

## Research Article

### **Acute Toxic Impacts of Three Heavy Metals (Copper, Zinc and Cadmium) on *Ceriodaphnia reticulata* (Cladocera)**

Ayda Telliöglü

Department of Biology, Mustafa Kemal University, Hatay, Turkey

#### **\*Corresponding author**

Ayda Telliöglü

Email: [atellioglu69@gmail.com](mailto:atellioglu69@gmail.com)

---

**Abstract:** *Ceriodaphnia reticulata* (Cladocera: Daphniidae) is a common limnetic species in summer-temperate and tropical water bodies. In this study, that performed acute toxicity tests of three heavy metals, copper (Cu), zinc (Zn), and cadmium (Cd), to *C. reticulata*. For *C. reticulata*, the lethal concentration (LC<sub>50</sub>) values of Cu (24-h LC<sub>50</sub>=18.7 µg/L, 48-h LC<sub>50</sub>= 12.7 µg/L) and Zn (24-h LC<sub>50</sub>=258.5 µg/L, 48-h LC<sub>50</sub>=182.1 µg/L) were lower than those for *D. magna* and *L. Kindtii*, esed test organisms for toxic chemicals. On the other hand, for *C. reticulata*the 24-h LC<sub>50</sub> of Cd (183.2 µg/L) was much greater than that for *D. magna* and *L. Kindtii*, and the 48-h LC<sub>50</sub> of Cd (82.1 µg/L) was comparable. This results indicate that *C. reticulata* may be more strongly influenced by Zn and Cu than is *D. magna* and *L. kindtii*. It is likely that the summer plankton community in which *Ceriodaphnia* species is dominant is more sensitive to heavy metals than a community in which *Daphnia* species are dominant.

**Keywords:** Cladocera, Ceriodaphnia, Heavy metal, Toxicity

---

## **INTRODUCTION**

Aquatic ecosystems are the final sink for all potentially toxic metals in the environment via transfer from natural and/or anthropogenic sources. The increasing use of contaminating chemicals in many industrialised parts of the world makes the development of ecotoxicity measurement techniques an absolute necessity [1].

Cladoceran species are important aquatic organisms because they transfer energy and materials from primary producers to higher trophic feeders, such as fish. At the same time, they are also one of the most sensitive species to toxic chemicals [2]. Life table demographic responses of cladocerans are used for the assessment of water quality, including metal toxicity [3]. Most ecotoxicological works have considered continuous exposure of the test species to toxicants [4]. However, since the release of industrial effluents into natural waterbodies is pulsed and subsequently diluted, their impact on aquatic organisms remains mostly unknown [5]. Consequently, zooplankton that predominate in these altered systems, are in a constant physiological challenge as well as subjected to different levels of stress during their life cycle [6, 7]. While *Daphnia*, mainly *D. magna* has received considerable importance as a test organism in ecotoxicological evaluations [8], in terms of sensitivity, *Ceriodaphnia* is comparable to daphnids [9].

*Ceriodaphnia reticulata*, littoral and limnetic, usually nearshore or in the warmer upper layers of water. Reproduction during most of the year is parthenogenetic, but when environmental conditions became unfavorable, males are produced and sexual reproduction occurs in *C. reticulata*, widespread usage in aquatic eco-toxicity laboratories.

The sensitivity of *C. reticulata* to toxic chemicals may differ from that of temperate *Daphnia* species. Investigating the impact of heavy metals on *Ceriodaphnia* species may be important for understanding the impact of toxic chemicals on the summer-temperate community with dominance of non-*Daphnia* species. I carried out acute toxicity tests of three heavy metals, copper (Cu), zinc (Zn), and cadmium (Cd), to examine how these metals affect the survival of *D. reticulata*.

Although short-term toxicity tests involve the derivation of median lethal concentrations consider various experimental procedures including quantification of population growth responses and life table demographic variables [10]. It is generally believed that population Dynamics act as a magnifying glass to facilitate detection of small changes in the survival and/or reproduction of a given zooplankton species under stressful conditions [11].

Despite the important role of cladoceran species as the summer plankton in the temperate zone and a common species in tropical regions, few studies have investigated the sensitivity of cladoceran species to toxic chemicals. The sensitivity of *C. reticulata* to toxic chemicals may differ from that of temperate *Daphnia* species. Investigating the impact of heavy metals on *Ceriodaphnia* species may be important for understanding the impact of toxic chemicals on the summer-temperate community with dominance of non *Daphnia* species. I carried out acute toxicity tests of three heavy metals, copper (Cu), zinc (Zn), and cadmium (Cd), to examine how these metals affect the survival of *C. reticulata*.

## MATERIALS AND METHODS

The test species *C. reticulata* was originally isolated from the Hazar Lake (Elazığ/Turkey) and was cultured using the single celled green alga *Chlorella vulgaris*. The medium and food were replaced daily in the stock culture, with food being added at a concentration of approximately  $0.3 \times 10^6$  cells/ml. The stock culture was kept under constant laboratory conditions ( $23 \pm 1$  °C, 16:8 h light:dark) for at least 1 month before the start of the bioassays. Bioassays were conducted for Cu, Zn, and Cd using the following test substances;  $\text{CdCl}_2 \cdot 2\frac{1}{2}\text{H}_2\text{O}$  (CAS No. 10108642),  $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$  (CAS No. 7758998), and  $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$  (CAS No. 773020). A stock solution of each chemical (5.000 mg/L) was prepared by dissolving 50 mg of the chemical in aged tap water to a final volume 10 mL. The tests were conducted under the same laboratory conditions as those for the stock cultures ( $23 \pm 1$  °C, 16:8 h light:dark, pH 7-8, dissolved oxygen (DO) > 4 mg/L, and total hardness 67.5 mg/L). Control and exposure treatments were prepared for each metal by diluting each stock solution of the chemical with aged tap water. Concentrations for Cd exposure were 60, 80, 105, 135, 178, and 230.5 µg/L; for Cu 7, 10, 14, 18.5, 25.5, and 38 µg/L; for Zn, 95, 135.5, 190, 280, and 360.5 µg/L. Two glass beakers containing 30 mL of each solution were prepared for the control and the exposure concentrations for each metal (40 beakers in total). Five neonates or adult females of *Ceriodaphnia reticulata* were introduced into each beaker. Test individuals were not fed during the assays. Based on results of the tests, we determined 24- and 48- h median lethal concentration ( $\text{LC}_{50}$ ) values with 95% confidence intervals (CI) using the Probit method.

## RESULTS AND DISCUSSION

Acute toxicity tests indicated that lead at higher concentrations had a detrimental effect on the survival of *C. reticulata*. In this study, we determined the impacts of Cu, Zn, and Cd on *C. reticulata* survival (Table 1). The  $\text{LC}_{50}$  values of Cu and Zn for *C. reticulata* were lower than those for *Daphnia magna*, and *Leptodora kindtii* one of the global Standard test organisms for toxic chemicals because it is relatively more sensitive to *C. reticulata* is more strongly influenced by Zn, and Cu than

is *D. magna* and *L. kindtii*. In contrast, the  $\text{LC}_{50}$  value of Cd for *C. reticulata* was more than or nearly comparable with that of *D. magna* and *L. kindtii* (Table 1), indicating that *C. reticulata* is less sensitive to Cd than is *D. magna* and *L. kindtii*.

There have been no reports addressing the acute toxicity of heavy metals to *C. reticulata*. These results clearly demonstrated that *C. reticulata* was more vulnerable to Cu and Zn from heavy metals than was *D. magna* and *L. kindtii* (Table 1). In *D. magna* and *L. kindtii*, the  $\text{LC}_{50}$  values for Cu (120 µg/L [12]; 19.69 µg/L [13]) were six times higher than that in *C. reticulata* (19.7 µg/L) (Table 1). Similarly, in *D. magna* and *L. kindtii*  $\text{LC}_{50}$  values for Zn (1800 µg/L [14], 2332 µg/L [13]) were seven and nine times higher than that in *C. reticulata* (258.5 µg/L) (Table 1). Results indicate that *C. reticulata* is more strongly influenced by Zn and Cu than *D. magna* and *L. kindtii*. In contrast, the  $\text{LC}_{50}$  values of Cd for *C. reticulata* was more than or nearly comparable with that of *D. magna* and *L. kindtii* (Table 1), indicating that *C. reticulata* is less sensitive to Cd than is *D. magna* and *L. kindtii*.

The main intake route of the soluble toxicants is through contact with body surface, and thus large cladocerans (subsequent lower to-ratio) tend to be more tolerant to the chemicals than the small ones [15].

Freshwater zooplankton community is composed of cladocerans, copepods, and rotifers. Species sensitivity distributions of cladocerans and copepods to Cd, Cu, and Zn have been reported (Wong *et al.*, 2009). The 5% hazardous concentrations (HC5) of cladocerans are 8.16 µg/L for Cd, 2.72 µg/L for Cu, and 30.8 µg/L for Zn. The HC5 copepods are 19.6 µg/L for Cd, 23.4 µg/L for Cu, and 32.0 µg/L for Zn. These indicate that *C. reticulata* may show moderate sensitivities to Cd, Cu, and Zn in cladoceran species, but *C. reticulata* has a slightly lower sensitivity to Cu than copepod species. If *C. reticulata* is a dominant species in a zooplankton community, relatively low concentrations of Cu may affect the zooplankton community and functions such as the energy and material flow to higher trophic organisms due to the decrease in *C. reticulata*.

Although Zn is an essential heavy metal, it is required only in low concentrations for zooplankton. Thus, the cuticular structure and life span of cladocerans are affected if Zn is not available in sufficient quantity. For example, when the availability of Zn is < 25 µg per daphnid, reduced survival and reproduction has been reported for *D. magna* and *D. pulex* [16]. However, Zn is also toxic to a variety of aquatic organisms including both phyto- and zooplankton [17].

Freshwater zooplankton community is composed of cladocerans, copepods, and rotifers. Species sensitivity distributions of cladocerans and copepods to Cd, Cu,

and Zn have been reported [18]. The 55% hazardous concentrations (HC5) of cladocerans are 8.16 µg/L for Cd, 2.72 µg/L for Cu, and 30.8 µg/L for Zn. The HC5 of copepods are 19.6 µg/L for Cd, 23.4 µg/L for Cu, and 32.0 µg/L for Zn. These indicate that *C. reticulata* may show moderate sensitivities to Cd, Cu, and Zn in cladoceran species.

The presence of heavy metals has been reported in natural lakes. For example, Zn, Cu, and Cd in water of Lake Hazar, Demirköprü Dam Lake, and Avşar Lake [19, 20]. In Demirköprü Dam Lake, the concentration of Cu (0.02 mg/L) was higher than in other in Turkey, such as Hazar Lake, Avşar Lake, but was lower than the LC<sub>50</sub> value for *C. reticulata* as were the concentrations of Cd (0.7 – 1 µg/L) and Zn (25 – 98 µg/L). The *C.*

*reticulata* population may be affected not only by acute effects of heavy metals on individual mortality but also by chronic effects on the demography. Although in this study, I could not examine chronic effects of heavy metals on *Diaphanosoma* population, I could estimate roughly the chronic effects on *Diaphanosoma* population by dividing LC<sub>50</sub> values obtained in my study by acute –to- chronic ratio [21, 22].

When the acute –to- chronic ratio was assumed to be a factor of 10 that is used in risk assessment [21], the estimated no –observed- effect concentrations (NOECs) of Cd, Cu, and Zn were 6.98, 1.04, and 17.41 µg/L, respectively. The estimated NOEC value of Cu was lower than the concentration in Hazar Lake, and Avşar Lake.

**Table 1. Acute toxicities (24- and 48-h lethal concentration (LC50) values, µg/L) and 95% confidence intervals (CI) of cadmium (Cd), copper (Cu), and zinc (Zn) for *C. reticulata*, with values included for *D. magna* and *L. kindtii* as a reference species.**

Species	Metal	24-h LC <sub>50</sub> (95%CI)	48-h LC <sub>50</sub> (95%CI)
<i>C. reticulata</i>	Cd	183.2 (150.4-410.7)	82.1 (51-120.8)
	Cu	18.7 (14.3-22.4)	12.7 (6.3-13.8)
	Zn	258.5 (122.4-331.6)	182.1 (87.4-310.6)
<i>D. magna</i>	Cd	92 <sup>a</sup>	43 <sup>a</sup>
	Cu	120 <sup>b</sup>	24.93 <sup>c</sup>
	Zn	1800 <sup>d</sup>	690 <sup>e</sup>
<i>L. kindtii</i>	Cd	20.14 <sup>f</sup>	Not determined
	Cu	19.69 <sup>f</sup>	Not determined
	Zn	2332 <sup>f</sup>	Not determined

Note: <sup>a</sup>Lewis & Weber [24], <sup>b</sup>Milam CD *et al.* [12], <sup>c</sup>McWilliam and Baird [25], <sup>d</sup>Cairns *et al.* [14], <sup>e</sup>Khangarot *et al.* [23], <sup>f</sup>Sakamoto & Ogamino [13]

## CONCLUSION

*C. reticulata* population may be affected by a relatively lower concentration of Cu in temperate lakes. Further studies will be needed to evaluate the chronic effect of Cu on the *C. reticulata* population.

## REFERENCES

1. Brando C, Bohets HL, Vyer IE, Dierickx PJ; Correlation between the in vivo cytotoxicity to cultured fathead minnow fish cells and fish lethality data for 50 chemicals. *Chemosphere*, 1992; 25; 553 – 562.
2. Hanazato T; Pesticide effects on freshwater zooplankton: an ecological perspective. *Environ Pollut.*, 2001; 112; 1-10.
3. Stark JD, Banks JE; Population-level effects of pesticides and other toxicants on arthropods. *Anno Rev Entomol.*, 2003; 48: 505-519.
4. Sharma RK, Agrawal M; Biological effects of heavy metals: An overview. *J Environ Biol.*, 2005; 26: 301-313.
5. Pascoe D, Shazile NAM; Episodic pollution. A comparison of brief and continuous exposure of rainbow trout to cadmium. *Ecotoxicol Environ Saf.*, 1986; 12: 189-197.
6. Nandini S, Mayeli M, Sarma SSS; Effects of stress on the life table demography of *Moina macrocopa*. *Hydrobiologica*, 2004; 526: 245-254.
7. Park KS, Shin HW; Studies on phyto-and-zooplankton composition and its relation to fish productivity in a west coast fish pond ecosystem. *J Environ Biol.*, 2007; 28: 415-422.
8. Sarma SSS, Nandini S; Review of recent ecotoxicological studies on cladocerans. *J Environ Sci Heal B.*, 2006; 41: 1417-1430.
9. Versteeg DJ, Stalmans M, Dyer SD, Janssen CR; *Ceriodaphnia* and *Daphnia*: A comparison of their sensitivity to xenobiotics and utility as a test species. *Chemosphere*, 1997; 34: 869-892.
10. Calow P; Handbook of ecotoxicology. Blackwell Sci. Publ., London, 1993.
11. Halbach U, Siebert M, Westermayer M, Wissel C; Population Dynamics of rotifers as a bioassay tool for ecotoxicological tests in aquatic environments. *Ecotoxicol Environ Safety*, 1983; 7: 484-513.
12. Milam CD, Farris JL, Hardesty DK; Acute toxicity of six freshwater mussel

- species(Glochidia) to six chemicals: implications for daphnids and *Utterbackia imbecillis* as surrogates for production of freshwater mussels (Unionidae). Arch Environ Contam Toxicol., 2005; 48: 166-173.
13. Sakamoto M, Ogamino Y; *Leptodora kindtii*: a cladoceran species highly sensitive to toxic chemicals. Limnology, 2010; 11: 193-196.
  14. Cairns J Jr., Buikema AL, Heath AG; Parker BC; Effects of temperature on aquatic organisms sensitivity to selected chemicals. In Virginia Water Resources Research Center, Bulletin, Volume 106, Virginia Polytechnic Institute, Blacksburg, 1978: 1-88 .
  15. Vesela S, Vijverberg J; Effects of body size on toxicity of zinc in neonates of four differently sized *Daphnia* species. Aquat Ecol., 2007; 41; 67-73.
  16. Keating KI, Caffrey PB; Selenium deficiency induced by zinc deprivation in a crustacean. Proc Nat Acad Sci USA, 1989; 86: 6436-6440.
  17. Duttagupta S, Gupta S, Gupta A; Euglenoid blooms in the floodplain wetlands of Barak Valley, Assam, North eastern India. J Environ Biol., 2004; 25: 369-373.
  18. Wong LC, Kwok KWH, Leung KMY, Wong CK; Relative sensitivity distribution of freshwater planktonic crustaceans to trace metals. Hum Ecol Risk Assess., 2009; 15: 1335-1345.
  19. Alp MT, Şen B, Özbay Ö; Heavy metal levels in *Cladophora glomerata* which seasonally occurrence in the Lake of Hazar. Ekoloji, 2011; 20(78): 13-17.
  20. Öztürk M, Özözen G, Minareci O, Minareci E; Determination of heavy metals in fish water and sediments of Avşar Dam Lake in Turkey. Iran J Environ Health Sci Eng., 2009; 6(2): 73-80.
  21. Okkerman PC, Van der Plassche EJ, Slooff W, Van Leeuwen CJ, Canton H; Ecotoxicological effects assessment: a comparison of several extrapolation procedures. Ecotoxicol Environ Saf., 1991; 21: 182-193.
  22. Roex EWM, Van Gestel CAM, Van Wezel AP, Van Straalen NM; Ratios between acute aquatic toxicity and effects on population growth rates in relation to toxicant mode of action. Environ Toxicol Chem., 2000; 19: 685-693.
  23. Khangarot BS, Ray PK, Chandra H; *Daphnia magna* as a model to assess heavy metal toxicity: comparative assessment with Mouse system. Acta Hydrochim. Hydrobiol., 1987; 101: 143-178.
  24. Lewis PA, Weber CI; A study of the reliability of *Daphnia* acute toxicity tests. In Cardwell RD, Purdy R, Bahner RC editors; Aquatic toxicology and hazard assessment: seventh symposium, ASTM STP 854, Philadelphia, 1985: 73-86.
  25. McWilliam RA, Baird DJ; Postexposure feeding depression: a new toxicity endpoint for use in laboratory studies with *Daphnia magna*. Environ Toxicol Chem., 2002; 21: 1198-1205.