$ =	0.022 ng L^{-1}
$MAC_{freshwater}$ =	0.22 ng L^{-1}
$MAC_{saltwater}$ =	0.022 ng L^{-1}
$EQS_{sediment}$ =	28.0 ng kg^{-1} organic carbon (dry weight)
EQS_{biota} =	15.9 mg kg^{-1} lipid (wet weight)
$EQS_{human\ helath}$ =	0.6 mg kg^{-1} lipid (wet weight)

1 Indledning

Identiteten af gamma-cyhalothrin fremgår af tabel 1.

Cyhalothrin er et insekticid af pyrethroid-typen bestående af tre chirale centre og fire isomerer, herunder gamma-cyhalothrin (GHT) i en ligelig fordeling på ca. 20-25% (EU 2012). Gamma-cyhalothrin i den rene version består næsten udelukkende (98%) af 1R,cis,Z-S' isomeren og regnes for den økotoksikologiske mest potente af de fire isomerer. Beslægtede stoffer er lambda-cyhalothrin, der indeholder mere end 40-50% gamma-cyhalothrin foruden samme mængde af 1S,cis,Z-R' isomeren. Isomeren 1R,cis,Z-S' (gamma-cyhalothrin) er estimeret til at være minimum 162 gange mere giftigt for zebra fisk end den anden isomer, der udgør lambda-cyhalothrin (1S,cis,Z-R') (Xu m.fl. 2008). Kendte cyhalothrin-baserede pesticider, som er baseret alene på gamma-cyhalothrin, inkluderer f.eks. GF-317 (Corello) produceret af Cheminova.

Tabel 1.1. Identitet og struktur af gamma-cyhalothrin (EU 2012)

IUPAC navn	[(S)-cyano-(3-phenoxyphenyl)methyl] (1*R*,3*R*)-3-[(*Z*)-2-chloro-3,3,3-trifluoroprop-1-enyl]-2,2-dimethylcyclopropane-1-carboxylate
Strukturformel	
CAS nr.	76703-62-3
EINECS nr.	616-373-3
Kemisk formel	C23H19ClF3NO3
SMILES	CC1(C(C1C(=O)OC(C#N)C2=CC(=CC=C2)OC3=CC=CC=C3)C=C(C(F)(F)F)Cl)C

2 Fysisk kemiske egenskaber

De fysisk kemiske egenskaber for gamma-cyhalothrin fremgår af tabel 2.

Tabel 2.1. Fysisk kemiske egenskaber for gamma-cyhalothrin

Parameter	Værdi	Reference
Molekylevægt, Mw (g·mol-1)	449,85	EU 2012
Smeltepunkt, Tm (°C)	55,6	EU 2012
Kogepunkt, Tb (°C)	#1	EU 2012
Damptryk, Pv (Pa) (20 °C)	3,45 x 10-7	EU 2012
Henry's konstant, H (pa·m3·mol-1)	0,0221	EU 2012
Vandopløselighed, Sw (mg·L-1)	0,002	EU 2012
Dissociationskonstant, pKa	NA	
Octanol/vand fordelingskoefficient, log Kow	4,96	EU 2012
Koc (L·kg-1)	151200	DK QSAR data base

[#1]Nedbrydes inden kogepunktet
NA = not assessed

3 Skæbne i miljøet

3.1 Nedbrydelighed

Gamma-cyhalothrin absorberer qua sin lave vandopløselighed relativt stærkt til organisk materiale og derved også potentielt til jord, hvilket medfører, at stoffet ikke kan karakteriseres som let nedbrydeligt.

I fire jordtyper blev der fundet Kd værdier (fordelingskoefficienter mellem jordvæske og jordpartikler) ved hjælp af OECD 106 metoden på i gennemsnit 622 (spænd på 610-908), hvilket indikerer en moderat adsorption (EU 2012). Alle jorder havde dog relativt lavt indhold af organisk materiale (0,57-1,97 % med et gennemsnit på 1,1%).

Nedbrydningstider - målt som den primære nedbrydning - i tre jorder er rapporteret som DT50 mellem 23 og 33 dage med et geometrisk middel på 25 dage efter konversion til 20 °C (EU 2012). I sediment blev der i tre forskellige typer (organisk kulstof 0,6-2,7 og 7,4 %) målt DT50 på 11, 17 og 53 dage. De tilknyttede DT90 var >100, >100 og 86 dage, hvilket indikerer en moderat persistens af GHT i sediment.

3.2 Bioakkumulering

Der er ikke fundet eksperimentelle data for bioakkumulering af gamma-cyhalothrin, dog findes der enkelte bioakkumuleringsstudier med lambda-cyhalothrin, der kan give et indblik i potentialet for akkumulering i biota (EU 2012). BCF for fisk blev i et ældre studie med en række mangler bestemt til 2240, mens der i et 300 dages livscyklusstudie blev målt BCF i F0 generation af tykhovedet elritse (*P. promelas*) mellem 3952-6691 kg/L ved de forskellige testkoncentrationer. Gennemsnitlig BCF var 4982. Disse BCF var højere end dem, der blev målt i æg og larver fra F1 generationen. Alt i alt opfylder lambda-cyhalothrin således kriterierne for at være karakteriseret som ”bioakkumulerende”, men ikke ”meget bioakkumulerende” under REACH forordningen, hvor grænsen går ved henholdsvis 2000 og 5000 kg/L. Det formodes, at BCF for gamma-cyhalothrin vil være sammenlignelig med den for lambda-cyhalothrin, hvorfor den gennemsnitlige BCF på 4982 kg/L for lambda-cyhalothrin også anvendes for gamma-cyhalothrin.

3.3 Naturlig forekomst

Cyhalothrin er et såkaldt pyrethroid. Pyrethroider er baseret på naturlige insektgifte, som dannes i bestemte arter af Krysanthemum (f.eks. *Tanacetum cinerariifolium* eller *Tanacetum coccineum*), der ikke er naturligt hjemmehørende i Danmark.

4 Giftighedsdata

4.1 Giftighed over for vandlevende organismer

Der foreligger en del økotoksikologiske ferskvandsdata for cyhalothriner, de fleste dækker dog produkter med lambda-cyhalothrin, hvor isomeren gamma-cyhalothrin kun udgør en andel på 30-50% og resten typisk inaktive isomerer. Dette underbygges af et større sammenlignende studie, som finder, at gamma-cyhalothrin er omtrent dobbelt så giftigt som lambda-cyhalothrin (Gidding m.fl. 2008). Schroer m.fl (2004) undersøgte giftigheden af lambda-cyhalothrin i en lang række laboratoriestudier med vandlevende invertebrater (krebsdyr, insekter, snegle og fladorme), der må formodes at være den mest følsomme organismegruppe over for insekticider som cyhalothriner. Dyr blev indsamlet enten fra midlertidige populationer holdt i laboratorier eller direkte fra felten. I begge tilfælde blev de akklimatiseret i minimum 2 dage inden forsøgsstart. Eksponering til lambda-cyhalothrin skete gennem en kommerciel formulering (Karate) og koncentrationer verificeret gennem kemiske analyser. Generelt var der et markant fald i vandkoncentrationerne gennem de 96 timer, som forsøgene typisk varede. For 13 forskellige akvatiske insektarter var EC50 mellem 2,8 og 957 ng/L med dansemyggelarven *Chaoborus obscuripes* som den mest følsomme. *Daphnia magna* blev ikke testet af Schroer m.fl., hvorfor en direkte sammenligning mellem gamma- og lambda-cyhalothrin er vanskelig, men EC50(48 h) for *Dapnia galeata* blev rapporteret til 117 ng/L.

For det rene stof gamma-cyhalothrin er de fleste data fra standard-lignende tests desuden baseret på korttidsstudier. Derudover er mange NOEC værdier fra ECOTOX databasen f.eks. baseret på et semi-felt studie (Wijngaarden m.fl. 2009). Dette studie diskuteres nærmere i afsnit 4.2

Tabel 4.1 lister de brugbare NOEC/EC10 fra langtidsstudier med fisk og invertebrater samt autotrofe organismer (vandplanten andemad). Derudover foreligger der i EU (2012) et studie (OECD 201) med algen *Selenastrum capricornutum,* hvor der blev observeret en lav giftighed af gamma-cyhalothrin, hvilket understreger den forventede lave giftighed af stoffet for autotrofe organismer. I en dosis-serie fra 0,156-5,0 mg/L var der således en ikke-signifikant hæmning på antal alger på mellem 22,2 og 34%, mens der ingen dosis-respons sammenhæng kunne ses (EU 2012). I et tilsvarende forsøg med en gamma-cyhalothrin formulering (GF-317) kunne der etableres en NOEC på 4,0 mg aktivt stof per L. Altså væsentligt højere værdier end for fisk og invertebrater.

Effektkoncentrationer over for vandlevende organismer er sammenstillet i tabel 4.1 (NOEC/EC10) og tabel 4.2 (EC50/LC50).

Tabel 4.1. NOEC og EC10 værdier (ng/L) fra langtidsstudier anvendt i fastsættelsen af vandkvalitetskriterium

Arter / Species	**NOEC/ EC10**	**Eksponeringstid**	**Effektmål / End-point**	**Kilde [CRED]@**
Fathead minnow (Pimephales promelas) (Early life stage)	37,9	35 d	Larvae survival / NOEC	1 [1]
Waterflea (Daphnia magna)	2,18	21 d	Reproduction / NOEC	1 [1]
Midge (Chironomus riparius)*	46,9	28 d	Development / NOEC	1 [1]
Duck weed (Lemna gibba)	508	7 d	Growth / NOEC	2 [1]

* Forsøg udført som spiket vand med tilstedeværelsen af sediment

@ Data er kvalitetsvurderet efter CRED metoden (Moermond m.fl. 2016), hvor koderne "1" og "2" dækker over henholdsvis pålidelig uden restriktioner og pålidelig med restriktioner. Se Bilag A for mere information.

Kilder: 1= EU 2012; 2= OPP. Se Bilag A.

4.2 Giftighed over for sedimentlevende organismer

Som en del af validering af vandkvalitetskriterier baseret på laboratoriedata kan disse bl.a. sammenlignes med effekter observeret i felten under mere realistiske forhold. I et hollandsk studie med udendørs beholdere blev ændringer i samfundsstrukturen af makro-invertebrater, zooplankton, fytoplankton og makrofytter fulgt over en periode på cirka 70 dage (van Wijngaarden m.fl. 2009). Organismerne var primært indsamlede arter undtagen to arter (*Gammarus pulex* og *Asellidae*), der blev introduceret ved forsøgsstart. I alt 71 makro-invertebrater (primært insekter) indgik i den endelige dataanalyse. Konklusionen på dette studie var, at der - med en enkelt midlertidig undtagelse – ikke kunne ses effekter på de forskellige populationer og den overordnede samfundsstruktur ved koncentrationer under 5 ng/L. Glasmyg, *Chaoborus sp*, var den mest følsomme art, og her var effekter ved 5 ng/L undervejs i studiet, f.eks. en forbigående 25% reduktion på dag 17. Ved 10 ng/L var effekterne markante (50%) og længerevarende. Alt i alt blev en NOEC for samfundet af invertebrater på 5 ng/L anbefalet.

En opsummering af engelske og amerikanske feltstudier af Maund m.fl. (1998) for lambda-cyhalothrin konkluderede, at NOEC var at finde ved omtrent 20 ng/L. I det amerikanske mesokosmos studie (Hill m.fl. 1994) blev artssammensætningen af invertebrater og fisk i store damme fulgt efter adskillige simulerede pesticidsprøjtninger. NOEC for samfundet af invertebrater blev estimeret til 17 ng/L baseret på nominelle startkoncentrationer. For fisk var den højere. I det engelske mesokosmos-studie (Farmer m.fl.1995) blev en række 25 m^3 store kunstige damme fire gange med to ugers mellemrum tilført lambda-cyhalotrin i mængder, der svarede til 17 eller 170 ng/L. Ved den laveste dosis var der kun få effekter i gruppen af insekter og isopoder, mens at der ved slutning af forsøget stadig var målbare effekter på gruppen *Gammaridae* (tanglopper).

Tabel 4.2. EC50/LC50 værdier (ng/L) fra korttidsstudier anvendt i fastsættelsen af korttids-vandkvalitetskriterium

Arter / Species	EC50/ LC50	Eksponerings-tid	Effektmål / Endpoint	Kilde [CRED]@
Fish, Rainbow trout (Oncorhynchus mykiss)	72,1	96 h	Mortality	1 [1]
Fish, Rainbow trout (Oncorhynchus mykiss)	170	96 h	Mortality	1 [1]
Fish, Blugill sunfish (Lepomis macrochirus)	35,4	96 h	Mortality	1 [1]
Fish, Blugill sunfish (Lepomis macrochirus)	63,1	96 h	Mortality	1 [1]
Fish, Fathead minnow (Pimephales promelas)	340	96 h	Mortality	1 [1]
Fish, Zebra fish (Brachydanio rerio)	270	96 h	Mortality	1 [1]
Fish, Guppy (Poecilia reticulata)	170	96 h	Mortality	1 [1]
Crustacea (Daphnia magna)	99,4	48 h	Immobility	1 [1]
Crustacea (Daphnia magna)	45	48 h	Immobility	1 [1]
Crustacea (Gammarus pseudolimnaeus)	3,05	96 h	Immobility	1 [1]
Crustacea (G. pseudolimnaeus) - Neonates	0,446	96 h	Immobility	1 [1]
Crustacea (G. pseudolimnaeus) - Juveniles	3,58	96 h	Immobility	1 [1]
Crustacea (G. pseudolimnaeus) - Adults	6,32	96 h	Immobility	1 [1]
Crustacea (Gammarus pulex)	9,2	96 h	Immobility	2 [2]
Crustacea (Asellus aquaticus)	23,7	96 h	Immobility	2 [2]
Crustacea (Prosellus coxalis)	16,6	96 h	Immobility	2 [2]
Insecta (Chaoborus obscurripes)	3,8	96 h	Immobility	2 [2]
Insecta (Cloeon dipterum)	23,4	96 h	Immobility	2 [2]
Insecta (Notonecta maculata)	4,6	96 h	Immobility	2 [2]
Insecta (Corisa punctata)	12,3	96 h	Immobility	2 [2]
Insecta (Coenagrionidae sp.)	322	96 h	Immobility	2 [2]
Insecta (Chinonomini sp.)	145	96 h	Immobility	2 [2]
Algae (Selenastrum capricornutum)*	>2,85	96 h	Growth (Biomass)	1 [1]
Algae (Selenastrum capricornutum)*	>2,85	96 h	Growth (Growth rate)	1 [1]
Algae (Selenastrum capricornutum)	6,99	96 h	Growth (Cell density)	1 [1]

@ Data er kvalitetsvurderet efter CRED metoden (Moermond m.fl. 2016), hvor koderne ”1” og ”2” dækker over henholdsvis pålidelig uden restriktioner og pålidelig med restriktioner. Se Bilag A for mere information.

* Disse ”>” data bruges ikke i evalueringen af hvilken sikkerhedsfaktor, der skal bruges i KVKK (se afsnit 5.2)

Kilder: 1= EU 2012; 2= Wijngaarden m.fl. (2009). Se Bilag A.

4.3 Giftighed over for sedimentlevende organismer

Der findes meget få økotoksikologiske studier med sedimentlevende organismer eksponeret for gamma-cyhalothrin. Således er kun ét brugbart studium fundet (EU 2012). Dette er gennemført efter forskrifter i OECD 219 og under GLP. *Chironomus riparius* er i 28 dage eksponeret til en koncentrationsserie på 0,0469-1,5 µg/kg. De subletale målparametre var fremkomst (emergence) og udvikling (development), med det sidste som den mest følsomme parameter. NOEC blev bestemt til 46,9 ng/kg i et sediment med et rapporteret kulstofindhold på 1,7%.

4.4 Giftighed over for pattedyr og fugle

Gamma-cyhalothrin er relativt ufarligt for fugle. Et studie fra EU (2012) fandt således LD50 for *Colinus virginianus* (Nothern Bobwhite Quail) til at være > 2000 mg/kg kropsvægt. Et fodringsstudie med samme art fandt en LC50 på 2644 mg/kg føde. Begge studier var udført efter gældende OECD standarder og fulgte GLP. Giftigheden for pattedyr er beskrevet i afsnit 4.5.

4.5 Giftighed over for mennesker

Gamma-cyhalothrin er giftig ved indtagelse (fareklassificering H301, H312 og H330).

Der er fastsat en ADI (acceptable daily intake) for gamma-cyhalothrin på 0,0025 mg/kg kropsvægt/dag. Denne er baseret på en NOAEL (No Observed Adverse Effect Level) på 0,5 mg/kg kropsvægt/dag og en sikkerhedsfaktor på 200 (EU 2012). NOAEL stammer fra et 12 måneders eksponeringsstudie med hund og er udført med lambda-cyhalothrin, hvorfor sikkerhedsfaktoren er dobbelt så høj som normalt for at kompensere for den højere giftighed af gamma-cyhalothrin.

5 Andre effekter

Foruden ovennævnte fareklassificering kan det nævnes, at Gamma-cyhalothrin kan forårsage hudallergi (H317) og organskader ved længerevarende eller gentagen eksponering (H373).

6 Udledning af vandkvalitetskriterier

6.1 Vandkvalitetskriterier (VKK)

Samlet set har det ikke været muligt at beregne et vandkvalitetskriterium (VKK) baseret på arternes følsomhedsfordeling (Species Sensitivity Distribution, SSD, metoden), eftersom der var data fra færre end 10 arter fra 8 forskellige taksonomiske grupper.

Baseret på fire NOEC/EC10 fra langtidsstudier (tabel 4.1) er der fastsat et vandkvalitetskriterium for ferskvand baseret på kroniske data fra fisk, invertebrater og en vandplante. VKK fastsættes på baggrund af en sikkerhedsfaktor på 10 og den laveste NOEC/EC10, hvilket er NOEC for *Daphnia magna* på 2,18 ng/L.

$VKK_{(ferskvand)}$ = 2,18 ng/L/10 = 0,22 ng/L

De tilgængelige økotoksikologiske data fra subletale langtidsstuder (tabel 4.1) indeholder ikke studier med saltvandsorganismer. Vandkvalitetskravet for saltvand findes derfor iflg. EU's retningslinjer (EU TGD#27) ved brug af en højere sikkerhedsfaktor på de samme ferskvandsdata.

Baseret på fire NOEC/EC10 fra langtidsstudier i ferskvand (tabel 4.1) er der fastsat et vandkvalitetskriterium for saltvand baseret på kroniske data fra fisk, invertebrater og en vandplante. VKK fastsættes på baggrund af en sikkerhedsfaktor på 100 og den laveste NOEC/EC10, hvilket er NOEC for *Daphnia magna* på 2,18 ng/L.

$VKK_{(saltvand)}$ = 2,18 ng/L/100 = 0,022 ng/L

6.2 Korttidsvandkvalitetskriterier (KVKK)

Baseret på EC50/LC50 fra korttidsstudier (tabel 4.2) kan der fastsættes et korttidskvalitetskriterium (KVKK). Da der foreligger akutte EC50/LC50 data fra fisk, invertebrater og alger, kan KVKK i udgangspunktet fastsættes på baggrund af en sikkerhedsfaktor på 100 og den laveste EC50/LC50. Det er dog muligt at reducere sikkerhedsfaktoren til 10, såfremt det undersøgte stof har en specifik virkningsmekanisme, og der foreligger et antal relevante studier med den mest følsomme målgruppe inkluderet i datasættet. For insekticidet gamma-cyhalothrin vil dette være insekter eller leddyr generelt. Her foreligger studier med 11 arter, hvilket er vurderet til at være tilstrækkeligt. En anden mulighed for at reducere sikkerhedsfaktoren er, hvis variationen i det fulde datasæt er begrænset, dvs. at standardafvigelsen i de log-transformerede data er mindre end 0,5, hvilket imidlertid ikke er tilfældet for gamma-cyhalothrin, hvor standardafvigelsen er 0,77. Alt i alt vurderes det dog, at sikkerhedsfaktoren kan reduceres til 10. KVKK kan derfor fastsættes på baggrund af en sikkerhedsfaktor på 10 og den laveste EC50, hvilket er en EC50 på 0,446 ng/L for nyfødte *Gammarus pseudolimnaeus*. KVKK baseret på korttidseksponering kan derfor beregnes til 0,446 ng/L/10 = 0,045 ng/L, hvilket rundt regnet er 50 gange lavere end det estimeret VKK på 0,22 ng/L baseret på langtidseksponering. Økotoksikologisk anses dette ikke for at være meningsfuldt og skyldes formodet, at

Gammarus som art er mere følsomme end *Daphnia*. I overensstemmelse med EU's retningslinjer i TGD#27 fastsættes KVKK i sådanne tilfælde på niveau med VKK.

$KVKK_{(ferskvand)}$ = $VKK_{(ferskvand)}$ = 0,22 ng/L

Samlet set har det ikke været muligt at beregne et korttidsvandkvalitetskriterium (KVKK) baseret på SSD metoden, eftersom de 17 arter i tabel 4.2 dækker mindre end de anbefalede 8 forskellige taksonomiske grupper. F.eks. mangler der data fra diverse ormerækker (ledorme, fladorme, rundorme m.fl.), bløddyr og højere planter. En fuldt normalfordelt SSD med 17 arter dækkende fisk, krebsdyr, insekter og alger resulterer i en HC5 på 1,70 [0,39-4,48] ng/L. I SSD er anvendt det geometriske gennemsnit, hvis der er flere sammenlignelige data for én art (se tabel 4.2). For *Gammarus pseudolimnaeus* er den laveste EC50 dog anvendt, eftersom data stammer fra forskellige livsstadier. Den maksimale anbefalede sikkerhedsfaktor i forbindelse med SSD er ifølge TGD#27 på 5, dvs. PNEC=HC5/5. Anvendes denne sikkerhedsfaktor pga. de relativt få taksonomiske grupper, vil det resultere i en PNEC på 0,34 ng/L, hvilket er på niveau med det fastsatte KVKK på 0,22 ng/L.

Der findes ikke studier med saltvandsorganismer, uagtet at enkelte af testorganismerne kan findes i brakvand. KVKK for saltvand fastsættes derfor på baggrund af KVKK for ferskvand med en yderligere sikkerhedsfaktor på 10. Også for saltvand fastsættes KVKK på niveau med VKK i overensstemmelse med EU's retningslinjer i TGD#27.

$KVKK_{(saltvand)}$ = $VKK_{(saltvand)}$ = 0,022 ng/L

6.3 Kvalitetskriterium for sediment (SKK)

Generelt anses stoffer med en adsorptionskoefficient til organisk karbon (Koc) lavere end1000 L/kg ikke for at ophobes i sediment. SKK skal derfor iflg. TGD#27 kun beregnes for stoffer med en log Koc eller log Kow ≥3. Med en log Kow på 4,96 (tabel 2.1) er det derfor relevant at beregne SKK for gamma-cyhalothrin.

Ifølge TGD#27 fastsættes PNEC for sediment, når der kun findes én NOEC med en sikkerhedsfaktor på 100, efter at NOEC er kalibreret til et standard sediment med 5% organisk kulstof.

Den fundne NOEC på 46,9 ng/kg (1,7 % OC) konverteres til en NOEC i standard sediment (5%OC) på 137,9 ng/kg, hvilket med en sikkerhedsfaktor på 100 resulterer i et sedimentkvalitetskriterium på 1,4 ng/kg tv. Dette kvalitetskriterium gælder for både ferskvands- og marine sedimenter.

SKK = 137,9 ng/kg/100 = 1,4 ng/kg tv (5% OC)

SKK = 28 ng/kg OC

6.4 Kvalitetskriterium for biota (BKK)

For stoffer med en Kow > 3 bør der fastsættes et kvalitetskriterium, der beskytter mod sekundær forgiftning af fugle, pattedyr og andre top-predatorer gennem ophobning i fødekæderne.

Som beskrevet i afsnit 4.4 er ADI for gamma-cyhalthrin baseret på en NOAEL (No Observed Adverse Effect Level) på 0,5 mg/kg kropsvægt/dag i et studie med hund over 12 måneder. NOAEL kan omregnes til NOEC for føde ved at gange med faktoren BW/DFI, hvor BW er kropsvægt og DFI er det daglige fødeindtag. For hundeforsøg er denne konversionsfaktor (BW/DFI) sat til 40 ifølge TGD#27. Det vil sige NOEC for føde er 20 mg/kg.

BKK beregnes ud fra NOEC i fodringsforsøg, hvor $NOEC_{foder}$ angiver den højeste koncentration i foderet, hvor der ingen statistisk signifikant forskel var mellem eksponerede og ueksponerede (kontrol) dyr. NOEC bestemmes ud fra toksikologiske dosis-respons data og en sikkerhedsfaktor, som beror på typen af underliggende data. Når NOEC stammer fra et kronisk toksikologisk forsøg med et pattedyr (i dette tilfælde hund), er sikkerhedsfaktoren ifølge TGD#27 på en faktor 10.

PNEC skal ifølge TGD#27 energinormaliseres for at kalibrere energiindholdet i føden, som er brugt i de bagvedliggende toksikologiske undersøgelser (her hundefoder) med energiindholdet i føden for de organismer, der ønskes beskyttet (her rovfisk fra øverste trofisk niveau).

NOEC = 20 mg/kg foder

$PNEC_{foder}$ = NOEC/10 = 2 mg/kg foder

TGD#27 angiver desuden, at:

Energiindhold i (hunde)foder = 15.100 kJ kg^{-1} tørvægt

Tørstofindhold i (hunde)foder = 92 %

Derved kan PNEC udtrykt i energienhed beregnes som

$$PNEC_{foder} = \frac{2{,}0\ mg/kg}{15100\ KJ/kg * 0{,}92} = 0{,}000144\ mg/kJ$$

TGD#27 angiver samtidig at:

Energiindhold i fisk = 21.000 kJ kg^{-1} tørvægt

Tørstofindhold i fisk = 26,3 %

Energiindholdet i frisk fisk kan følgelig beregnes som: 21.000 kJ kg^{-1} tørvægt * 0,263 = 5523 kJ kg^{-1} fisk, vådvægt.

PNEC udtrykt som indhold i frisk fisk, $PNEC_{fisk,\ vådvægt}$ er derved:

$PNEC_{fisk}$ = 0,000144 mg kJ^{-1} * 5523 kJ kg^{-1} fisk,vådvægt = 0,8 mg kg^{-1} fisk,vådvægt.

Med et standard fedtindhold (lipid) i fisk på 5% (vådvægt) kan en lipidnormaliseret $PNEC_{fisk}$ beregnes som

$$PNEC_{fisk,lipid} = \frac{0{,}8\frac{mg}{kg\ fisk}}{0{,}05\ \frac{kg\ lipd}{kg\ fisk}} = 15{,}9\ \frac{mg}{kg\ lipid}$$

BKK = 0,8 mg/kg fisk

BKK = 15,9 mg/kg lipid

6.5 Kvalitetskriterium for human konsum af vandlevende organismer (HKK)

Da gamma-cyhalothrin er klassificeret H302, giftigt ved indtagelse, skal der beregnes et kvalitetskriterium for human konsum af vandlevende organismer, HKK.

Der er fastsat et maksimalt acceptabelt dagligt indtag for mennesker (ADI - Acceptable Daily Intake) for gamma-cyhalothrin på 0,0025 mg/kg kropsvægt/dag.

Ifølge TGD#27 beregnes HKK ud fra en forudsætning om, at maksimalt 20% af ADI må stamme fra fisk og skalddyr, og at et standard fødeindtag fra denne kilde svarer til 0,00163 kg fisk (vv)/kg kropsvægt/dag.

Herved kan HKK beregnes som:

$$\text{HKK} = \frac{0{,}2 * \text{ADI}}{0{,}00163} = \frac{0{,}2 * 0{,}0025}{0{,}00163} = 0{,}03 \text{ mg/kg fisk (vv)}$$

Med et standard fedtindhold (lipid) i fisk på 5% kan en lipidnormaliseret $PNEC_{fisk}$ beregnes som

$$\text{PNEC}_{\text{fisk,lipid}} = \frac{0{,}03 \frac{mg}{kg\ fisk}}{0{,}05 \frac{kg\ lipd}{kg\ fisk}} = 0{,}6 \frac{mg}{kg\ lipid}$$

HKK = 0,03 mg/kg fisk vv

HKK = 0,6 mg/kg lipid

6.6 Vandkvalitetskriterier baseret på BKK og HKK

Ifølge TGD#27 bør de fundne kvalitetskriterier for sekundær forgiftning af biota (BKK) og mennesker (HKK) konverteres til vandkvalitetskriterier for at sikre, at de fundne VKK baseret på direkte effekter er tilstrækkelige konservative til at beskytte mod sekundære effekter gennem bioakkumulering i fødekæder.

VKK_{biota} og $VKK_{human\ konsum}$ beregnes som BKK/BAF og HKK/BAF, hvor BAF er bioakkumuleringsfaktoren. Kendes denne ikke, kan den beregnes som $BCFxBFM_{1\text{-}2}$, hvor BCF er biokoncentrationsfaktoren og BMF_1 er den (tredje) trofiske biomagnifikationsfaktor dækkende fødekæden fra alge til rovfisk, og BMF_2 er biomagnifikationsfaktoren fra rovfisk til toppredatorer som rovfugle og pattedyr. Det sidste led (BMF_2) for toppredatorer anvendes typisk kun for det marine og ikke det ferske vandmiljø. BMF1 og BMF2 kan bestemmes eksperimentelt, men i mangel af stofspecifikke værdier kan generiske værdier fra TGD#27 anvendes. Da biomagnifikationspotentialet afhænger af stoffets fysisk-kemiske egenskaber, er de generiske værdier fastsat i forhold til stoffets log Kow og/eller målte BCF. For gamma-cyhalothrin, med en log Kow på 4,96, er BMF_1 og BMF_2 fastsat til 2 i begge tilfælde. For stoffer med en log Kow > 5 er de to BMF fastsat til

10. BCF i fisk er angivet som 4982 (afsnit 3.2), hvilket også lige akkurat medfører en placering i gruppen af stoffer (2000 ≤ BCF< 5000), hvor BMF er angivet som 2 og ikke 10.

Da HKK er mindre end BKK beregnes VKK på basis af HKK som følger:

Ferskvand:

$VKK_{fw,human\ konsum}$ = HKK/(BCF x BMF_1) = 0,03 mg/kg/(4982 L/kg x 2) = 3,0 x 10^{-6} mg/L = 3,0 ng/L

Saltvand:

$VKK_{sw,human\ konsum}$ = HKK/(BCF x BMF_1 x BMF_2) = 0,03 mg/kg/(4982 L/kg x 2 x 2) = 1,5 x 10^{-6} mg/L = 1,5 ng/L

I begge tilfælde er de ovenstående VKK, tilbageberegnet fra BKK og HKK, højere end de VKK, der er beregnet på baggrund af akvatiske økotoksikologiske data (afsnit 5.1 og 5.2), hvorfor disse fastholdes som de endelige VKK for fersk- og saltvand.

7 Konklusion

Cyhalothrin er et insekticid af pyrethroid-typen, hvor stoffet i sin struktur består af tre chirale centre og fire isomerer, herunder gamma-cyhalothrin i en ligelig fordeling på ca. 20-25%. Gamma-cyhalothrin i den rene version består næsten udelukkende (98%) af 1R,cis,Z-S' isomeren og regnes for den økotoksikologiske mest potente af de fire isomerer. Gamma-cyhalothrin er som insekticid stærkt giftigt overfor invertebrater, men også relativt giftigt over for fisk. Mængden af pålidelige og relevante økotoksikologiske data er generelt høj, og det har været muligt at fastsætte kvalitetskriterier for alle miljømatricer.

Følgende kvalitetskriterier er fastsat:

Vandkvalitetskriterium, ferskvand (VKK) = 0,22 ng/L

Vandkvalitetskriterium, saltvand (VKK) = 0,022 ng/L

Korttidsvandkvalitetskriterium, ferskvand (KVKK) = 0,22 ng/L

Korttidsvandkvalitetskriterium, saltvand (KVKK) = 0,022 ng/L

Sedimentkvalitetskriterium (SKK) = 28 ng/kg OC dw

Kvalitetskriterium for biota (BKK) = 15,9 mg/kg lipid ww

Sundhedskvalitetskriterium (HKK) = 0,6 mg/kg lipid ww

8 Referencer

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9 Bilag 1. Kvalitetsevaluering af data

Evaluering af studier og data fundet i EU's risikovurderingsrapport for gamma-cyhalothrin forelagt for Kommissionen og forfattet af UK i 2008 og opdateret i 2012.

Species	**Comments from the Reporting Member State (RMS) (UK) on Study Quality. From EU (2012)**
Rainbow Trout (*O. mykiss*)	The study is GLP compliant and was conducted in line with current EPA/OECD guidelines. No significant deviations from the guidelines were noted. The results are therefore considered suitable for use in the regulatory risk assessment.
Bluegill sunfish (*L. macrochirus*)	The study is GLP compliant and was conducted in line with current EPA/OECD guidelines. The RMS notes that the mortality level recorded in the water control group 20%, greater than that normally accepted for a valid study. However, since there were no mortality recorded in the vehicle control, which is considered more representative of the test substance concentrations dissolved in solvent, the RMS considers this study suitable for use in the regulatory risk assessment.
Zebra fish (*B. rerio*)	The study is GLP compliant and was conducted in line with current OECD guideline. No significant deviations from the guideline were noted. The results are therefore considered suitable for use in the regulatory risk assessment.
Fathead minnow (P. promelas) (Early life stage)	The study is GLP compliant and was conducted in line with current guideline. No significant deviations from the guideline were noted. The results are therefore considered suitable for use in the regulatory risk assessment
Guppy (*P. reticulata*)	The study is GLP compliant and was conducted in line with current OECD guideline. No significant deviations from the guideline were noted. The results are therefore considered suitable for use in the regulatory risk assessment.
Waterflea (D. magna)	The study is GLP compliant and was conducted in line with current guideline. No significant deviations from the guideline were noted. The results are therefore considered suitable for use in the regulatory risk assessment
Amphipod *(G.pseudolimnaeus - neonates)*	The study was adopted from the US EPA FIFRA Standard Experimental Procedure 540/9-85-006 and was conducted to GLP compliant. No significant deviations from the guidelines or study plan were noted. The results are therefore considered suitable for use in the regulatory risk assessment.
Midge (*C. riparius*)*	The study is GLP compliant and was conducted in line with current guideline. No significant deviations from the guideline were noted. The results are therefore considered suitable for use in the regulatory risk assessment.

* Sediment study

OPP Pesticide Ecotoxicity Database

I USA har det regionale Centre for Integrated Pest Mangement (IPM) under United States Department of Agriculture samlet en database med økotoksikologiske data, der har været indsendt, gennemgået og kvalitetsmæssigt anerkendt i forbindelse med de amerikanske godkendelsesprocedure (Se Boks 1).

I fastsættelsen af vandkvalitetskriteriet for gamma-cyhalothrin er der anvendt data fra OPP databasen (tabel 2.1). Disse er præsenteret i Figur 1.

Kvalitetsevalueringen i OPP opererer med forskellige kvalitetskategorier[1]. De relevante i denne sammenhæng er således:

"The three study categories used by the Agency to classify studies are core, supplemental, and invalid are represented by a letter code as C, S, or IN".

Eller mere specifikt:

Core: *All essential information was reported and the study was performed according to recommended EPA or ASTM methodology. Minor inconsistencies with standard recommended procedures may be apparent; however, the deviations do not detract from the study's soundness or intent. Studies within this category fulfil the basic requirements of current FIFRA guidelines and are acceptable for use in a risk assessment.*

Supplemental*: Studies in this category are scientifically sound; however, they were performed under conditions that deviated substantially from recommended protocols. Results do not meet guideline requirements; however, the information may be useful in a risk assessment. Some examples of the conditions that may place a study in a supplemental category include:*

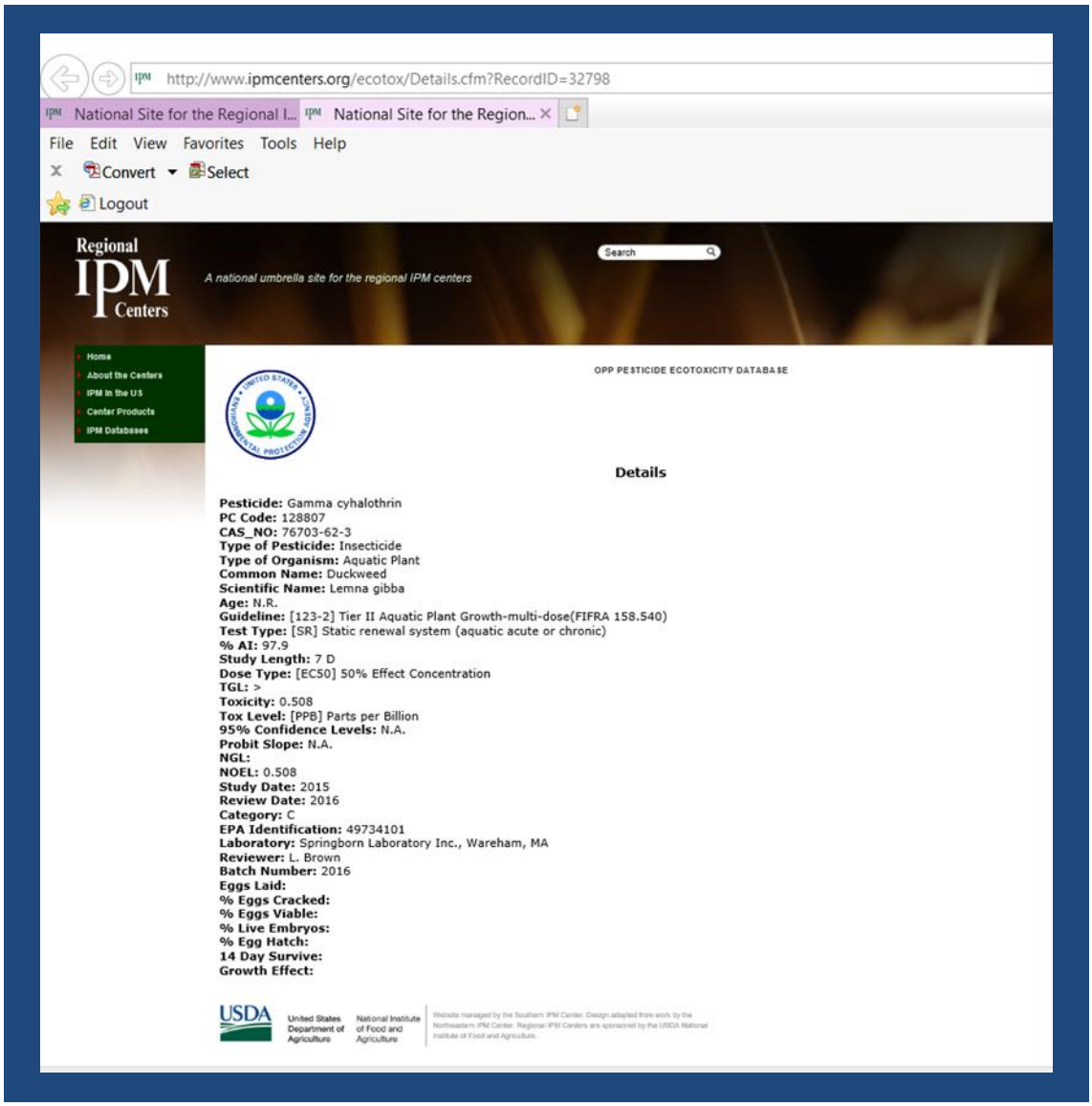
- *Unacceptable or non-native test species*
- *Test material not properly identified*
- *Dosage levels tested were less than 5000 ppm (or 100 ppm for aquatics), but not high enough to produce an effect on the tested organisms or a precise LC50/EC50(exceptions sometimes made for highly insoluble chemicals).*
- *Deviations from recommended diet preparation measures*
- *Deviations from recommended water quality characteristics which may have stressed test organisms and affected toxicological response(e.g., low D.O. in aquatic studies)*
- *Tested organisms were older or younger than required age.*

Som det kan ses på de efterfølgende sider, er studiet med andemad (*Lemna gibba*) karakteriseret under kategori "C", hvilket tolkes som fuldt acceptabelt.

[1] http://www.ipmcenters.org/ecotox/DatabaseGuidance.pdf

Tekst fra OPP Pesticide Ecotoxicity Database. http://www.ipmcenters.org/ecotox/

The Ecological Fate and Effects Division of the USEPA Office of Pesticide Programs is continuing efforts to update the database with all EPA reviewed ecotoxicity endpoints for pesticides registered or previously registered in the U.S. Toxicity data on over 4,000 active ingredients, metabolites, and multi- ingredient formulations are presently included in the database. The toxicity data inputed into the database is compiled from actual studies reviewed by EPA in conjunction with pesticide registration or reregistration and studies performed by USEPA, USDA and USFWS laboratories, which have been reviewed by Agency biologists and judged acceptable for use in the ecological risk assessment process. The database presently contains over 30,000 records for acute and chronic toxicity endpoints on terrestrial and aquatic plants, aquatic invertebrates, terrestrial invertebrates, insects, amphibians, fish, birds, reptiles, and wild mammals. The database is presented in Microsoft ACCESS and contains 35 fields per record entry. Each record entry summarizes one ecotoxicity study for a single species or one toxicity endpoint from a multiple species study and includes EPA tracking information regarding that study submission.



FIGUR 1. Screen dump af informationer fra databasen med økotoksikologisk data fra den Amerikanske Miljøstyrelse (USEPA) – Office of Pesticide Program (OPP).

EVALUATION SCHEME FOR CRED ASSESSMENTS

Remark: If a study includes data on several tests / test species / test endpoints, relevance and reliability of these endpoints may differ. In this case, separate sheets should be completed for each endpoint.

Evaluated study (full reference):	van Wijngaarden, R.P.A., Barber, I. & Brock, T.C.M. 2009. Effects of the pyrethroid insecticide gamma-cyhalothrin on aquatic invertebrates in laboratory and outdoor microcosm tests. Ecotoxicology 18: 211-224. https://doi.org/10.1007/s10646-008-0274-1
Test substance:	Gamma-cyhalotrin (CAS:76703-62-3)
Evaluated test:	Short term exposure of fresh water species to Gamma-cyhalotrin
Evaluated test species:	*Chaoborus obscuripes* *Chironomini Sp.* *Cloeon dipterum* *Notonecta maculata* *Corixa punctata* *Coenagrionidae sp.* *Gammarus pulex* *Asellus aquaticus* *Proasellus coxalis*
Evaluated test endpoint(s):	Immobility / Mortality

Evaluator (institution):	John Jensen, Department of Bioscience, Aarhus University

For each question, mark one appropriate answer with x. NR = Not Reported. NA = Not Applicable.

A. Relevance of the data

Remark: Relevance of a study mainly depends on the scope of the assessment / the regulatory framework, for which the study is evaluated. The following 12 questions should therefore be answered in the context of the overall assessment. NR= Not Reported. NA= Not Assessed.

	Yes	No	NR	NA
1. Is the tested species relevant for the compartment under evaluation?	**X**			
Example: An aquatic species should be tested to evaluate risks for the aquatic environment.				
Remarks:				

	Yes	No	NR	NA
2. Are the tested organisms relevant for the tested compound?	**X**			
Example: In case of an ERA for an antibiotic, cyanobacteria should be used as test species instead of algae.				
Remarks:				

	Yes	No	NR	NA
3. Are the reported endpoints appropriate for the regulatory purpose?	**X**			
Example: Acute effects on aquatic organisms are not relevant for the environmental risk assessment of human pharmaceuticals				
Remarks:				

	Yes	No	NR	NA
4. Are the reported endpoints appropriate for the investigated effects or the mode of action of the test substance?	**X**			
Explanation: When a risk assessment is performed for a substance, for which information is available on a specific mode of action that is considered relevant for environmental organisms, studies including endpoints assessing this particular mode of action are most appropriate. For instance, if an API is known to affect reproduction of vertebrates, the endpoints of the fish early life stage test may not be appropriate. Instead, fish tests should include endpoints such as vitellgenin levels, secondary sex characteristics, sex ratio and reproduction depending on the specific mode of action of the substance (OECD 2012).				
Remarks:				

	Yes	No	NR	NA
5. Is the effect relevant on a population level?	**X**			
Explanation: Endpoints of the guideline studies, on which the ERA of human pharmaceuticals is based, are generally population relevant. For non-standard tests, population relevance has to be evaluated on a case by case basis.				
Remarks:				

	Yes	No	NR	NA
6. Is the recorded effect statistically significant, biologically relevant and appropriate for the regulatory purpose?	**X**			
Explanation: In the context of environmental risk assessment, a biologically relevant effect is an effect that is important and meaningful for environmental health (EFSA 2011). In a test system with relatively little control variation, minor changes may be statistically significant without necessarily being biologically relevant. To evaluate risks caused by chronic exposure, NOEC or EC_{10} values are used, while EC_{50} values are not appropriate. For the EC_{10}, it has to be evaluated on a case-by-case basis, if the effect is within biological variation of the control response. To				

evaluate risks caused by acute exposure (note that this is only relevant for some terrestrial tests with human pharmaceuticals), EC_{50} values are preferred.
Remarks:

	Yes	No	NR	NA
7. Are appropriate life-stages studied?	**X**			
Explanation/example: The tested life stage should be (a) appropriate for the selected test and test design and (b) relevant for the expected effect of the API. For instance, fish early life stages are not appropriate for studying effects on reproduction.				
Remarks:				

	Yes	No	NR	NA
8. Are the test conditions appropriate for the tested species and relevant for the assessment?	**X**			
Explanation/example: Test organisms should be tested under appropriate conditions. For instance, freshwater species should be tested in freshwater and saltwater species in saltwater. If a test with freshwater or saltwater species is required depends on the scope of the assessment.				
Remarks:				

	Yes	No	NR	NA
9. Is the timing and duration of exposure relevant and appropriate for the studied endpoints and species?	**X**			

Explanation: The required exposure time should be appropriate for the test organism and the studied endpoint. Chronic studies should include sensitive life stages or cover the whole life cycle.
Remarks: Duration was 96 h

	Yes	No	NR	NA
10. If recovery is studied, is this relevant for the framework for which the study is evaluated?				**X**
Explanation: In most regulatory frameworks (including the environmental risk assessment of human pharmaceuticals), recovery is not relevant (exception: authorization of plant protection products).				
Remarks:				

	Yes	No	NR	NA
11. Is the substance tested representative and relevant for the substance being assessed?	**X**			
Explanation: Sufficient information should be provided to allow a clear identification of the test item. A substance may be tested as pure active substance or in a formulation. Tests performed with formulations are relevant for plant protection products, but less relevant within many other regulatory frameworks. Studies with mixtures of different substances are relevant for assessing toxicity of these mixtures, but not for assessing the individual substances contained in the mixture. For salts, the counter ion may influence toxicity. For pro-drugs, the active moiety and, if entering the environment in >10% of the administered does, the pro-drug need to be assessed (EMA/CHMP 2011). Depending on the regulatory framework, effects of transformation products may need to be considered. If the substance causing the effect is not the substance being assessed, expert judgement is needed to decide on how to deal with the results of the study and the resulting risk assessment.				
Remarks:				

	Yes	No	NR	NA
12. Is the tested exposure route relevant for the assessment?	**X**			
Explanation/example: The exposure route should be appropriate for the assessment. For instance, exposure by injection is generally not appropriate (Harris et al. 2014). For pharmaceuticals, exposure should be continuous. Intermittent exposure is generally not relevant. Exposure duration has to be sufficiently long. However, note that acute tests with some terrestrial organisms are also required in the environmental risk assessment of human pharmaceuticals.				
Remarks:				

Assigned Relevance Class: R1

Justification: All essential relevance criteria fulfilled

B. Reliability of the data

For each question, mark one appropriate answer with x. NR= Not Reported. NA= Not Assessed.

Remark: Before evaluating the test, please check the physico-chemical characteristics of the test substance (what is the solubility, log K_{OW}, pK_a, is the compound volatile, does it hydrolyse, photolyse etc.?)

1. Is a standard method (e.g. OECD, ISO, US EPA) or modified standard used? Please specify:

	Yes	No	NR	NA
1a. A standard method is used.				
1b. A slightly modified standard method is used.	**X**			
1c. A substantially modified standard method is used.				
Remarks 1b: *The protocol partly resembles OECD 202 for Daphnia magna*				

	Yes	No	NR	NA
2. Is the test, including chemical analysis of the test substance where required, performed under GLP conditions?		**X**		
Remarks:				

3. Validity criteria:

	Yes	No	NR	NA
3a. Are all validity criteria fulfilled if applicable?	**X**			
Explanation: For standard tests, compliance with the validity criteria of the guideline is crucial for a study to be considered as reliable. Please check the corresponding test guideline where relevant. For non-guideline tests with standard species, validity criteria as described in a guideline for a similar test should be met if applicable.				

	Yes	No	NR	NA
3b. Are validity criteria clearly failed?		**X**		
Explanation: If validity criteria are clearly failed, a test is classified as '3' (not reliable).				
Remarks:				

4. Inclusion of appropriate control replicates:

	Yes	No	NR	NA
4a. Was a negative control included, and was its performance acceptable?	**X**			
4b. Was a positive control included, if required, and was its performance acceptable?			**X**	
4c. Was a solvent control included, if a solvent was used, and was its performance acceptable?	**X**			
Explanation: It depends on the test substance and test type which controls should be included; please check the corresponding test guideline where relevant. In addition to the negative control, a solvent control has to be included in all cases where a solvent is used. The concentration of solvent in the solvent control should correspond to the highest solvent concentration used in the test treatments. In some tests, a positive control with a reference substance is required. For standard tests, the corresponding guidelines provide information on how the controls should perform, e.g. with regard to survival, growth or reproduction. For non-standard tests and non-standard test organisms, expert judgement is needed to decide if performance of the controls is acceptable. Performance of the solvent control should preferably not differ significantly from performance of the negative control.				
Remarks:				

Test Substance

	Yes	No	NR	NA
5. Is the test substance clearly identified with name, CAS-number or SMILES code and, where relevant, information on stereochemistry?	**X**			
Explanation/example: If the salt of an API was tested, information on the type of salt should be provided. It should be specified if test concentrations relate to free acid / free base or salt. If the test substance is not clearly identified, a test is classified as '3' (not reliable).				
Remarks:				

	Yes	No	NR	NA
6a. Is the purity of the test substance reported and in an acceptable range (>95%)?			**X**	
6b. Is the source of the test substance reported and trustworthy?			**X**	
Remarks:				

7. If a formulation is used or if impurities are present:

	Yes	No	NR	NA
7a. Can it be excluded that other ingredients in the formulation or impurities exert an effect?				**X**
7b. Is the amount of test substance in the formulation indicated?				**X**

Remarks:

Test organism

8. Description of the test organisms:

	Yes	No	NR	NA
8a. Is the test species clearly identified?	**X**			
8b. For algae: is mean cell density at the test start within an appropriate range? For other test organisms: Is mean body weight/length of the test organism in an appropriate range?	**X**			
8c. Is age/life stage of the organisms at test start reported and in the required range, where appropriate (e.g. not for algae)?	**X**			
8d. Is sex of the test organisms reported and is sex ratio appropriate, where relevant (e.g. when evaluating sexual-endocrine effects)?				**X**
8e. Is the species strain reported where required?	**X**			
Explanation for 8a: If the test species is not clearly identified, a test is classified as '3' (not reliable). *Explanation for 8 b-e: For standard tests, the corresponding guidelines provide information on required range of mean cell densities, age / life stage of the test organisms etc. at the test start.*				
Remarks 8c: No algae included				

	Yes	No	NR	NA
9a. Are the test organisms from a reliable source? For field collected organisms: is the site of origin well-described?	**X**			

9b. Have the organisms been acclimatized to test conditions (e.g. water type, temperature) before the start of exposure, where relevant? For tests with embryonic stages: have the parental organisms been held at appropriate conditions?	**X**			
9c. Are the test organisms exempt from previous exposure or any other kind of stressor?				**X**
Remarks 9b: All species were acclimated to laboratory conditions for at least 2 days, during which suitable food material was provided				

Test conditions and chemical analysis

10. Appropriateness of the experimental system for the test substance:

	Yes	**No**	**NR**	**NA**
10a. Is the type of exposure (e.g. static, semi-static, flow-through) appropriate for the test substance, taking its physico-chemical characteristics into account?	**X**			
10b. In case that the test substance is a difficult substance as defined in OECD (2000): is the selected test system appropriate for testing of this substance?	**X**			
10c. For ionisable substances: has the test been performed in an appropriate pH-range?				**X**
Explanation 10a: Static systems are in most cases only appropriate for short-term tests (exception: water/sediment tests). Where appropriate, guideline requirements should be followed. *Explanation 10b: Difficult test substances are substances, which are e.g. poorly water soluble, volatile, photo-degradable, hydrolytically unstable, oxidizable, biodegradable, complexing or strongly adsorbing to surfaces of test vessels etc. In order to obtain reliable test results with such substances, test systems generally have to be adapted to take the difficult properties of the substance into account (e.g. by using a closed test*				

system without headspace for volatile substances). For further details, please see OECD (2000). It has to be verified on a case-by-case basis, if the used test system is appropriate for the test substance. *Explanation 10c: Relatively small changes in pH can significantly alter the balance between dissociated and non-dissociated forms of some substances. An altered dissociation equilibrium may significantly affect the water solubility and the partition coefficient of the substance and hence, its bioavailability and toxicity. Tests with such substances should therefore be performed at a pH, within the pH range required for maintaining the health of the test organisms, at which the more toxic form of the test substance prevails (as far as possible). For further guidance see OECD (2000).*
Remarks:

	Yes	No	NR	NA
11. Is the experimental system appropriate for the test organism (e.g. choice of medium / test water or soil, feeding, water or soil characteristics, temperature, light/dark conditions, pH, oxygen content)? Have conditions been stable during the test?	**X**			
Explanation: The general requirements of the test species should be considered with regard to the characteristics of the selected test medium etc. Temperature, pH and oxygen content should be stable and within the appropriate range for the organism (where applicable, check the corresponding guideline). If control performance is not good (e.g. high mortality), this may indicate that test conditions were not appropriate. Where applicable, feeding should follow the guideline requirements, and all excess should be removed after feeding to avoid decreased bioavailability of the test substance.				
Remarks:				

	Yes	No	NR	NA
12a. For aquatic tests: were exposure concentrations below the limit of water solubility?	**X**			

12b. For aquatic tests: if a solvent was used, was solvent concentration within the appropriate range (i.e. not higher than 0.01%)?				**X**
Remarks:				

	Yes	**No**	**NR**	**NA**
13. Is a correct spacing between exposure concentrations applied?	**X**			
Explanation: For standard tests, the corresponding guidelines provide information on the spacing factor. A factor of 3.2 is often recommended. As rule of thumb, the spacing factor should not be >10.				
Remarks:				

	Yes	**No**	**NR**	**NA**
14. Is the exposure duration defined and appropriate?	**X**			
Remarks:				

15. Chemical analysis

	Yes	**No**	**NR**	**NA**
15a. Are chemical analyses performed to verify test substance concentrations over the duration of the study where required?	**X**			
15b. Is an appropriate analytical method used to measure test substance concentrations?	**X**			

15c. Are the measured test substance concentrations within the calibration range of the analytical method?	**X**			
15d. Are samples analyzed from a sufficient number of treatments and controls, and from a sufficient number of time intervals?	**X**			
15e. Are test substance concentrations sufficiently stable during the course of the exposure?		**X**		
Explanation 15a: If required in the corresponding test guideline, nominal test substance concentrations should be verified by chemical analysis. Non-guideline test should be evaluated based on test guidelines for similar tests where appropriate. *Explanation 15d: The frequency of chemical analyses should be evaluated based on the requirements of the corresponding test guideline or, for non-guideline studies, on a guideline for a similar test if appropriate.* *Explanation15e: Please evaluate according to the requirements of the corresponding test guideline or, for non-guideline studies, a test guideline for a similar test where appropriate.*				
Remarks 15e: After 96 hour the dose was reduced to 16% of the initial in average				

	Yes	**No**	**NR**	**NA**
16. Is the biomass loading of the organisms in the test system within an appropriate range?	**X**			
Explanation: For standard tests, the corresponding guidelines provide information on maximum biomass loading. For non-standard tests / non-standard test species, expert knowledge is required to decide if the loading rate is appropriate.				
Remarks:				

Statistical design

	Yes	No	NR	NA
17a. Is a sufficient number of replicates used for all controls and treatments?	**X**			
17b. Is a sufficient number of organisms per replicate used for all controls and test concentrations?	**X**			
Explanation for 17 ab: For standard tests, the guideline requirements should be followed. When a non-guideline study is evaluated, expert judgement is needed to assess if the study design is appropriate to obtain statistically reliable results.				
Remarks:				

	Yes	No	NR	NA
18. Are appropriate statistical methods used to derive the effect concentrations?	**X**			
Explanation: Generally, a description of the statistical methods is needed to assess the reliability of the test results. For standard tests, the corresponding guideline requirements should be followed. Further guidance is e.g. provided by OECD (2006). When a non-guideline study is evaluated, expert judgment may be needed. EC_x values should not be extrapolated considerably beyond the range of tested concentrations.				
Remarks:				

	Yes	No	NR	NA
19a. Is a concentration-response curve observed?	**X**			

19b. Is the observed effect statistically significant?				**X**
Explanation 19a: The requirement for a concentration-response relationship depends on the objective of the study. If a limit test is performed at one (or two) concentration(s) to verify the lack of toxicity and no toxicity is recorded, a concentration-response relationship is obviously not needed to conclude that the LC_{50} or NOEC is above the highest tested concentration. However, if the intention of the study is to demonstrate an effect, reliability of the test results is higher, if (1) a sufficient number of concentrations have been tested and (2) the observed effect is regularly increasing (or regularly decreasing) with increasing test concentration (i.e. the concentration-response relationship is monotonous). Expert knowledge is needed, if an effect is only observed at the highest tested concentration. Expert knowledge is also needed in the case of non-monotonous concentration-response curves (e.g. U-, J- or inverted U-shaped curves). In such cases, the underlying mechanisms of effects and the reproducibility of the results should be considered (Harris et al. 2014). *Explanation 19b: The significance level and the statistical method used to evaluate the specific effect should be indicated.*				
Remarks 19b: The lower and upper confidence limit around ECx differed by less than a factor of 2.				

	Yes	**No**	**NR**	**NA**
20. Are sufficient data available to check the calculation of endpoints and (if applicable) fulfilment of the validity criteria (e.g. control data, concentration-response curves)?			**X**	
Explanation: If enough data are presented, additional endpoints may be calculated by the assessor if not reported by the author of the study.				
Remarks: Raw data is not available.				

Assigned Reliability Class: R2

Justification: Overall, the study is trustworthy and reliable. However, the study is not using a standard test system, it is not conducted under GLP and not all information is presented regarding e.g. origin and purity of chemical substance. Finally, no raw data and dose-response curves was presented.

Overall CRED Score: R2

Justification: Based upon reliability score of R2.