



## RESEARCH ARTICLE

### Acute Toxicity of Aluminium to the Fish (*Catla catla*, *Labeo rohita* and *Cirrhina mrigala*)

Hamda Azmat\*, Muhammad Javed and Ghazala Jabeen

Department of Zoology & Fisheries, University of Agriculture, Faisalabad, Pakistan

\*Corresponding author: hamda.azmat@gmail.com

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#### ABSTRACT

Acute toxicity tests (96-hr LC<sub>50</sub> and lethal concentration) of aluminium (Al) were conducted with three fish species viz. *Catla catla*, *Labeo rohita* and *Cirrhina mrigala* of 60, 120 and 240 days age groups at constant water temperature (30°C), pH (7.50) and total hardness (300 mg.L<sup>-1</sup>) in the wet laboratory. At termination of each trial, the fish were dissected and their organs viz. bones, gills, gut, intestine, kidney, liver, scales, skin, muscles and fats isolated for the determination of Al concentrations. At 60 days, all the three fish species showed significantly (P<0.05) higher sensitivity to Al while 240 days fish were significantly least sensitive. Among the three fish species, *Catla catla* were significantly (P<0.05) more sensitivity to Al with the mean 96-hr LC<sub>50</sub> and lethal concentration of 81.68±28.54 and 129.81±30.95 mg.L<sup>-1</sup>, respectively. Fish organs showed significantly variable ability to concentrate metal during acute exposure of Al. However, liver and kidneys exhibited significantly higher potentials for metals accumulation. From the study it was concluded that all the three fish species responded differently towards Al toxicity.

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#### INTRODUCTION

Metals are the common pollutants of the rivers in Punjab province entering them with industrial and municipal waste waters. The heavy loads of metals in the rivers have adversely affected the indigenous fish fauna, including major carps viz. *Catla catla*, *Labeo rohita* and *Cirrhina mrigala* that are on the verge of extinction in the aquatic habitats (Rauf *et al.*, 2009a; 2009b; Azmat and Javed, 2011). Therefore, heavy metals are considered as chief environmental pollutants and have long been known as staid contaminants of aquatic environments (Javed, 2004). Metals contamination of the environment results both from natural sources and industrial activities besides an additional contribution from air (Vutukuru, 2003). Heavy metals can cause severe destruction in the physiological functions of aquatic animals when present in elevated concentrations (Javed, 2003) directly due to their amassing in body (Waqar, 2006) and transferring them to the trophic level further up in the food chain (Chen and Folt, 2000). One of the most serious impacts of metals persistence in aquatic environment is their biological magnification through the food chain (Ubaidullah *et al.*, 2004). Therefore, heavy metals enrichment of recipient

waters may cause adverse effects on the sustainability of the ecosystem (Farombi *et al.*, 2007).

Acute toxicity bioassays (LC<sub>50</sub> and lethal concentration) are used to evaluate the toxicity of heavy metals and to assess the potentials of various fish species to the toxicity of metals (Abdullah *et al.*, 2007). Acute toxicity tests permit rapid assessment of the impacts of various toxicants on organisms. The criterion of lethal toxicity is mortality, the final response of an organism. However, in natural waters, fish are mostly affected by long-term influence of low concentrations of pollutants or their mixtures. Therefore, chronic effects of sub-lethal toxicity of various metals are applied. The studies on metallic toxicity make possible to assess the sub-lethal (1/3 LC<sub>50</sub>) effects on physiology, growth, biology and behavior of various organisms, to predict probable toxic effects and their consequences on adaptive abilities of various animals including fish (Pane *et al.*, 2004). Aluminium is found as its oxides and silicates on the Earth (Scancar *et al.*, 2004). The solubility of Al escalated linearly with the drop in water pH causing escalated concentrations of inorganic Al. Inorganic form of Al has been reported the most injurious to various fish species (Camargo *et al.*, 2009). Therefore, present work was conducted to determine the acute toxicity of Al to the three indigenous

fish species along with their growth responses under sub-lethal exposures under static bioassay.

## MATERIALS AND METHODS

Fingerlings of three fish species viz. *Catla catla*, *Labeo rohita* and *Cirrhina mrigala* were obtained from the Fish Seed Hatchery, Faisalabad and kept in holding tanks, supplied with flow through aerated water and acclimated for 48 hours, in the laboratory before conducting toxicity tests.

**Metals acute toxicity assays:** Laboratory tests were conducted to determine the acute toxicity of Al in terms of 96-hr LC<sub>50</sub> and lethal concentrations to three age groups (60, 120 and 240 days) of *Catla catla*, *Labeo rohita* and *Cirrhina mrigala* at constant water temperature (30°C), pH (7.50) and total hardness (300 mg.L<sup>-1</sup>). These tests were performed in glass aquaria containing 50-liter water. Ten fish of each age group and species were tested against various concentrations of Al with three replications for each test dose. Extra pure compound of Al (Al(NO<sub>3</sub>)<sub>3</sub>.9H<sub>2</sub>O) was dissolved in deionized water and stock solution prepared for required metal concentrations. The exposure concentration of Al for each fish species was started from zero with an increment of 0.05 and 0.50 mg.L<sup>-1</sup> (as total concentration) for low and high concentrations, respectively.

**Tissue distribution assays:** After acute toxicity trials, the dead fish were replaced from the medium, weighed after being lightly blotted dry and dissected. The fish body organs viz. bones, gills, gut, intestine, kidneys, liver, scales, skin, muscles and fats were isolated and analyzed for Al concentrations by following the methods (SMEWW, 1989).

Mean values for 96-hr LC<sub>50</sub> and lethal concentrations were determined by using three replications for each treatment/test dose with 95% confidence intervals. MINITAB computer program based on Probit Static Bioassay test system was used to statistically analyze the fish mortality data. Analysis of Variance and Tuckey's Student Newman-Keul test (Steel *et al.*, 1996) were used to find-out statistical differences among various variables under study.

## RESULTS

The 96-hr tests (both LC<sub>50</sub> and lethal) were performed to evaluate the sensitivity of three fish species viz. *Catla catla*, *Labeo rohita* and *Cirrhina mrigala* to short term effects of Al. Among the three fish age groups, at 240-day, all the three fish species showed significantly least sensitivity to Al while 60-day fish were significantly more sensitive. Among the three fish species, *Catla catla* were

significantly (P<0.05) more sensitive to Al, followed by that of *Labeo rohita* and *Cirrhina mrigala* for both 96-hr LC<sub>50</sub> and lethal concentrations (Table 1).

Aluminium exposure at 96-hr LC<sub>50</sub> caused significant amassing of Al in all the measured tissues of three fish species (Table 2). Among the three fish species, *Catla catla* and *Cirrhina mrigala* showed maximum accumulation of Al in their liver followed by that of kidney and gills where as both the fish species showed least tendency of accumulation in their fats. *Labeo rohita* showed maximum accumulation of Al in its kidney followed by that of liver, however, fats showed least accumulation of Al. The exposure of Al at lethal concentrations to the three fish species caused significantly higher accumulation of Al in the organs of all the three fish species. *Catla catla* showed highest tendency of accumulation of Al in its liver (128.25±41.98 µg.g<sup>-1</sup>) followed by that of kidney (125.74±52.96 µg.g<sup>-1</sup>) and gills (102.56±39.08 µg.g<sup>-1</sup>) with statistically significant differences. Same patterns of accumulation was shown by *Cirrhina mrigala* with the mean values of 176.54±35.35, 163.55±34.48 and 102.88±2.98 µg.g<sup>-1</sup> for liver, kidney and gills, respectively. In case of *Labeo rohita*, for lethal concentration trials, it showed maximum accumulation in its kidney followed by that of liver and gills with the mean values of 152.17±39.32, 122.20±78.06 and 83.28±41.97 µg.g<sup>-1</sup>, respectively. All the three fish species showed least accumulation of Al in their fats (Table 2).

## DISCUSSION

Among the three fish age groups, 240-day all the three fish species showed significantly least sensitivity to Al while 60-day fish were significantly more sensitive. Acute exposure of Al caused significant sensitivity to all the three age groups of fish predicting age related tolerance in three fish species (Kazlauskiene and Stastinaite, 1999). Javed and Abdullah (2006) reported acute toxicity of iron and nickel to *Catla catla*, *Labeo rohita* and *Cirrhina mrigala* that varied significantly with age also. Among the three fish species, *Catla catla* were significantly more sensitive to Al, followed by that of *Labeo rohita* and *Cirrhina mrigala* for both 96-hr LC<sub>50</sub> and lethal concentrations. Javid *et al.* (2007) observed higher sensitivity of *Catla catla* against nickel than that of *Labeo rohita* and *Cirrhina mrigala*. However, Abdullah *et al.* (2007) reported *Labeo rohita* as the least sensitive species to manganese toxicity while it was more sensitive to nickel. Acute exposure of Al caused significantly higher amassing of metal in fish kidney and liver. The extent of metal accumulation differed significantly in various organs of fish that may basically be attributed to their different physiological functions (Karuppasamy, 2004). Ahmed and Bibi (2010) reported significantly

**Table 1:** Responses of three fish species for their 96-hr LC<sub>50</sub> and lethal concentrations (mg.L<sup>-1</sup>) of aluminium

96-hr	Age groups*	Fish Species		
		<i>Catla catla</i>	<i>Labeo rohita</i>	<i>Cirrhina mrigala</i>
LC <sub>50</sub>	1	59.56±0.53c	78.32±0.60 b	91.77±0.42 a
	2	71.58±0.61c	97.49 ±0.57 b	114.17±0.38 a
	3	113.89± 0.60c	140.58±0.59 b	149.14±0.60 a
Lethal concentration	1	104.58±0.56c	115.84±0.40 b	137.28±0.43 a
	2	120.50±0.26c	137.32±0.35 b	177.45±0.41 a
	3	164.34±0.59c	182.74±0.40 b	221.82±0.29 a

\*Age groups (1=60-day; 2=120 day; 3=240-day). Means with the same letters in a single row are statistically similar at P<0.05.

**Table 2:** Accumulation of aluminium ( $\mu\text{g g}^{-1}$ ) in fish body organs during 96-hr acute toxicity trials

Organs	LC <sub>50</sub>			Lethal concentration		
	<i>Catla catla</i>	<i>Labeo rohita</i>	<i>Cirrhina mrigala</i>	<i>Catla catla</i>	<i>Labeo rohita</i>	<i>Cirrhina mrigala</i>
Bones	10.77±4.19g	9.69±4.73h	11.87±1.93g	17.06±7.10h	15.63±5.69h	14.43±2.35h
Gills	56.54±23.32c	5.3.03±22.24c	68.60±10.72c	102.56±39.08c	83.28±41.97c	102.88±2.98c
Gut	31.49±20.67d	34.79±9.98e	15.72±5.32f	70.06±23.37d	49.23±9.15e	21.06±61.43g
Intestine	31.12±18.74d	35.88±9.53d	16.34±3.98f	56.85±35.83e	60.24±15.64d	24.22±2.81f
Kidney	75.25±20.11b	92.20±23.89a	97.85±22.36b	125.74±52.96b	152.17±39.32a	163.55±34.48b
Liver	78.41±26.78a	75.85±44.49b	106.12±24.16a	128.25±41.98a	122.20±78.06b	176.54±35.35a
Scales	15.01±3.73f	13.42±9.65g	17.88±3.37e	24.62±8.67g	17.99±11.94g	30.34±7.12e
Skin	26.38±8.20e	26.15±16.38f	30.38±11.35d	38.39±12.14f	34.11±16.36f	36.74±13.74d
Muscle	4.65±1.38h	3.14±0.45i	3.06±0.63h	6.66±1.38i	4.78±1.37i	8.01±0.34j
Fats	1.88±0.41i	1.58±0.44j	2.12±0.44i	2.89±0.78j	2.90±0.12j	3.53±1.03j

Means with same letters in a single row are statistically similar at  $P < 0.05$ .

higher lead in the liver and intestine of *Catla catla*. The present results clearly reveal liver as the most important site for metal accumulation (Murugan *et al.*, 2008).

Al induced DNA damage in fish has been reported to be dose dependent as cells treated with aluminium exhibited decrease in their capacity predicting Al inhibited DNA repair in Al treated cells (Lankoff *et al.*, 2006). Exposure of fish to Al has also been reported to promote iron induced reactive oxygen species and lipid peroxidation causing reduced metabolism resulted in decreased fish growth (Sarnowski, 2003). Al accumulation was significantly higher in liver, followed by that in kidney of all the three fish species. Metals can enter the fish through skin, gills, oral intake of water, food and non-food particles. After absorption, metals are transported via blood stream to either a storage organ i.e. liver for their transformation and/or bio-accumulation in various organs of fish (Nussey *et al.*, 2000).

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