中国科学: 地球科学 2012 年 第 42 卷 第 5 期: 673 ~ 679
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《中国科学》杂志社
SCIENCE CHINA PRESS

论 文

基于基因表达效应的物种敏感度分析初探

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收稿日期: 2012-02-28; 接受日期: 2012-04-20
国家水体污染控制与治理科技重大专项(编号: 2012ZX07501-003-006)和中国环境科学研究院改革启动经费专项项目(编号: 2011GQ-02)

摘要 物种敏感度分析是水质基准推算的主要技术之一, 现阶段应用于物种敏感度分析的主要是生物个体水平的毒性数据. 随着基因组学数据的日渐丰富, 基因表达效应数据能否应用于水质基准的推算值得研究. 本研究以重金属镉、铜和锌为例, 广泛搜集了相关生物毒性数据, 通过构建物种敏感度分布曲线对三种重金属的毒性效应进行了分析. 结果表明, 镉的急性、慢性和基因表达效应数据的物种敏感度排序为慢性>基因>急性, 铜和锌的基因表达效应数据不足, 但从趋势上看, 锌的三种类型数据的物种敏感度与镉类似, 铜与二者不同, 其基因表达效应较为敏感, 表明基因表达效应具有作为慢性水质基准数据的应用潜力, 但现有数据仍然不够丰富. 针对在基准推导中的应用, 基因表达效应研究中也存在污染物暴露浓度设置不合理以及目标基因的选择范围较小的问题.

关键词
水质基准
重金属
基因表达效应
物种敏感度分布
数据分析

水质基准(Water Quality Criteria, WQC)是制/修订水质标准(Water Quality Standard, WQS)的科学依据和理论基础, 在水环境管理中具有重要作用. 水质基准限值的推导有多种方法, 其中物种敏感度分布法(Species Sensitivity Distribution, SSD)于 20 世纪 70 年代到 80 年代由欧美科学家独立提出[1], 发展至今, 该方法已经成为推导水质基准的主流技术之一[2]. SSD 方法的原理是利用不同的函数, 如对数-三角函数[3]、对数-逻辑斯谛函数[4]或对数-正态分布[5]等, 对众多生物对同一种污染物的敏感度的分布状况进行拟合, 通过数据分析结合生态风险评估理论求得保护一定比例生物的污染物浓度作为水质基准限值, 目前 SSD 方法使用的主要是个体水平的污染物毒性数据, 如计算急性基准通常使用半致死浓度(LC_{50})和半效应浓度(EC_{50})等[6], 计算慢性基准通常使用无观测效应浓度(No Observed Effect Concentration, NOEC)和最低可见效应浓度(Low Observed Effect Concentration, LOEC)等[7]. 除个体水平外, 其他生物组织水平的毒性效应数据在水质基准推导中应用很少, 在少数情况下, 偶尔作为验证数据使用[3].

随着环境污染物的广泛迁移与分布, 长期以来, 人们在分子、细胞、组织、个体、种群、群落甚至生态系统水平上开展了广泛的生态毒理学研究[8], 深入分析环境污染物对生态环境的危害, 积累了大量的生态毒理学数据. 20 世纪 90 年代以来, "组学"技术开始应用于生态毒理学研究领域[9], 其中包括"生态毒理基因组学", 在基因水平上表征环境污染物或化学物质对生物体的毒性效应成为研究热点[10], 人们普遍认为环境毒物对基因表达的影响要灵敏于个体水平的效应指标, 基因表达效应终点对环境毒物的应

英文引用格式: Yan Z G, Yang N Y, Wang X N, et al. Preliminary analysis of species sensitivity distribution based on gene expression effect. Sci China Earth Sci, 2012, doi: 10.1007/s11430-012-4425-0

激响应远远早于生物个体的存活和生殖效应指标, 因此污染物胁迫下的基因表达效应成为环境风险早期预警的重要研究对象[11~14], 同时也为这些数据在水质基准推导中的使用提供了可能. 但是, 也有部分学者对基因表达效应数据在环境风险早期预警中的应用存疑[15,16].

为评估基因表达效应数据在风险评估中的应用, Fedorenkova 等[17]利用荷兰国立公共卫生与环境研究院(RIVM)的毒性数据库(e-toxBase)和发表的文献数据, 以基因毒性数据最丰富的重金属镉为例, 对比分析了生物个体水平的急性、慢性毒性数据与基因表达效应数据, 出人意料地发现基因表达效应对镉的灵敏度虽然大于急性毒性指标, 但同时显著低于慢性毒性指标, 因而提出分子水平上的基因表达效应并不一定是最敏感的毒理学终点, 但因为该文数据分析的不确定性, 其结论受到一定的质疑[18]. 本文利用数据量更丰富的 ECOTOX 毒性数据库(http://cfpub.epa.gov/ecotox/)、TOXNET 数据库(http://toxnet.nlm.nih.gov)、GEO 数据库(www.ncbi.nlm.nih.gov/geo)和中国知网(www.cnki.net)以及公开发表的文献数据, 重新分析了重金属镉的急性、慢性和基因表达效应数据的敏感性, 同时, 利用铜和锌的毒性数据对镉的研究结果进行了验证, 对基因表达效应数据在环境风险评估与水质基准中的应用进行了探讨.

1 材料与方法

1.1 数据搜集与筛选

利用上述数据库广泛搜集重金属镉、铜和锌对水生生物的毒性数据, 同时, 利用 Web of Science, Scopus 和 Scirus 等文献数据库搜索公开发表的文献数据, 搜索所用关键词包括“镉”、“铜”、“锌”、“重金属”、“生态毒性”、“基因表达”、“基因芯片”、“基因组学”、“蛋白组学”、“转录组学”、“风险评估”、“风险分析”和“生态风险”等, 并以所得文献的引用文献作为补充.

将搜集到的文献数据进行筛选, 筛选标准为: 急性毒性数据指标为 LC_{50} 和 EC_{50}, 暴露时间为 2~4 d, 不同暴露时间的数据以时间较长的优先; 慢性毒性数据指标为 NOEC 和 LOEC, 暴露时间为 7 d 以上, 不同暴露时间的数据以时间较长的优先; 同一物种不同生命阶段的毒性数据以敏感生命阶段的数据优先; 基因表达效应数据不考虑暴露时间, 以敏感数据优先, 为方便统一比较分析, 将获取的能引起基因表达的污染物最小浓度作为基因表达效应的 LOEC.

1.2 数据分析

将搜集且筛选过的数据进行合并、排序, 同一物种的多个同类毒性数据取几何平均值作为该物种的急性、慢性或基因毒性值. 将排序后的毒性数据利用对数-logistic 函数进行拟合, 构建 SSD 曲线, 函数方程为: $y = 1/[1+\exp((\alpha-x)/\beta)]$[19]. 水生生物被危害的比例为 5%时的污染物浓度(HC_5)依据文献方法[5,19]求出.

2 结果

2.1 重金属对基因表达的毒性效应

针对镉、铜和锌 3 种重金属, 本研究广泛搜集了急性、慢性和基因效应毒性数据, 其中包括镉的 82 种生物的急性毒性数据、12 种生物的慢性毒性数据, 铜的 118 种生物的急性毒性数据和 35 种慢性毒性数据, 锌的 81 种急性毒性数据和 15 种慢性毒性数据(见电子版附表 1~6, http:earth.scichina.com).

本文搜集的基因表达效应数据明显少于急性和慢性毒性数据, 共涉及 19 种水生生物, 包括 18 种生物的镉毒性数据、7 种生物的铜毒性数据和 3 种生物的锌毒性数据, 其中以镉的基因表达毒性数据最为丰富(表 1), 18 种水生生物中, 鱼类占 8 种, 表明人们对于环境污染物对鱼类的影响比较关注. 现有数据显示, 爪哇青鳉在基因水平上对镉的反应最灵敏, 最不敏感的是日本虎斑猛水蚤, 两者相差 5 万倍; 铜毒性相关的 5 种水生生物中, 最敏感的是大型溞, 最不敏感的是鲫鱼, 两者相差 50 倍; 锌毒性数据较少, 其中大型溞和太平洋牡蛎的敏感性相差近 10 倍.

19 种生物的基因表达毒性数据中, 暴露浓度的组数设置不一, 有 14 篇文献仅设置了 1 组暴露浓度, 仅有 1 篇文献设置了 5 组暴露浓度, 其他都不超过 5 组暴露浓度, 与标准毒性测试方法(急性或慢性)相比[44], 暴露浓度的设置很少. 而且从实验结果可以看出, 对于设计了多组暴露浓度的实验, 基于现有数据, 最小的暴露浓度往往就是基因表达效应的 LOEC,

表 1　镉、铜和锌诱导的水生生物的基因表达效应

重金属	物种	暴露浓度(μg/L)	暴露时间	LOEC(μg/L)(暴露时间)	文献
Cd	嗜热四膜虫(*T. thermophila*)	495, 989, 2473, 3956, 4946	1 h	3956(1 h)	[20]
	日本虎斑猛水蚤(*T. japonicus*)	5000, 10000, 15000	96 h	5000(96 h)	[21]
	大型溞(*D. magna*)	6, 20, 37	24 h	6(24 h)	[22]
	摇蚊幼虫(*C. tentans*)	200, 2000, 20000	24 h	200(24 h)	[23]
	鸟蛤(*A.granosa*)	250	4, 8, 12, 16 d	250(4 d)	[24]
	贻贝(*M. edulis*)	200	4, 21 d	200(4 d)	[25]
	牡蛎(*C. virginica*)	50	48 h	50(48 h)	[26]
	鲫鱼(*C. auratus*)	50, 500, 2500	12, 24, 36 h	50(12 h)	[27]
	比目鱼(*P. flesus*)	50	1, 2, 4, 8, 16 d	50(1 d)	[28]
	鲤鱼(*C. carpio*)	9.4, 105, 480	3, 42 h, 7, 28 d	9.4(3 h)	[29]
	鳉鱼(*K. marmoratus*)	610	6, 12, 24, 48, 96 h	610(48 h)	[30]
	爪哇青鳉(*O. javanicus*)	0.1, 10, 1000	24 h	0.1(24 h)	[31]
	河鲀(*T. obscurus*)	2050	6, 12, 24, 48, 96 h	2050(6 h)	[32]
	虹鳟(*O. mykiss*)	560	6 h	560(6 h)	[33]
	紫贻贝(*M. galloprovincialis*)	200	2 , 9 d	200(2 d)	[34]
	环节动物(*Laeonereis acuta*)	100, 1000	14 d	100(14 d)	[35]
	斑马鱼(*D. rerio*)	1.9, 9.6	7, 21 d	1.9(7 d)	[36]
	太平洋牡蛎(*C. gigas*)	5.32	14 d	5.32(14 d)	[37]
Cu	大型溞(*D. magna*)	2, 4	24 h	2(24 h)	[38]
	中华绒螯蟹(*E. sinensis*)	50, 100, 500, 1000	7 d	50(7 d)	[39]
	鲫鱼(*C. auratus*)	100, 1000, 5000	12, 24, 36 h	100(12 h)	[27]
	日本虎斑猛水蚤(*T. japonicus*)	4.74	6, 12, 24 h	4.74(6 h)	[40]
	虹鳟(*O. mykiss*)	19.2	6, 12, 24, 48 h	19.2(24 h)	[41]
	斑马鱼(*D. rerio*)	8, 15	21 d	8(21 d)	[42]
	紫贻贝(*M. galloprovincialis*)	40	2, 9 d	40(2 d)	[34]
Zn	大型溞(*D. magna*)	100, 200, 500, 1000	24 h	100(24 h)	[43]
	虹鳟(*O. mykiss*)	915.46	6, 12, 24, 48 h	915.46(24 h)	[41]
	太平洋牡蛎(*C. gigas*)	994.5	14 d	994.5(14 d)	[37]

更低的污染物浓度下是否有基因表达效应未知. 按照推算LOEC的国际标准试验方法, 想要科学推算出基因表达效应的 LOEC, 污染物浓度的设置还需进一步改善. 另外, 基因效应研究中暴露时间的设置也差异很大, 最短为 1 h(嗜热四膜虫), 最长为 7 d(中华绒螯蟹), 基于作为早期预警指标的考虑, 在筛选数据时, 暴露时间的长短没有被作为筛选的指标之一.

2.2　急性、慢性与基因表达效应数据的对比

为比较急性、慢性和基因表达效应数据之间的敏感性差异, 利用对数-logistic 函数分别对重金属镉、铜和锌的急性、慢性和基因表达 3 类数据进行分布拟合, 结果示于图 1, 其中锌的基因表达效应数据由于太少(仅有 3 个)没有进行拟合. 由图 1 可以看出, 镉和铜的 SSD 曲线斜率相对较小, 而锌的 SSD 曲线斜率则较大, 表明对于锌来说, 大部分水生生物的敏感度相对接近, 而不同水生生物对于镉和铜的敏感度则差异较大. 就急性、慢性和基因表达效应这 3 类数据的敏感性来说, 不同的重金属显示出差异性, 镉的数据敏感度排序为“慢性>基因>急性”; 铜和锌的基因毒性数据不足, 但从趋势上看, 锌与镉类似; 铜则表现为基因表达效应数据更敏感, 其中在高浓度区间, 基因表达效应数据的灵敏度与慢性数据接近. 3 种重金属的共同之处都是急性毒性数据最不敏感, 但慢性毒性数据与基因表达效应数据的敏感性不统一.

为便于对 3 种重金属的急性、慢性和基因表达效应数据进行量化对比, 分别对能保护 95%的水生生物的污染物浓度(HC_5)进行计算, HC_5 是计算保护水生生物的水质基准的重要依据, 计算结果示于表 2. 由表中可以看出, 对于镉和铜来说, 慢性效应指标的灵敏度远远大于急性指标, 分别为 27 倍和 6 倍, 表明对于水环境的日常管理与保护, 慢性指标远比急性指标重要. 对于锌, 慢性效应的灵敏性不到急性效应

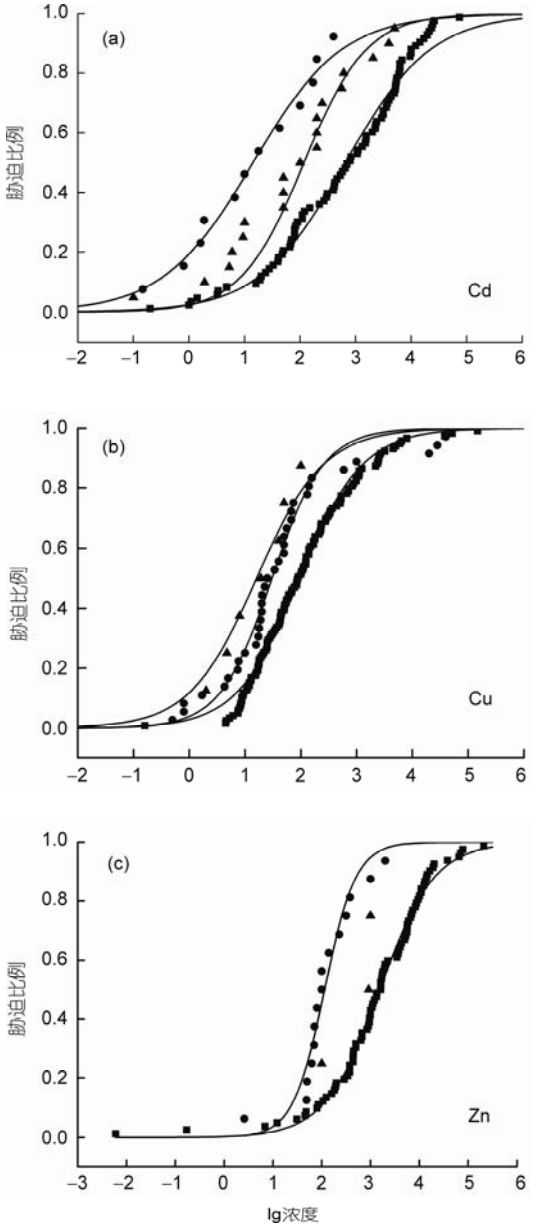

图 1　镉、铜和锌对水生生物的急性、慢性和基因表达效应数据的 SSD 曲线

■表示急性毒性数据, ●表示慢性毒性数据, ▲表示基因表达效应数据, 曲线由对数-logistic 函数拟合

的 2 倍, 差异性比镉和铜要小. 由于基因表达效应的数据欠缺, 只有镉比较丰富, 可以进行拟合, 计算 HC_5, 计算结果表明在保护水环境中 95%的水生生物的水平上, 基因表达效应指标比急性效应指标灵敏 7 倍多, 但却比慢性指标迟钝 3 倍多. 对于铜和锌来说, 基因表达效应指标数据太少而无法准确计算 HC_5, 其

表 2　镉、铜、锌对水生生物的 HC_5[a)]

重金属	数据类别	HC_5(μg/L)	90%置信区间(μg/L)
Cd	个体水平-急性	5.26	2.23~10.8
	个体水平-慢性	0.197	0.0147~0.938
	基因表达效应	0.717	0.0907~2.91
Cu	个体水平-急性	2.98	1.70~4.82
	个体水平-慢性	0.529	0.140~1.45
	基因表达效应*	/	/
Zn	个体水平-急性	16.6	7.39~32.8
	个体水平-慢性	9.09	2.36~21.6
	基因表达效应*	/	/

a) *, 因数据量太少(n<10), 不进行拟合[1]

中铜的基因表达效应数据有 7 个, 本文构建了其 SSD 曲线以分析敏感度趋势(图 1), 锌的基因表达效应数据只有 3 个, 仅在图 1 中显示位置, 没有构建 SSD 曲线.

3　讨论

随着生态毒理学研究的深入, 个体水平毒性效应数据在水质基准推算中的应用越来越体现出局限性. 基于更宏观、更微观或者更综合的毒性效应指标对水质基准进行表征是新趋势. 通常慢性毒性效应指标比急性毒性效应指标更加灵敏, 在日常环境管理也具有更重要的作用, 本文研究也验证了这一点. 随着“组学”技术的发展及其在环境科学领域的应用, 对环境污染物发生应激响应的基因表达量的变化成为生态风险评估的研究热点, 一般认为在微观水平上的基因表达量的变化要早于细胞、组织、个体、种群以及群落等诸多水平上的效应变化[11,13], 因此, 近年有大量文献在基因水平上进行生态毒理学的研究, 并且将之与个体、种群等水平的研究结果相联系, 希望能够搭建起微观与宏观预警指标之间的桥梁, 将各水平上的研究结论统一起来, 使基因表达效应指标在环境风险评估中发挥更大作用[23,45~47].

随着生态毒理学中基因毒性数据的积累, 使得利用数据分析方法(如 SSD 法)研究基因效应指标与更宏观水平效应指标之间的关系成为可能. 最近 Fedorenkova 等[17]通过广泛的文献调研(同时基于 RIVM 的毒性数据库 e-toxBase[17]), 以基因表达效应数据最丰富的镉为例, 分析了镉的急性、慢性和基因表达效应数据三者之间的关系, 并且出人意料地发现, 在镉的现有毒性数据中, 基因表达效应指标远没

有慢性毒性效应指标敏感, 因此对基因作为生态风险早期预警指标的适用性提出了商榷, 当然, 作者也指出, 由于基因表达效应的数据还不够丰富和完善, 仅通过镉的毒性数据分析得出的结论还需要进一步验证与研究[17]. 基于此, 本研究选择了更为丰富的生物毒性数据, 对镉的急性、慢性和基因表达效应 3 类数据重新进行了分析, 另外, 还选择了基因效应数据相对丰富的铜和锌两种重金属对镉的研究结论进行验证. 结果表明, 对镉的 3 类数据进行分析得出的结论与 Fedorenkova 等[17]得出的结论相同, 敏感性排序都是“慢性>基因>急性”. 由于铜和锌的基因数据不足, 只能从趋势上大致判断, 锌与镉的数据敏感度类似; 但铜的基因表达效应数据的灵敏度从趋势上大于慢性毒性效应数据, 特别是在对环境管理有重要意义的低浓度区间, 差异较为明显, 显示了基因表达效应指标可能具有作为生态风险评估指标的潜力, 其分布的逻辑性也为在水质基准研究中的应用提供了可能, 但还需要更进一步的验证研究.

镉和锌的数据分析显示基因指标的敏感性低于慢性指标, 即使是铜, 基因指标的灵敏性优势与慢性指标相比也不十分明显. 仔细分析表 1 可知, 除嗜热四膜虫外, 暴露浓度的最小值就是基因表达指标的 LOEC, 这表明在基因表达上, 几乎可以肯定还存在更小值的 LOEC, 现在能从文献中获取的所谓的基因表达的“LOEC”很可能不是真正的 LOEC, 如果实验中继续设置更低浓度的暴露浓度, 基因表达效应的 LOEC 值将继续降低, 即基因表达效应的灵敏度还会提升. 另一个重要原因是, 生物体内的基因上万甚至十几万, 目前文献中所选择研究的基因很可能并不是对该污染物最早应激响应的靶标基因, 因此, 只有通过基因组学技术进行高通量地筛选目标基因, 才有可能找到最敏感的响应基因, 正确评价基因表达效应指标的灵敏性.

在灵敏度的定量分析中, 本文选择了对于确定水质基准具有重要参考价值的 HC_5 进行对比分析, 遗憾的是铜和锌的基因表达效应数据不足而无法计算 HC_5, 但就现阶段而言, 镉、铜和锌已经是基因效应数据相对丰富的污染物[17], 目前没有更多有效的基因效应数据可以用于分析计算. 由于数据的限制, HC_5 计算过程中也没有严格依照水质基准技术中的“最小毒性数据需求”原则[3,48], 仅进行大概推算以比较数据灵敏性. 与 Fedorenkova 等[17]计算的镉的 3 类数据的 HC_5(急性: 18.3 μg/L; 慢性: 0.23 μg/L; 基因: 0.72 μg/L[17])相比, 本研究得出的镉急性 HC_5 明显降低(5.26 μg/L), 慢性 HC_5 相差不大(0.197 μg/L), 基因效应 HC_5 基本相同(0.717 μg/L). 这主要与分析所利用的数据组有关, 比如本文筛选得到的镉的急性数据量(82 组数据)远远大于 Fedorenkova 等[17]从 e-toxBase 数据库中选择的急性数据量(20 组数据), 由此也大大影响了急性 HC_5 的计算. 然而, 虽然镉的数据选择有差异, 但得出的结论是相同的.

一般认为, 在分子水平上对污染物胁迫发生应激响应的基因表达效应具有良好的生态风险早期预警指标的应用潜力, 但现有数据尚难以大范围地支持其在实践中应用, 还需要通过各种“组学”技术对目标基因进行高通量筛选, 才能确定对特定污染物发生特异性响应的靶标基因, 针对基因表达效应终点的毒性试验方法也需进一步改进, 以便使基因表达效应真正在生态风险早期预警中发挥重要作用, 同时也能有效促进其中水质基准研究中的应用.

致谢 感谢审稿人提出的修改建议.

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附表 1　镉对水生生物的急性毒性

物种名称	终点	暴露时间(d)	毒性值(μg/L)	文献
Acanthopagrus schlegeli	LC_{50}	2	2300	[1]
Acartia tonsa	LC_{50}	4	90	[2]
Americamysis bahia	LC_{50}	4	110	[3]
Americamysis bahia	LC_{50}	4	17.93	[4]
Americamysis bahia	LC_{50}	4	11.1	[5]
Ampelisca abdita	LC_{50}	4	1320	[6]
Amphiascus tenuiremis	LC_{50}	4	224	[7]
Anguilla rostrata	LC_{50}	4	820	[8]
Anodonta cygnea	EC_{50}	3	4.8	[9]
Artemia salina	LC_{50}	2	1540	[10]
Asellus aquaticus	LC_{50}	4	60	[11]
Asellus aquaticus	LC_{50}	4	53	[12]
Asellus aquaticus	LC_{50}	4	160	[13]
Baetis rhodani	LC_{50}	4	50	[11]
Brachionus calyciflorus	EC_{50}	2	70	[14]
Brachionus plicatilis	LC_{50}	3	5200	[15]
Ceriodaphnia reticulata	LC_{50}	2	66	[16]
Channa punctata	LC_{50}	4	11200	[17]
Channa punctata	LC_{50}	4	18500	[18]
Channa striata	LC_{50}	3	19400	[19]
Chasmichthys dolichognathus	LC_{50}	4	5500	[1]
Chironomus plumosus	LC_{50}	4	400	[20]
Chironomus riparius	LC_{50}	4	4320	[21]
Chironomus tentans	LC_{50}	4	80	[21]
Chrysophrys major	LC_{50}	4	270	[22]
Crangon crangon	LC_{50}	4	460	[23]
Crassostrea rhizophorae	LC_{50}	4	5000	[24]
Culicoides furens	LC_{50}	4	300	[20]
Cyprinus carpio	LC_{50}	4	3000	[25]
Cyprinus carpio	LC_{50}	4	4260	[26]
Daphnia lumholzi	LC_{50}	4	83	[27]
Daphnia magna	EC_{50}	2	24.4	[28]
Daphnia magna	LC_{50}	2	24	[29]
Daphnia magna	LC_{50}	2	118	[16]
Daphnia obtusa	EC_{50}	2	580	[30]
Daphnia pulex	LC_{50}	2	68	[16]
Dendrocoelum lacteum	LC_{50}	4	23220	[13]
Dugesia tigrina	LC_{50}	4	2250	[31]
Ephemera japonica	LC_{50}	2	6000	[1]
Ephemerella ignita	LC_{50}	4	12000	[11]
Eurytemora affinis	LC_{50}	4	120	[32]
Fundulus diaphanus	LC_{50}	4	110	[8]
Gammarus lacustris	LC_{50}	4	40.1	[33]
Gammarus pulex	LC_{50}	4	20	[11]
Garra mullya	LC_{50}	4	5400	[34]
Girella punctata	LC_{50}	4	17000	[1]
Grandidierella japonica	LC_{50}	4	3140	[6]
Haliotis rubra	LC_{50}	4	3700	[35]
Hexagenia rigida	LC_{50}	4	6200	[36]
Homarus americanus	LC_{50}	4	110	[37]
Lagodon rhomboides	LC_{50}	4	10000	[38]

续附表 1

物种名称	终点	暴露时间(d)	毒性值(μg/L)	文献
Lamellidens marginalis	LC_{50}	4	10000	[39]
Lamellidens marginalis	LC_{50}	4	10000	[40]
Lates calcarifer	LC_{50}	4	9600	[41]
Lepomis gibbosus	LC_{50}	4	1500	[8]
Lepomis macrochirus	LC_{50}	4	21100	[42]
Leuctra inermis	LC_{50}	4	26000	[11]
Limnodrilus hoffmeisteri	LC_{50}	4	2400	[11]
Litopenaeus vannamei	LC_{50}	4	1070	[43]
Liza macrolepis	LC_{50}	4	2940	[44]
Liza vaigiensis	LC_{50}	4	5250	[45]
Lophopodella carteri	LC_{50}	4	150	[46]
Lutjanus argentimaculatus	LC_{50}	4	6300	[47]
Menidia peninsulae	LC_{50}	4	310	[48]
Morone americana	LC_{50}	2	1100	[8]
Morone saxatilis	LC_{50}	4	19	[49]
Mugil cephalus	LC_{50}	4	6600	[1]
Mugil cephalus	LC_{50}	4	7079	[50]
Mytilus edulis	LC_{50}	4	1550	[51]
Notemigonus crysoleucas	LC_{50}	4	3150	[52]
Oncorhynchus mykiss	LC_{50}	4	3	[53]
Oncorhynchus mykiss	LC_{50}	4	6	[54]
Oncorhynchus mykiss	LC_{50}	4	2.1	[55]
Oncorhynchus tshawytscha	LC_{50}	4	1.41	[56]
Oryzias latipes	LC_{50}	4	16	[57]
Pagrus major	LC_{50}	4	650	[1]
Palaemon serratus	LC_{50}	4	4000	[58]
Palaemonetes pugio	LC_{50}	4	1.1	[59]
Paralichthys olivaceus	LC_{50}	2	2000	[60]
Pectinatella magnifica	LC_{50}	4	700	[46]
Penaeus chinensis	LC_{50}	4	33	[61]
Penaeus indicus	LC_{50}	2	420	[10]
Penaeus merguiensis	LC_{50}	2	800	[41]
Penaeus monodon	LC_{50}	4	48	[41]
Periophthalmus waltoni	LC_{50}	4	3.25	[62]
Perna viridis	LC_{50}	2	6300	[10]
Physa fontinalis	LC_{50}	4	80	[11]
Physa gyrina	LC_{50}	4	410	[63]
Pimephales promelas	LC_{50}	4	1	[64]
Platichthys flesus	LC_{50}	4	24400	[65]
Polycelis tenuis	LC_{50}	4	74000	[11]
Rhepoxynius abronius	LC_{50}	4	530	[6]
Rhithropanopeus harrisii	LC_{50}	4	0.2	[59]
Simocephalus vetulus	LC_{50}	2	24	[16]

附表 2　镉对水生生物的慢性毒性

物种名称	终点	暴露时间(d)	毒性值(μg/L)	文献
Brachionus calyciflorus	NOEC	4	10	[66]
Crassostrea gigas	LOEC	21	200	[67]
Crassostrea virginica	LOEC	20	42.9	[68]
Cyprinus carpio	LOEC	28	6.7	[69]
Daphnia magna	LOEC	14	36.4	[70]
Littorina littorea	LOEC	46	400	[71]
Mytilus edulis	LOEC	8	250	[51]
Mytilus edulis	LOEC	21	200	[67]
Mytilus edulis	LOEC	7	100	[72]
Mytilus galloprovincialis	LOEC	41	0.8	[73]
Mytilus galloprovincialis	LOEC	41	0.8	[74]
Oncorhynchus mykiss	LOEC	30	1.5	[53]
Oncorhynchus mykiss	LOEC	100	1.74	[55]
Oncorhynchus tshawytscha	LOEC	133	1.88	[56]
Ruditapes decussatus	LOEC	7	100	[75]
Salmo trutta	LOEC	7	0.1473	[76]
Salvelinus fontinalis	LOEC	10	18	[77]

附表 3　铜对水生生物的急性毒性

物种名称	终点	暴露时间(d)	毒性值(μg/L)	文献
Acantholeberis curvirostris	EC_{50}	2	11.9	[78]
Acartia tonsa	LC_{50}	4	17	[2]
Acroperus elongatus	EC_{50}	2	15.2	[78]
Acroperus elongatus	EC_{50}	2	27.7	[79]
Acroperus harpae	EC_{50}	2	14.4	[78]
Acroperus harpae	EC_{50}	2	11.5	[80]
Adenophlebia auriculata	LC_{50}	4	180	[81]
Alona quadrangularis	EC_{50}	2	28.2	[78]
Americamysis bahia	LC_{50}	4	181	[3]
Amphiascus tenuiremis	LC_{50}	4	124.6	[82]
Anadara granosa	LC_{50}	4	170	[83]
Anguilla rostrata	LC_{50}	4	6000	[8]
Artemia salina	LC_{50}	2	440	[10]
Astacus astacus	LC_{50}	4	148400	[84]
Balanus improvisus	LC_{50}	4	100	[85]
Barbus gonionotus	LC_{50}	4	530	[86]
Bosmina longirostris	EC_{50}	2	9.2	[78]
Brachionus calyciflorus	EC_{50}	2	26	[14]
Carcinus maenas	LC_{50}	4	51800	[87]
Catostomus commersoni	LC_{50}	4	886	[88]
Ceriodaphnia dubia	EC_{50}	2	1.6	[89]
Ceriodaphnia dubia	EC_{50}	2	1.6	[90]
Ceriodaphnia dubia	EC_{50}	2	11.8	[80]
Ceriodaphnia dubia	EC_{50}	2	1.9	[91]
Ceriodaphnia dubia	LC_{50}	2	12	[92]
Ceriodaphnia dubia	LC_{50}	2	14	[93]
Ceriodaphnia pulchella	EC_{50}	2	12	[78]
Ceriodaphnia pulchella	EC_{50}	2	1.7	[79]
Ceriodaphnia quadrangula	EC_{50}	2	34.6	[94]
Ceriodaphnia reticulata	EC_{50}	2	13.3	[78]

续附表 3

物种名称	终点	暴露时间(d)	毒性值(μg/L)	文献
Ceriodaphnia reticulata	EC_{50}	2	5.5	[79]
Ceriodaphnia reticulata	LC_{50}	2	17	[16]
Channa punctata	LC_{50}	4	339	[95]
Chironomus plumosus	LC_{50}	4	530	[21]
Chironomus riparius	LC_{50}	4	700	[96]
Chironomus riparius	LC_{50}	4	640	[21]
Chironomus tentans	LC_{50}	4	310	[21]
Chrysophrys major	LC_{50}	4	70	[22]
Chydorus ovalis	EC_{50}	2	33.4	[78]
Chydorus ovalis	EC_{50}	2	9.6	[79]
Chydorus sphaericus	EC_{50}	2	14.1	[78]
Chydorus sphaericus	EC_{50}	2	11.2	[79]
Chydorus sphaericus	EC_{50}	2	23.1	[80]
Cirrhinus mrigala	LC_{50}	4	110	[97]
Clarias gariepinus	LC_{50}	4	4540	[98]
Crassostrea rhizophorae	LC_{50}	7	2500	[24]
Culicoides furens	LC_{50}	4	38400	[20]
Cyprinodon variegatus	LC_{50}	4	368	[99]
Cyprinus carpio	LC_{50}	4	300	[100]
Cyprinus carpio	LC_{50}	4	50	[101]
Cypris subglobosa	LC_{50}	4	277.3	[27]
Daphnia galeata	EC_{50}	2	22.6	[78]
Daphnia galeata	EC_{50}	2	3.2	[79]
Daphnia longispina	EC_{50}	2	9.89	[78]
Daphnia longispina	EC_{50}	2	2.1	[79]
Daphnia longispina	EC_{50}	2	11	[94]
Daphnia lumholzi	LC_{50}	4	9.4	[27]
Daphnia magna	EC_{50}	2	30	[78]
Daphnia magna	EC_{50}	2	66.8	[102]
Daphnia magna	EC_{50}	2	26.8	[80]
Daphnia magna	EC_{50}	2	31.8	[103]
Daphnia magna	EC_{50}	2	36	[104]
Daphnia magna	LC_{50}	2	33	[105]
Daphnia magna	LC_{50}	2	54	[16]
Daphnia magna	LC_{50}	3	82	[106]
Daphnia magna	LC_{50}	3	84	[106]
Daphnia obtusa	EC_{50}	2	17	[30]
Daphnia pulex	LC_{50}	2	37	[107]
Daphnia pulex	LC_{50}	2	53	[16]
Disparalona rostrata	EC_{50}	2	43.3	[78]
Donax faba	LC_{50}	4	170	[83]
Dugesia tigrina	LC_{50}	4	2450	[31]
Epioblasma capsaeformis	EC_{50}	4	16	[108]
Etroplus maculatus	LC_{50}	4	50	[109]
Eurycea bislineata	LC_{50}	2	1120	[107]
Eurycercus lamellatus	EC_{50}	2	12.9	[94]
Eurycercus lamellatus	EC_{50}	2	24.3	[80]
Eurytemora affinis	LC_{50}	4	7.5	[32]
Fundulus diaphanus	LC_{50}	4	840	[8]
Galaxea fascicularis	LC_{50}	4	32	[110]
Gambusia affinis	LC_{50}	4	56	[111]

续附表 3

物种名称	终点	暴露时间(d)	毒性值(μg/L)	文献
Gammarus lacustris	LC_{50}	4	212	[33]
Gammarus pulex	LC_{50}	4	37	[96]
Haliotis rubra	LC_{50}	4	87	[35]
Heliocidaris tuberculata	EC_{50}	3	8	[112]
Homarus americanus	LC_{50}	4	85	[37]
Hydra oligactis	LC_{50}	4	84	[113]
Hydra viridissima	LC_{50}	4	25	[113]
Hydra vulgaris	LC_{50}	4	42	[113]
Ictalurus punctatus	LC_{50}	4	51	[114]
Isonychia bicolor	LC_{50}	2	223	[107]
Labeo rohita	LC_{50}	4	234	[115]
Lamellidens marginalis	LC_{50}	4	5000	[39]
Lamellidens marginalis	LC_{50}	4	5000	[40]
Lampsilis fasciola	EC_{50}	4	25	[108]
Lampsilis rafinesqueana	EC_{50}	4	43	[108]
Lates calcarifer	LC_{50}	4	3200	[41]
Lecane hamata	LC_{50}	2	230	[116]
Lecane luna	LC_{50}	2	60	[116]
Lecane quadridentata	LC_{50}	2	330	[116]
Leiostomus xanthurus	LC_{50}	4	282	[48]
Lepomis gibbosus	LC_{50}	4	2700	[8]
Lepomis macrochirus	LC_{50}	4	1250	[117]
Leptodea leptodon	EC_{50}	4	22	[108]
Liza vaigiensis	LC_{50}	4	2550	[45]
Lophopodella carteri	LC_{50}	4	510	[46]
Macrobrachium rosenbergii	LC_{50}	4	55	[118]
Menidia beryllina	LC_{50}	4	123.04	[119]
Menidia peninsulae	LC_{50}	4	141	[48]
Monhystera disjuncta	EC_{50}	4	2200	[120]
Morone americana	LC_{50}	4	6400	[8]
Morone saxatilis	LC_{50}	4	24	[120]
Mytilus edulis	EC_{50}	4	8.29	[119]
Neanthes arenaceodentata	LC_{50}	4	77	[121]
Oncorhynchus kisutch	LC_{50}	4	61	[122]
Oncorhynchus mykiss	LC_{50}	4	89	[122]
Oncorhynchus mykiss	LC_{50}	4	52	[53]
Oncorhynchus tshawytscha	LC_{50}	4	10	[123]
Oncorhynchus tshawytscha	LC_{50}	4	33.1	[124]
Oreochromis mossambicus	LC_{50}	4	150	[125]
Pagrus major	LC_{50}	4	1100	[126]
Paphia textile	LC_{50}	4	25	[127]
Paracentrotus lividus	EC_{50}	2	32	[128]
Paracentrotus lividus	EC_{50}	3	52	[129]
Paratya australiensis	LC_{50}	4	99	[130]
Paratya australiensis	LC_{50}	4	108	[130]
Pectinatella magnifica	LC_{50}	4	140	[46]
Penaeus chinensis	LC_{50}	4	18	[61]
Penaeus indicus	LC_{50}	2	1120	[10]
Penaeus merguiensis	LC_{50}	5	90	[41]
Penaeus monodon	LC_{50}	4	12	[41]
Periophthalmus waltoni	LC_{50}	4	7.56	[62]

续附表 3

物种名称	终点	暴露时间(d)	毒性值(μg/L)	文献
Pimephales promelas	EC_{50}	4	201	[131]
Pimephales promelas	LC_{50}	4	300	[132]
Pimephales promelas	LC_{50}	4	200	[133]
Pleuroxus truncatus	EC_{50}	2	51.6	[78]
Plumatella emarginata	LC_{50}	4	140	[46]
Poecilia reticulata	LC_{50}	4	112	[134]
Procambarus clarkii	LC_{50}	4	720	[135]
Ruditapes decussatus	LC_{50}	4	715	[136]
Salmo salar	LC_{50}	4	125	[137]
Scapholeberis microcephala	EC_{50}	2	11.2	[78]
Scapholeberis mucronata	EC_{50}	2	5.3	[78]
Sciaenops ocellatus	LC_{50}	4	518.1	[138]
Simocephalus exspinosus	EC_{50}	2	16.6	[78]
Simocephalus exspinosus	EC_{50}	2	4.6	[79]
Simocephalus vetulus	EC_{50}	2	16.1	[78]
Simocephalus vetulus	EC_{50}	2	4.1	[79]
Simocephalus vetulus	EC_{50}	2	10.3	[80]
Strongylocentrotus intermedius	LC_{50}	4	30	[139]
Terapon jarbua	LC_{50}	4	2240	[140]
Terapon jarbua	LC_{50}	4	2500	[141]
Tetrahymena pyriformis	EC_{50}	2	8000	[142]
Tigriopus californicus	LC_{50}	4	19.2	[143]
Tilapia nilotica	LC_{50}	4	1000	[144]
Tubifex tubifex	LC_{50}	4	0.16	[145]
Villosa iris	EC_{50}	4	13	[108]

附表 4　铜对水生生物的慢性毒性

物种名称	终点	暴露时间(d)	毒性值(μg/L)	文献
Americamysis bahia	LOEC	4	140	[3]
Atherinops affinis	NOEC	12	55	[146]
Atherinops affinis	NOEC	7	100	[147]
Barbus gonionotus	LOEC	28	50	[86]
Bathymodiolus azoricus	LOEC	24	25	[148]
Cambarus bartonii	LOEC	7	20	[149]
Carcinus maenas	LOEC	4	40000	[87]
Ceriodaphnia dubia	LOEC	7	50	[131]
Clarias gariepinus	LOEC	8	590	[150]
Crassostrea gigas	LOEC	21	40	[67]
Crassostrea virginica	LOEC	96	4.3	[151]
Cyprinus carpio	NOEC	28	1.7	[69]
Danio rerio	LOEC	6	50	[152]
Danio rerio	LOEC	6	50	[153]
Daphnia magna	LOEC	31	20	[106]
Daphnia magna	LOEC	21	15	[154]
Daphnia pulex	NOEC	20	10	[155]
Fundulus heteroclitus	NOEC	30	22.58	[156]
Gammarus pulex	LOEC	10	7.7	[157]
Hyalella azteca	NOEC	10	13	[158]
Hyalella azteca	LOEC	35	31.56	[159]
Lepomis macrochirus	NOEC	4	131	[160]
Macrophthalmus erato	NOEC	14	67.5	[161]

续附表 4

物种名称	终点	暴露时间(d)	毒性值(μg/L)	文献
Mytilus edulis	LOEC	21	40	[67]
Mytilus edulis	LOEC	16	10	[51]
Mytilus galloprovincialis	LOEC	41	0.8	[74]
Oncorhynchus kisutch	NOEC	60	18	[122]
Oncorhynchus mykiss	LOEC	731	19	[53]
Oncorhynchus tshawytscha	LOEC	147	7.4	[124]
Pimephales promelas	NOEC	7	28300	[162]
Pomacea canaliculata	LOEC	10	67.5	[163]
Pomacea paludosa	LOEC	14	56	[164]
Rana ridibunda	LOEC	14	20000	[165]
Ruditapes decussatus	LOEC	7	0.5	[166]
Rutilus rutilus	LOEC	56	5	[167]
Salmo trutta	LOEC	15	15.76	[168]
Salvelinus fontinalis	LOEC	10	158	[77]
Terapon jarbua	LOEC	14	1000	[141]
Tigriopus angulatus	LOEC	28	34	[169]
Tilapia nilotica	LOEC	42	0.8	[170]

附表 5 锌对水生生物的急性毒性

物种名称	终点	暴露时间(d)	毒性值(μg/L)	文献
Acartia clausi	LC_{50}	4	950	[171]
Acartia tonsa	LC_{50}	4	290	[171]
Acroperus elongatus	EC_{50}	2	1614	[172]
Acroperus elongatus	EC_{50}	2	1614	[78]
Americamysis bahia	LC_{50}	4	499	[3]
Ampelisca abdita	LC_{50}	10	343	[173]
Amphiascus tenuiremis	LC_{50}	4	375.8	[82]
Anadara granosa	LC_{50}	4	1940	[83]
Anguilla rostrata	LC_{50}	4	14500	[8]
Anodonta cygnea	EC_{50}	3	45.8	[9]
Artemia salina	LC_{50}	2	1700	[10]
Callianassa kraussi	LC_{50}	4	31	[174]
Ceriodaphnia dubia	EC_{50}	2	416	[172]
Ceriodaphnia dubia	EC_{50}	2	70	[89]
Ceriodaphnia dubia	LC_{50}	2	65	[93]
Ceriodaphnia reticulata	LC_{50}	2	96	[175]
Ceriodaphnia reticulata	LC_{50}	2	76	[16]
Channa punctata	LC_{50}	4	0.006	[176]
Chironomus plumosus	LC_{50}	4	9500	[20]
Chironomus tentans	LC_{50}	10	1125	[177]
Chrysophrys major	LC_{50}	4	444	[22]
Chydorus ovalis	EC_{50}	2	1627	[172]
Chydorus ovalis	EC_{50}	2	1627	[78]
Chydorus sphaericus	EC_{50}	2	1326	[172]
Chydorus sphaericus	EC_{50}	2	1326	[78]
Cichlasoma managuense	LC_{50}	4	77980	[178]
Cirrhinus mrigala	LC_{50}	4	1633	[179]
Cottus bairdi	LC_{50}	4	439	[180]
Crassostrea gigas	EC_{50}	2	200	[181]
Culicoides furens	LC_{50}	4	3800	[20]
Cyprinus carpio	LC_{50}	4	450	[100]

续附表 5

物种名称	终点	暴露时间(d)	毒性值(μg/L)	文献
Cypris subglobosa	LC_{50}	4	8352	[27]
Daphnia galeata	EC_{50}	2	1001	[172]
Daphnia galeata	EC_{50}	2	1001	[78]
Daphnia longispina	EC_{50}	2	375	[172]
Daphnia longispina	EC_{50}	2	375	[78]
Daphnia lumholzi	LC_{50}	4	437.5	[27]
Daphnia magna	EC_{50}	2	354	[182]
Daphnia magna	LC_{50}	2	100	[105]
Daphnia magna	LC_{50}	2	68	[16]
Daphnia magna	LC_{50}	4	570	[183]
Daphnia pulex	LC_{50}	2	107	[16]
Donax faba	LC_{50}	4	3680	[83]
Dugesia tigrina	LC_{50}	4	7400	[31]
Eurytemora affinis	LC_{50}	4	4090	[171]
Fundulus diaphanus	LC_{50}	4	19200	[8]
Gammarus lacustris	LC_{50}	4	2240	[33]
Haliotis diversicolor	LC_{50}	4	1200	[184]
Haliotis rubra	LC_{50}	4	1730	[35]
Heliocidaris tuberculata	EC_{50}	3	160	[112]
Homarus americanus	LC_{50}	4	500	[37]
Hydra oligactis	LC_{50}	4	14000	[113]
Hydra viridissima	LC_{50}	4	11000	[113]
Hydra vulgaris	LC_{50}	4	13000	[113]
Labeo rohita	LC_{50}	4	10910	[115]
Leiostomus xanthurus	LC_{50}	4	37900	[48]
Lepomis gibbosus	LC_{50}	4	20100	[8]
Lepomis macrochirus	LC_{50}	4	3500	[185]
Litopenaeus vannamei	LC_{50}	4	1350	[43]
Liza vaigiensis	LC_{50}	4	12500	[45]
Lophopodella carteri	LC_{50}	4	5630	[46]
Menidia peninsulae	LC_{50}	4	5600	[48]
Monhystera disjuncta	EC_{50}	4	5700	[120]
Morone americana	LC_{50}	2	10100	[8]
Morone saxatilis	LC_{50}	2	670	[186]
Mystus vittatus	LC_{50}	4	209000	[187]
Mytilus edulis	LC_{50}	4	5000	[51]
Nereis diversicolor	LC_{50}	4	6000	[188]
Oncorhynchus clarki	LC_{50}	4	670	[53]
Oncorhynchus mykiss	LC_{50}	4	240	[53]
Oncorhynchus mykiss	LC_{50}	4	590	[183]
Oncorhynchus tshawytscha	LC_{50}	4	182	[56]
Oreochromis mossambicus	LC_{50}	4	16500	[101]
Paracentrotus lividus	EC_{50}	2	0.172	[189]
Paralichthys olivaceus	LC_{50}	2	6700	[60]
Pectinatella magnifica	LC_{50}	4	4310	[46]
Penaeus chinensis	LC_{50}	4	48	[61]
Penaeus indicus	LC_{50}	2	1990	[10]
Periophthalmus waltoni	LC_{50}	4	12.21	[62]
Perna viridis	LC_{50}	2	7000	[10]
Physastra gibbosa	LC_{50}	4	1000	[185]
Pimephales promelas	LC_{50}	4	6.9	[190]

续附表 5

物种名称	终点	暴露时间(d)	毒性值(μg/L)	文献
Plumatella emarginata	LC_{50}	4	5300	[46]
Pseudodiaptomus coronatus	LC_{50}	4	1783	[171]
Rana catesbeiana	LC_{50}	4	70000	[178]
Salmo trutta	LC_{50}	4	640	[53]
Salvelinus fontinalis	LC_{50}	4	960	[53]
Simocephalus exspinosus	EC_{50}	2	911	[172]
Simocephalus exspinosus	EC_{50}	2	911	[78]
Simocephalus vetulus	EC_{50}	2	847	[172]
Simocephalus vetulus	EC_{50}	2	847	[78]
Strongylocentrotus intermedius	LC_{50}	4	1080	[139]
Strongylocentrotus purpuratus	EC_{50}	4	80	[191]
Terapon jarbua	LC_{50}	4	8700	[140]
Tilapia nilotica	LC_{50}	3	65550	[192]

附表 6 锌对水生生物的慢性毒性

物种名称	终点	暴露时间(d)	毒性值(μg/L)	文献
Bufo arenarum	NOEC	22	2000	[193]
Cottus bairdi	LOEC	30	379	[180]
Crassostrea gigas	LOEC	21	1000	[67]
Cyprinus carpio	NOEC	28	2.6	[69]
Daphnia magna	NOEC	21	62.6	[182]
Lepidocephalichthys thermalis	LOEC	14	100	[194]
Macrophthalmus erato	NOEC	9	139	[161]
Melita plumulosa	NOEC	2	230	[195]
Mytilus edulis	LOEC	16	50	[51]
Mytilus galloprovincialis	LOEC	41	80	[74]
Oncorhynchus mykiss	LOEC	30	71	[53]
Oncorhynchus tshawytscha	LOEC	139	49	[56]
Oreochromis mossambicus	LOEC	7	100	[194]
Palaemon elegans	LOEC	21	316	[196]
Salmo trutta	LOEC	15	70	[76]

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