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## Growth Responses of *Catla catla*, *Labeo rohita* and *Cirrhina mrigala* for Bio-accumulation of Zinc During Chronic Exposure

Muhammad Javed

Department of Zoology and Fisheries, Fisheries Research Farms,  
University of Agriculture, Faisalabad, Pakistan

**Abstract:** In the present research, laboratory studies were conducted with nine life stages of three fish species viz. *Catla catla*, *Labeo rohita* and *Cirrhina mrigala* to determine their feed intake, weight gains and feed conversion ratios under sub-lethal chronic exposure of zinc toxicity. Zinc bioaccumulations in fish organs viz., gills, kidney, liver, skin, muscle and scales during chronic toxicity exposures, have also been determined. All the three fish species showed uniformity in their response towards feed intake, weight increment and feed conversion ratios under sub-lethal zinc chronic exposures. Feed intake by all the three fish species increased significantly with metal exposure time. However, this significant increase in feed intake did not result in fish weight escalation and hence resulted in low feed conversion ratios. Sensitivity of the fish to zinc decreased with fish age also. Different fish age groups showed statistical differences for the accumulation of zinc in their bodies. However, the patterns of these accumulations varied significantly among fish organs. Fish liver and kidney exhibited significantly higher tendency for the accumulation of metals.

**Key words:** Fish growth, bio-accumulation, zinc, *Catla catla*, *Labeo rohita*, *Cirrhina mrigala*

### INTRODUCTION

Carp and other cyprinids contribute significantly towards total global aquaculture production. Out of the total aquaculture production of 33.31 million tons (excluding aquatic plants), carp and other cyprinids formed 14.9 million tons i.e., 44.70% of the total production. Considering the freshwater fish production through farming alone (i.e., 18.5 million tons) it formed 80.10%<sup>[1]</sup>. Tremendously increasing population and establishment of industries in the urban areas of Punjab have resulted in the discharge of heavy metals and their compounds through industrial effluents and domestic sewage into the rivers. Heavy metals contamination usually showed depletion in food utilization in fish<sup>[2]</sup> suggested that any such disturbance could result in reduced fish metabolic rate and hence reduced their growth<sup>[3]</sup>. Therefore, nutritional status, fish size and growth rate are considered when comparing whole-body as well as tissue heavy metal concentration for bio-monitoring and risk assessment are considered. Despite growing interest in the fate of metals in the environment, relatively a little is known about their mechanistic actions of toxicity, particularly in aquatic ecosystems and zinc received little research attention addressing factors influencing its toxicity to freshwater fish. All this necessitates the importance to investigate the growth responses of major carps viz., *Catla catla*,

*Labeo rohita* and *Cirrhina mrigala* and bio-accumulation of zinc during chronic exposure to determine their tolerance against metal toxicity.

### MATERIALS AND METHODS

Fingerling *Catla catla*, *Labeo rohita* and *Cirrhina mrigala* of nine age groups i.e., 90 to 330 day were taken for this experiment. All the three fish species were reared in glass aquarium, separately, for 30-day interval under sub-lethal zinc concentrations. The parameters viz., feed intake, increase in average weight and feed conversion ratios of three fish species were monitored. The fish were kept under laboratory conditions in 500 L tanks for two weeks prior to the experiment. The fish in good condition and good state of health were acclimatized and then *Catla catla*, *Labeo rohita* and *Cirrhina mrigala* exposed to zinc ( $Zn\ Cl_2$ ) sub-lethal concentrations of 10.41, 13.33 and 16.67  $mg\ L^{-1}$ , respectively<sup>[4]</sup> in tap water, using a static water system with continuous aeration, under controlled laboratory conditions at room temperature. The exposure medium were continuously replenished and partly exchanged to maintain the above mentioned sub-lethal concentrations of zinc for three fish species throughout the experimental period of 30 days for each trial. The concentrations of zinc in the test medium were measured by Atomic Absorption Spectrophotometry through methods of SMEWW<sup>[5]</sup>. During each trial, fish were fed to

satiation daily at 9.00 and 17.00 h with the feed having Digestible Energy (DE) of 2.90 Kcal g<sup>-1</sup> and 35% Digestible Protein (DP) with the following composition:

Ingredient	(%)	Mineral and amino acids	(%)
Fish meal	40.00	Lysine	1.9365
Corn gluten (30%)	39.27	Methionine	0.8205
Rice polish	7.51	Ca <sup>++</sup>	2.2659
Wheat floor	5.00	PO <sub>4</sub> <sup>-</sup>	1.2023
Oil (Sun-flower)	3.22	Na <sup>+</sup>	0.3244
Vitamin and mineral mix	5.00		

The whole experiment was conducted at room temperature during August, 2003 July, 2004. Nine age groups of each fish species, starting from 90- to 330-day, were reared for 30 days each under the sub-lethal concentrations of zinc with three replications. However, the control fish were reared in clean tap water for comparison. After every trail of 30 days, the fish were dissected and their organs viz., gills, kidney, liver, skin, muscle and scales were isolated. The organs of fish were digested in nitric acid and perchloric acid for zinc determinations through methods of SMEWW<sup>[5]</sup>. Data were analyzed for statistical significances using Analysis of Variance (ANOVA) and Duncan's Multiple Range tests. Correlation coefficients were obtained to explain the relationships among various parameters under study<sup>[6]</sup>.

## RESULTS

**Growth performance of fish under chronic exposure of zinc:** Feed intake, weight increment and feed conversion ratio in nine fish age groups varied significantly (p<0.01).

However, three fish species showed non-significant differences for feed intake and feed conversion ratios while differences for weight increments were statistically variable at p<0.05. The variations in feed intake, weight increments and feed conversion ratio of three fish species were statistically different under sub-lethal zinc contamination when compared with control. The interaction (species x treatment) for all the three parameters under study were statistically at par. Table 1 shows significant increase in feed intake by the fish up to 5th age group while it decreased for 6th and 7th groups. Weight increments were higher for 300 and 330 day fish while 150 day fish showed negative growth. Feed conversion ratios were significantly higher for 90, 300 and 330 day fish groups with non-significant differences. Feed intake by the fish was higher under zinc sub-lethal medium than in control. All the three fish species performed similarly in both treated and control regimes for the pattern of their feed intake. *Catla catla* showed negative growth in terms of average weight increment (-0.37±1.80 g). However, *Labeo rohita* showed significantly higher average weight increment of 0.97±1.93 g, followed by that of *Cirrhina mrigala* (0.26±1.56 g). *Cirrhina mrigala* in control had significantly higher weight increment of 1.94±1.20g, followed by that of *Labeo rohita* and *Catla catla*. Treated fish showed significantly lower feed conversion ratios than that of control. Treated *Catla catla* exhibited negative feed conversion ratio of -12.45±64.87 % while *Cirrhina mrigala* in control showed the highest feed conversion ratio of 113.74±50.57% (Table 1).

Table 1: Analysis of variance on feed intake, increase in weight and feed conversion efficiencies of three fish species under zinc sub-lethal concentrations in water

MS						
SOV	DF	Feed intake (g)		Weight increment (g)		Feed conversion (%)
Fish Age	8	3.787**		7.849**		7835.529**
Species	2	0.032 <sup>NS</sup>		3.391*		4707.140 <sup>NS</sup>
Treatment	1	21.407**		23.947**		107466.826**
Species x Treatment	2	0.021 <sup>NS</sup>		1.595 <sup>NS</sup>		1337.006 <sup>NS</sup>
Error	40	0.129		0.890		1849.236
		Feed intake (g)		Weight increment (g)		Feed conversion ratio (%)
Fish age						
90 days	1.45d		1.18b		93.80a	
120 days	1.50d		0.85b		64.08ab	
150 days	1.39d		-0.61c		17.32b	
180 days	2.96b		0.18bc		27.62b	
210 days	3.54a		0.60b		23.71b	
240 days	2.31c		0.64b		27.67b	
270 days	2.42c		0.23bc		22.71b	
300 days	3.03b		2.96a		104.87a	
330 days	2.97b		2.56a		93.43a	
Fish species	Treated	Control	Treated	Control	Treated	Control
<i>Catla catla</i>	2.97±0.93a	1.78±0.68a	-0.37±1.80a	1.30±0.61a	-12.54±64.87a	80.81±39.86a
<i>Labeo rohita</i>	3.00±1.03a	1.74±0.67a	0.97±1.93a	1.62±1.06a	27.39±59.38a	97.68±51.26a
<i>Cirrhina mrigala</i>	3.11±1.08a	1.78±0.66a	0.26±1.56a	1.94±1.20a	9.72±50.74a	13.74±50.57a

Means with similar letters in a single column are statistically similar at p<0.05

Table 2: Accumulation of zinc (mg L<sup>-1</sup>) in the fish organs during sub-lethal chronic exposure

Fish age	mg L <sup>-1</sup>	Fish organs	mg L <sup>-1</sup>	Fish species	mg L <sup>-1</sup>
90 day	397.55b	Gills	215.28c	<i>Catla catla</i>	703.40a
120 day	329.38b	Kidney	1141.47a	<i>Labeo rohita</i>	469.09b
150 day	497.30b	Liver	1173.17a	<i>Cirrhina mrigala</i>	474.78b
180 day	228.73b	Skin	222.75c		
210 day	519.40b	Muscle	91.71d		
240 day	644.84b	Scales	450.16b		
270 day	681.13b				
300 day	359.07b				
330 day	1284.42a				

Means with similar letters in a single column are statistically similar at p<0.05

Table 3: Age related accumulation patterns of zinc (µg g<sup>-1</sup>) in fish organs under sub-lethal concentrations (mg L<sup>-1</sup>)

Fish organs	Regression equation (y = a + bx)
Gills	= 229.58-0.104 <sup>NS</sup> (Fish age)
Kidney	= -590.78+8.475* (Fish age)
Liver	= 380.68+3.798 <sup>NS</sup> (Fish age)
Skin	= 140.62+0.358 <sup>NS</sup> (Fish age)
Muscle	= 81.87+0.078 <sup>NS</sup> (Fish age)
Scales	= -109.80+2.561* (Fish age)

\* = Significant at p<0.05; NS = Non-significant

**Zinc accumulation in fish organs:** Non-significant differences existed among the eight fish age groups (from 90 to 300 day) for the accumulation of zinc. However, 330 day fish showed significantly higher tendency for the accumulation of zinc than rest of the age groups. Among the three fish species, *Catla catla* showed significantly higher tendency for the accumulation of zinc than that of *Labeo rohita* and *Cirrhina mrigala*. Fish kidney and liver showed significantly higher tendency for the accumulation of zinc. Accumulation of metal in fish gills, skin, muscle and scales varied non-significantly (Table 2). Zinc accumulation in all the fish organs showed positive dependence upon fish age except for gills and regression coefficients for fish kidney and scales were significant only (Table 3).

## DISCUSSION

The present study showed that feed intake by all the three fish species, stressed with sub-lethal level of zinc, increased significantly when compared with control fish. However, this significant increase in feed intake did not result in fish weight escalation and hence resulted in low feed conversion ratios. All the three fish species showed uniformity in their response towards feed intake, weight increment and feed conversion ratios during zinc exposures. However, feed intake, weight increment and feed conversion ratios of fish varied significantly among nine age groups as 210 day fish showed significantly higher feed intake. Rainbow trout exposed to half the lethal concentration of copper showed initial loss of appetite but compensated during 39 day experiment so that their growth rate almost equaled to that of control.

Furthermore, compensation was faster at low ration than at higher one<sup>[7]</sup>. Thus, toxicity would not only influence the fish appetite but acclimation and feed conversion ratio that ultimately affected the growth performance of fish.

The evidence collected from the present investigation demonstrated that metal concentrations in fish organs increased with exposure of particulate metal during nine growth trails. However, sensitivity of the fish to zinc decreased with fish age. Pelgrom<sup>[8]</sup> reported that exposure of immature tilapia to copper concentrations ranging from 0 to 400 µg L<sup>-1</sup> and cadmium concentrations ranging from 0 to 155 µg L<sup>-1</sup> resulted increased whole body cadmium contents. Copper exposure resulted in an accumulation of copper as well as in a significantly decreased cadmium content of fish, probably as a result of cadmium excretion. Three fish species performed similarly for the accumulation of zinc in their bodies. However, variations were significant among all the fish organs. Fish liver, as a major organ for metabolism and kidney accumulated significantly higher concentrations of zinc than that of gills, skin, muscle and scales. *Catla catla* exhibited significantly higher tendency for the accumulation of zinc in its body than that of *Labeo rohita* and *Cirrhina mrigala*. Under sub-lethal metal toxicity, the concentration of metals in fish body increased significantly with age probably because of limited ability to store metals as exposure persisted. A positive correlation between the increase in metallic ions in tissue and increased metal tolerance in fish has also been observed. The concentrations of zinc increased in fish body significantly during last trial (330 day). Nussey<sup>[9]</sup> addressed the dependence of chromium, manganese, nickel and lead bio-accumulations in *Labeo umbratus* on size, gender and season (age). Bioaccumulation of chromium, manganese, nickel and lead varied between the gills, liver, muscle and skin. The gills generally had the highest metal concentrations due to their intimate contact with the environment and their importance as an effector of ionic and osmotic regulations. The liver, in its role as a storage and detoxification organ, had also accumulated high levels of metals during this investigation. However, muscle and skin accumulated significantly less metals. Nussey<sup>[9]</sup> reported decreased metal accumulation in fish organs with an increase in fish length. Therefore, smaller fish had higher body loads of metals due to various bioaccumulation processes. Relationship between growth performance and heavy metal accumulation in different organs of common bream (*Abramis brama*) was studied by Farkas<sup>[10]</sup>. The average metals concentration in the organs of fish varied in the range of: Cd, 0.39-1.98; Cu, 1.73-57.3; Zn, 12.27-159.3; Pb, 0.39-3.15 and Ni, 0.02-0.13 µg g<sup>-1</sup> dry weight. Heavy metal concentration

was found highest in liver and gills, while in muscle it was below the maximum permissible levels for human consumption along with significantly positive correlation among the heavy metal loads and growth performance.

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