

International Journal of Zoological Investigations

Contents available at Journals Home Page: www.ijzi.net



Acute Toxicity of Chromium Chloride to the Freshwater Catfish, *Heteropneustes fossilis*

Ajai Kumar Srivastav*, Khushbu Agarwal, ManiRam Prasad and Abhishek Kumar

Department of Zoology, D.D.U. Gorakhpur University, Gorakhpur, 273009, India

*Corresponding author

Abstract: Four-day static renewal acute toxicity test was used for *Heteropneustes fossilis* to determine the LC₅₀ values for chromium chloride. Six replicates each containing ten fish (kept in glass aquarium containing 30 L of the test solution) were subjected to chromium chloride (50, 75, 100, 125, 150, 200 and 250 mg/L). The per cent mortality after exposure to various concentrations of chromium chloride to *H. fossilis* for 24, 48, 72 and 96 h has been observed and the LC₅₀ values at different exposure periods as well as their upper and lower confidence limits and slope functions were calculated by PoloPlus software version 2.0 Computer programme. The LC₅₀ values of chromium chloride for *H. fossilis* at 24, 48, 72, and 96 h were calculated as 138.50 mg/L; 119.38 mg/L; 92.77 mg/L and 63.26 mg/L, respectively.

Keywords: *Heteropneustes fossilis*, LC₅₀, Chromium, Catfish

Introduction

Fishes are the most important non-target inhabitants of the aquatic ecosystem which are affected by toxicants (Scott and Sloman, 2004; Kiaune and Singhasemanon, 2011; Chaudhary *et al.*, 2015; Prasad *et al.*, 2015). Fishes influence the human beings in various ways and are economically very important. Their food value is now well known as it provides the much needed protein, vitamin A and D, and other elements.

Chromium is the sixth most abundant heavy metal in the earth crust (U. S. Environmental Protection Agency, 1984; Velma and Tchounwou, 2010). Chromium

finds its way into the aquatic ecosystem through some industries mainly electroplating, polishing, paint, rubber, plastic, ceramics, fiberglass, chrome plating, chrome alloy making, welding and foundries. Chromium is fairly toxic to animals and human beings beyond its optimum concentration. In such instances chromium produced cytotoxic, haematological, histological, immunological and genotoxic effects to fish (Sastry and Sunita, 1982; Zhu *et al.*, 2004; Vutukuru, 2005; Prabakaran *et al.*, 2007; Bozcaarmutlu and Arinc, 2007; Goodale *et al.*, 2008; Tan *et al.*, 2008; Velma *et al.*, 2009; Palaniappan and

Karthikeyan, 2009; Velma and Tchounwou, 2010; Muthukumaravel and Rajaraman, 2013).

Earlier, the toxic effects of heavy metals on fish are not well studied. Hence, the present study was designed to investigate the toxic effect of heavy metal – chromium chloride on a freshwater catfish *H. fossilis*, in terms of fish mortality test.

Material and Methods

(A) Collection and handling of the fish:

Stinging catfish, *Heteropneustes fossilis* were used as test fish in this study because this species is hardy, readily available, easy to handle, can be held in captivity for long period, and form an important species in many water resources.

Adult *H. fossilis* (both sexes, body weight 30-35 g) were collected locally and inspected for external signs of injury and diseases. Those, which showed such symptoms, were discarded and only the healthy ones were selected for experiments. These fish were acclimatized to the laboratory conditions (under natural photoperiod and temperature 28.4 ± 1.2 C) for two weeks in plastic pools containing 500 L of dechlorinated tap water. During acclimatization the fish were fed daily with wheat flour pellets and ground dried shrimps, 2-3 times per day. Water was renewed daily after cleaning the fecal matter and leftover food. All care was taken to avoid giving stress to the fish. The fish were not fed 24 h before and during the experimental period so that excretory substances may not influence the toxicity of test solutions. The mortality rate during acclimatization was less than 4%.

(B) Determination of LC_{50}

Four-day static renewal acute toxicity test (APHA *et al.*, 1998) was used for the determination of LC_{50} values for chromium.

Six replicates each containing ten fish (kept in glass aquarium containing 30 L of the test solution) were subjected to chromium chloride (50, 75, 100, 125, 150, 200 and 250 mg/L). Chromium chloride was firstly dissolved in distilled water and then the desired volume of the solution was mixed in tap water to obtain the above mentioned toxicant concentration. A control group with six replicates (each containing 10 fish) kept in 30 L tap water was also run. The solutions of all the aquaria (control and experimental) were renewed daily. Precautions were taken to remove the dead fish immediately because dead fish deplete dissolved oxygen which greatly affects toxicity data (Schreck and Brouha, 1975). Death in fish was confirmed when the movement of the operculum was stopped and the fish remain unresponsive when gently prodded at the caudal peduncle. The concentration of the toxicants and their degradation products were not measured during the course of bioassay as the facilities for it were not available in the department.

The LC_{50} values at different exposure periods as well as their upper and lower confidence limits and slope functions were calculated by PoloPlus software version 2.0 Computer programme.

Results

The per cent mortality after exposure to various concentrations of chromium chloride to *H. fossilis* for 24, 48, 72 and 96 h has been depicted in Fig. 1 and the LC_{50} values are depicted in Table 1.

The LC_{50} values of chromium chloride for *H. fossilis* at 24, 48, 72, and 96 h are 138.50 mg/L; 119.38 mg/L; 92.77 mg/L and 63.26 mg/L, respectively. Table 1 shows the LC_{50} values, their upper and lower confidence limits and slope functions for *H. fossilis*.

Table 1: LC₅₀ value, slope function and confidence limits after exposure of chromium chloride at different intervals for the fish *H. fossilis*

Exposure Periods	Effective dose (mg/L)	Limits(mg/L)*		Slope Function	't' ratio	Hetero-geneity
		LCL	UCL			
24 h	LC ₁₀ =100.85	92.13	107.83	9.302	10.086	0.817
	LC ₅₀ =138.50	131.77	145.78	±		
	LC ₉₀ =190.21	177.13	209.71	0.922		
48 h	LC ₁₀ =70.71	32.93	90.61	5.635	9.741	4.274
	LC ₅₀ =119.38	94.34	157.39	±		
	LC ₉₀ =201.54	154.12	479.39	0.578		
72 h	LC ₁₀ =47.02	15.79	65.81	4.343	9.197	3.645
	LC ₅₀ =92.77	66.63	119.39	±		
	LC ₉₀ =183.02	136.67	445.45	0.472		
96 h	LC ₁₀ =40.71	24.44	50.63	6.695	8.441	1.4006
	LC ₅₀ =63.26	50.98	72.97	±		
	LC ₉₀ =98.29	84.12	132.94	0.793		

*The upper and lower confidence limits for LC₅₀ values calculated at 0.05 levels.

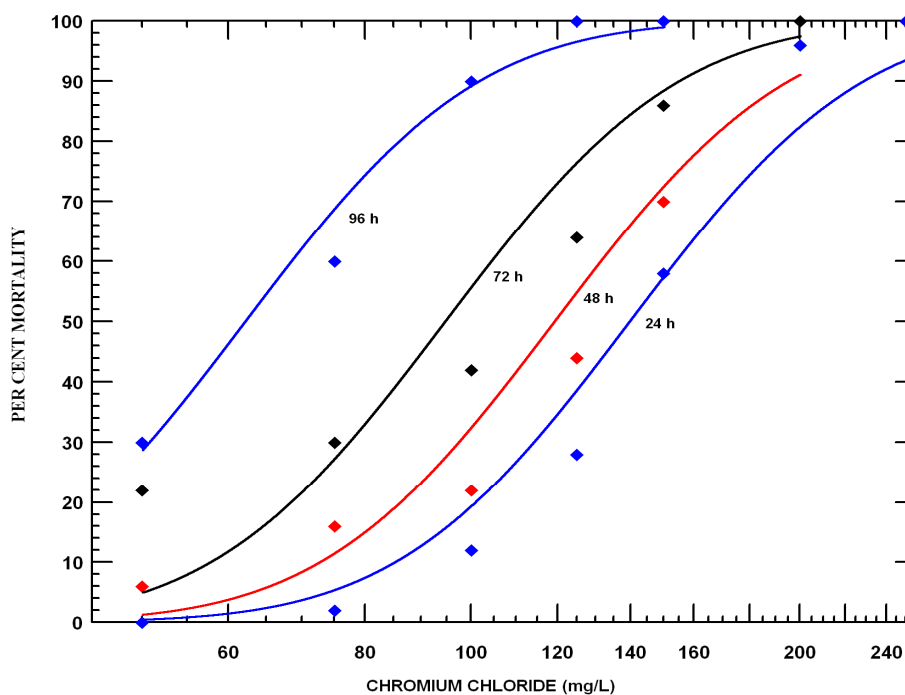


Fig. 1: Per cent mortality of the fish *Heteropneustes fossilis* after 24, 48, 72, and 96 h exposure to different concentrations of chromium chloride.

Discussion

The LC₅₀ values for *H. fossilis* for chromium chloride at 24, 48, 72 and 96 h are 138.50, 119.38, 92.77 and 63.26 mg/L, respectively. 96 h LC₅₀ values for chromium for fathead minnow (*Pimephales promelas*) and goldfish (*Carassius auratus*) have been reported as 48 and 120 mg/L, respectively (Adelman and Smith, 1976; Adelman *et al.*, 1976). For rainbow trout 24 h TLm value for K₂Cr₂O₄ has been estimated as 100 mg/L (Schiffman and Fromm, 1959). Pickering and Henderson (1966) have determined 24, 48 and 96 h TLm values for chromium salts for fathead, bluegills, goldfish and guppies and reported that for these fishes 96 h LC₅₀ value ranged from 3.33 mg/L to 118.0 mg/L in soft water and from 67.0 to 133 mg/L in hard water. Saxena *et al.* (1980) have reported 96 h LC₅₀ for the fish *Channa punctatus* as 45.2 ppm chromium at pH 7.5. According to Kaviraj and Konar (1982) 96 h LC₅₀ for chromium for *Tilapia mossambica* at pH 6.0, 7.0 and 8.0 were 179.0, 170.0 and 217.5 ppm, respectively.

Al-Kahem (1995) has reported 96 h LC₅₀ value for trivalent chromium as 23.7 mg/L for *Oreochromis niloticus*. 96 h LC₅₀ value for trivalent and hexavalent chromium has been reported as 53 and 24 mg/L, respectively for juvenile mullet (Negilsck, 1976). Al-Akel and Shamsi (1996) estimated the 96 h LC₅₀ for hexavalent chromium for *Cyprinus carpio* as 93.6 mg/L. Issac Arunkumar *et al.* (2000) have reported 96 h LD₅₀ for hexavalent and trivalent chromium as 75 and 1000 µg for African mouth breeder *Oreochromis mossambicus*. Vutukuru (2005) reported 96 h LC₅₀ value as 111.45 mg/L for Indian major carp *Labeo rohita*. In another study Vutukuru *et al.* (2007) found 96 h LC₅₀ for chromium as 61 mg/L for *Labeo rohita*. For *Channa punctatus* 96 h LC₅₀ value for chromium trioxide was determined as 64.2 mg/L (Chaudhary *et al.*,

2012). The 96 h LC₅₀ value for potassium dichromate was determined as 61.80 mg/L for *Channa punctatus* (Kumar *et al.*, 2012). Abedi *et al.* (2012) reported 96 h LC₅₀ as 17.05 and 7.46 mg/L for common carp (*Cyprinus carpio*) and Sutchi catfish (*Pangasius hypophthalmus*), respectively. Oliveira- Fitho *et al.* (2013) reported 96 h LC₅₀ for chromium as 123.1 mg/L for *Danio rerio*, 93.3 mg/L for *Hyphessobrycon eques* and 107.2 mg/L for *Oreochromis niloticus*. Shaukat and Javed (2013) estimated 96 h LC₅₀ for chromium for three fish species at various age groups- 60 day, 90 day and 120 day. These authors reported LC₅₀ value for 96 h as 53.57 mg/L, 74.56 mg/L and 96.86 mg/L for *Ctenopharyngodon idella* for 60, 90 and 120 days age, respectively. Further, they reported 87.93 mg/L, 102.87 mg/L and 128.09 mg/L as 96 h LC₅₀ value for *Cyprinus carpio* for 60, 90 and 120 days age, respectively. For *Tilapia nilotica* the values were 119.52, 139.29 and 164.36 mg/L at 60, 90 and 120 day age, respectively. They have concluded that 60 day fish were most sensitive to chromium than 90 and 120 day fish (Shaukat and Javed, 2013).

References

- Abedi Z, Khaleshi M, Eskandari SK and Rahmani H (2012) Comparison of lethal concentrations (LC₅₀-96 H) of CdCl₂, CrCl₃ and Pb (NO₃)₂ in common carp (*Cyprinus carpio*) and Sutchi catfish (*Pangasius hypophthalmus*) Iranian J. Toxicol. 6: 672-680.
- Adelman IR and Smith Jr LL (1976) Fathead minnows (*pimephales promelas*) and gold fish (*Carassius auratus*) as standered fish in bioassays and their reaction to potential reference toxicants. J. Fish Res. Board Can. 33: 209-214.
- Adelman IR, Smith Jr LL and Siesennop GD (1976) Acute toxicity of sodium chloride pentachlorophenol, guthian (R) and hexavalent chromium to fathead minnows (*Pimephales promelas*) and goldfish (*Carassius auratus*), J. Fish. Res. Bd. Can. 33: 203-208.

- Al-Akel AS and Shamsi MJK (1996) Hexavalent Cr toxicity and impact on carbohydrate metabolism and haematological parameters of carp (*Cyprinus carpio*) from Saudi Arabia. *Aquat. Sci.* 58: 24-30.
- Al-Kahem HF (1995) Behavioural responses and changes in some haematological parameters of the cichlid fish, *Oreochromis niloticus*, exposed to trivalent chromium. *JKAU: Sci.* 7: 5-13.
- APHA, AWWA and WPCF (1998) Standard methods for the examination of water and wastewater. 15th edition, APHA, Washington, DC.
- Bozcaarmutlu A and Arinc E (2007) Effect of mercury, cadmium, nickel, chromium and zinc on kinetic properties of NADPH-cytochrome P450 reductase purified from leaping mullet (*Liza saliens*), *Toxicol. In Vitro* 21: 408-416.
- Chaudhary J, Abha and Jha AM (2012) Cytogenetic effect of chromium trioxide in an air breathing teleost *Channa punctatus* (Bloch). *Int. J. Phar. Biol. Sci.* 2: 246-253.
- Chaudhary A, Prakash C and Srivastav SK (2015) Biochemical changes in blood of freshwater catfish *Heteropneustes fossilis* exposed to microcystin-LR. *Int. J. Zool. Invest.* 1: 72-76.
- Goodale BC, Walter R, Pelsue SR, Thompson WD, Wise SS, Winn RN, Mitani H and Wise Sr JP (2008) The cytotoxicity and genotoxicity of hexavalent chromium in medaka (*Oryzias latipes*) cells. *Aqua. Toxicol.* 87: 60-67.
- Issac Arunkumar R, Rajasekaran P and Michael RD (2000) Differential effect of chromium compounds on the immune response of the African mouth breeder *Oreochromis mossambicus* (Peters). *Fish & Shellfish Immunology.* 10: 667-676.
- Kaviraj A and Konar SK (1982) Acute toxicity of mercury, chromium and cadmium to fish, plankton and worm. *Geobios.* 9: 97-100.
- Kiaune L and Singhasemanon N (2011) Pesticidal copper (I) oxide: environmental fate and aquatic toxicity. *Rev. Environ. Contam. Toxicol.* 213: 1-26.
- Kumar P, Kumar R, SahebraoNagpure N, Nautiyal P, Dabas A, Kushwaha B and Lakra WS (2012) Genotoxic and mutagenic assessment of hexavalent chromium in fish following in vivo chronic exposure. *Human Ecol. Risk Asses.: Int. J.* 18: 855-870.
- Muthukumaravel K and Rajaraman P (2013) A study on the toxicity of chromium on the histology of gill and liver of freshwater fish *Labeo rohita*. *J. Pure Appl. Zool.* 1: 122-126.
- Negilski DS (1976) Acute toxicity of zinc, cadmium and chromium to the marine fishes, yellow-eye mullet (*Aldrichetta forsteri* C. & V.) and small-mouthed hardyhead (*Atherinasoma microstoma* Whitley). *Aus. J. Marine Freshwat. Res.* 27: 137-149.
- Olivieira-Filho EC, Muniz DHF, Freire IS, Aquino FG and Andrade LRM (2013) Comparative susceptibility of freshwater fish species to metals from ultramafic soils. *Ecotoxicol. Environ. Chem.* 8: 119-123.
- Prabakaran M, Binuramesh C, Steinhagen D and Michael RD (2007) Immune response in the tilapia, *Oreochromis mossambicus* on exposure to tannery effluent. *Ecotoxicol. Environ. Saf.* 68: 372-378.
- Prasad M, Kumar A, Suzuki N and Srivastav AK (2015) Botanical Pesticide *Nerium indicum* Alters Prolactin Cells of Stinging Catfish, *Heteropneustes fossilis*. *Int. J. Zool. Invest.* 1: 77-84.
- Pickering QH and Handerson C (1966) The acute toxicity of some heavy metals to different species of warmwater fishes. *Air Water Poll. Int. J.* 10: 453-463.
- Palaniappan PL and Karthikeyan S (2009) Bioaccumulation and depuration of chromium in the selected organs and whole body tissues of freshwater fish *Cirrhinus mrigala* individually and in binary solutions with nickel. *J. Environ. Sci. (China).* 21: 229-236.
- Sastry KV and Sunita KM (1982) Effect of cadmium and chromium on the intestinal absorption of glucose I the snakehead fish, *Channa punctatus*. *Toxicol. Lett.* 10: 293-296.
- Saxena OP Parashari A and yadav RS (1980) Toxicity of few heavy metals to freshwater fish *Channa punctatus* (Bl.). *J. Ichthyol.* 1: 37-40.
- Schiffman RH and Fromm PO (1959) Chromium induced changes in the blood of rainbow trout, *Salmo giardneri*. *Sewage Ind. Wastes.* 31: 205-211.
- Schreck CB and Brouha P (1975) Dissolved oxygen depletion in static bioassay system. *Bull. Environ. Contam. Toxicol.* 14: 149-152.

- Scott GR and Sloman KA (2004) The effects of environmental pollutants on complex fish behaviour: integrating behavioural and physiological indicators of toxicity. 68: 369-92.
- Shaukat T and Javed M (2013) Acute toxicity of chromium for *Ctenopharngodon idella*, *Cyprinus carpio* and *Tilapia nilotica*. Int. J. Agric. Biol. 15: 590-594.
- Tan F, Wang M, Wang W and Lu Y (2008) Comparative evaluation of the cytotoxicity sevsitivity of six fish cell lines to four heavy metals in vitro. Toxicol. In. Vitro. 22: 164-170.
- Velma V and Tchounwou PB (2010) Chromium-induced biochemical, genotoxic and histopathoogic effects in liver and kidney of goldfish, *Carassius auratus*. Mut. Res. 698: 43-51.
- Velma V, Vutukuru SS and Thounwou PB (2009). Ecotoxicology of hexavalant chromium in freshwater fish: A critical review. Ren. Environ. Health. 24: 129-145.
- Vutukuru SS (2005) Acute effects of hexavalent Cr on survival, oxygen consumption, haematological parameters and some biochemical profile of the Indian major carp, *Labeo rohita*. Int. J. Environ. Res. Pub. Health. 2: 456-462.
- Vutukuru SS, Prabhath NA, Raghavender M and Yerramilli A (2007) Effect of arsenic and chromium on the serum amino-transferases activity in Indian major carp, *Labeo rohita*. Int. J. Environ. Res. Pub. Health. 4: 224-227.
- Zhu Y, Wang Y and Zhang R (2004) Cadium, chromium and copper induced polychromatocyte micronuclei in Carp (*Cyprinus carpio L*). Comp. Anim. Physiol. 72: 78-86.