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RESEARCH ARTICLE

Effects of Zinc and Lead Toxicity on the Growth and their Bioaccumulation in Fish

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ABSTRACT

This study evaluated the impacts of chronic exposure of waterborne zinc (Zn) and lead (Pb) on the growth and their bioaccumulation in three fish species viz. *Catla catla, Labeo rohita* and *Cirrhina mrigala.* Three fish species responded similarly for their feed intakes while weight increments and feed conversion efficiency (FCE) varied significantly due to Zn and Pb exposures. Younger fish were significantly more sensitive to metallic ion toxicity. Chronic exposure of both Zn and Pb (at 1/3rd of LC_{50}) to the fish caused significantly lesser gain in weight, feed intakes and FCE than that of control (un-stressed) fish. Amongst 9 age groups, 330-day fish exhibited significantly better growth in terms of weight gain and feed intake than the other age groups. Both Zn and Pb bioaccumulations varied significantly among fish organs while the patterns of their bioaccumulation did not vary significantly within three fish species. Fish liver and kidney accumulated significantly higher Zn and Pb during chronic exposures. However, Zn accumulation was significantly more than that of Pb in the fish body. Amongst three fish species, *Labeo rohita* exhibited significantly higher tendency to accumulate Zn while *Catla catla* amassed higher Pb in its body. The bioaccumulation of both Zn and Pb was positively dependent upon fish age and exposure concentration of metals. Zn bioaccumulation in fish body followed the order: liver>kidney>skin>gills>scale=muscle while that of Pb was: kidney>liver>gills>skin>muscle=scales.

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INTRODUCTION

The behavior of toxicants in water envisages biological response of an aquatic ecosystem. Different toxicants act through their toxicity, fate and specific nature while the response of biological system involves adaptations, defense, stress response and recuperation. An incredible use of metals in industries has resulted huge production of waste water and its discharge into aquatic bodies has become a major threat to the indigenous fish fauna in Pakistan (Rauf *et al*., 2009; Hussain *et al*., 2010). Since organisms are chronically exposed to low concentration of heavy metals in natural habitats, therefore, it is pertinent to evaluate the chronic effects (sub-lethal) of metals on the fish and other aquatic organisms. Growth is a straightforward indicator that can envisage all the impacts within the fish. The exposure of fish to metallic toxicity can cause loss of appetite (Javed and Saeed, 2010) that is not only associated with ration but also involve acclimation, appetite and FCE if it is principally desired to realize how a toxicant could affect the fish growth. Therefore, it is essential to know about fish nutrition, appetite, acclimation and FCE during growth trials under metal stress conditions.

Zinc as a trace element is essential for electron transfer in catalytic reactions. However, chronically higher intake of metals would lead towards bioaccumulation in the body organs (Nussey *et al*., 2000) that may become toxic to the fish (Kim and Kang, 2004). The fish inhabiting contaminated waters can take up metals from water, diet and skin (Rauf *et al*., 2009). During acute exposure, waterborne Pb can cause disruption of Ca^{2+}, chlorides, sodium, spinal deformities, black tail disease and disruption in the synthesis of hemoglobin in fish (Dallinger *et al*., 2002). Fish intestine can also accumulate higher amounts of Pb indicating a site of dietary Pb uptake. Pb, Zn and Cd are considered Ca^{2+} antagonist. Therefore, dietary intake of Ca^{2+} protects the body against Cd and Zn uptakes (McGeer *et al*., 2000). Cd works as a cumulative toxin, however, in low concentration it is an essential element for both animals and plants (Waqar, 2006). Metal compounds are potentially injurious or toxic and their sub-lethal exposures could cause reduced growth, behavioral and physiological disorders in the fish (DeBoeck *et al*., 1997). Due to rapid industrialization in the Punjab province, liquid wastes are generating and dumped untreated in to the aquatic habitats that are causing serious threats to the indigenous fish fauna, particularly *Catla catla*, *Labeo rohita* and *Cirrhina mrigala* are on the threshold of extinction (Rauf *et al*., 2009). This necessitates the evaluation of growth energetics of these cyprinids and their ability to bio-accumulate Zn and Pb during chronic exposures. This study will help sustainable conservation of indigenous fish fauna in the country.

MATERIALS AND METHODS

Growth experiments were conducted with 9 age groups (90- to 330-day old) of juvenile fish in glass aquaria under controlled laboratory conditions. The groups of three fish species viz. *Catla catla*, *Labeo rohita* and *Cirrhina mrigala* were grown, separately, under sub-lethal (1/3rd of LC_{50}) zinc concentrations of 10.41±0.10, 13.33±0.10 and 16.67±0.08 mg L^{-1} while the other groups were exposed to lead concentrations of 11.67±0.09, 11.68±0.11 and 16.64±0.06 mg L^{-1}, respectively as determined by Javed and Abdullah (2004). Ten fish of each species were grown, separately, after being acclimatized to laboratory conditions, in 50 liter glass aquaria with three replications for each treatment for 60 days. However, control fish were grown in metal free water for comparison. Fish aquaria were supplied with continuous aeration through capillary system. All growth trials were conducted under constant water pH (7.2), hardness (250 mgL^{-1}) and temperature (30^{o}C). The exposure media were replenished to sustain the required concentration of each metal in the aquaria throughout the period of this investigation. The increase/decrease in fish weights, feed intakes and FCE of each treated and control fish species were monitored on weekly basis. The water of each aquarium was checked for the desired metal concentration, as mentioned above, on daily basis (Anonymous, 1989). All fish groups were fed a feed (30% C.P. and 2.90 Kcal g^{-1} DE), to satiation, twice a day throughout the growth period of 60 days. At the end of each 60-day growth trial, fish were sacrificed and organs isolated for the determination of their respective metal (Anonymous, 1989). Data were statistically analyzed for significant differences by following Steel *et al*. (1996) while correlation coefficients were computed to elucidate relationships among variables defined for this study.

RESULTS

Fish growth under metal stress: Zinc exposure to all the nine age groups of three fish species exerted significant effects on their average weight gains, feed intakes and FCE values. Weight increments were significantly higher in 300-day age group, followed by 330-day fish. Age groups viz. 3, 4, 5 and 7 exhibited significantly least and negative growth while that of control was significantly higher. Average feed intake was significantly higher in 300-day fish, followed by 330- and 270-day age groups. All control (un-stressed) age groups demonstrated significantly higher FCE than the both Zn and Pb exposed fish (Table 1). Regarding overall performance of nine age groups, three fish species illustrated significantly inconsistent weight increments with the highest average increment of 0.97±0.93g by *Labeo rohita* while *Catla catla* exhibited negative growth. Feed intakes did not vary significantly among three fish species. However, treated fish showed significantly higher feed intakes than that of control. Zn treatment caused significant variations in FCE of three fish species with the mean highest FCE being observed for *Labeo rohita* (27.39±9.38%), followed by that of *Cirrhina mrigala* and *Catla catla*. The control *Cirrhina mrigala* demonstrated higher ($p<0.05$) FCE of 113.74±8.75%, followed by that of *Labeo rohita* (97.68±9.26) and *Catla catla* (80.81±11.86%).

Pb exposure to 300-day age group of fish caused significantly higher weight gains, followed by that of 9th age group. However 3rd, 4th, 5th and 7th age groups showed reduction in their average weights. Both 300 and 330-day control fish groups showed significantly higher weight incrementss. Pb exposure caused significantly higher feed intakes in both 210- and 240-day fish groups. However, feed intakes were significantly lesser in control than that of Pb exposed three fish species. Amongst treated age groups, 300-day (8th group) fish exhibited significantly higher FCE, followed by that of 9th age group. *Labeo rohita* accomplished significantly higher average weight of 0.54±0.25g, followed by that of *Catla catla* (0.07±0.33g) while *Cirrhina mrigala* gave significantly negative growth (–0.18±0.14g). Pb exposure caused non-significant variations in feed intakes of all the three fish species while it was significantly higher in control fish. Pb exposed *Labeo rohita* exhibited significantly higher FCE of 13.69±4.50%, followed by that of *Catla catla* (2.46±1.60%) and *Cirrhina mrigala* (-6.68±2.26%). Amongst three control fish species, *Cirrhina mrigala* gave significantly higher average FCE of 113.75±10.57% while *Labeo rohita* and *Catla catla* had the values of 97.68±5.26 and 80.81±9.86%, respectively with statistically significant differences (Table 2).

Bioaccumulation of metals in fish body: All age groups of fish illustrated significantly variable abilities for the uptake and accumulation of both Zn and Pb in their kidney, gills, liver, scales, skin and muscle. Zn accumulation was significantly higher in fish liver, followed by that in kidney, skin, gills, scales and muscle. However, these accumulations were non-significantly different in the fish scales and muscle. Three control fish species showed non-significant differences in their ability to accumulate Zn that followed the order: liver> gills> kidney> muscle>skin>scales (Table 3). Pb exposure caused significantly higher amassing in the kidney of both *Cirrhina mrigala* and *Catla catla*. All organs, except muscle and scales, build up significantly inconsistent Pb concentrations: kidney>liver>gills>skin>muscle=scales. The ability of three control fish species to accumulate Pb did not vary significantly however its amassing was significantly higher in liver, followed by that in kidney, skin, muscle, gills and scales (Table 4). The accumulation of both Zn and Pb in fish were directly correlated ($P<0.05$) with metallic ion exposure concentration and fish age. Zn accumulation in all the fish organs, except

Table 1: Growth performance of fish exposed to sub-lethal concentrations of Zn

Age Group	Days	Fish Species	Initial Average Weights(g)		Average Weight Increment (g)		Feed Intake (g)		FCE (%)	
			Treated	Control	Treated	Control	Treated	Control	Treated	Control
I	90-120	*Catla catla*	9.37±1.25	9.78±2.21	0.69b	1.12c	2.23a	1.01a	30.94c	110.89c
	90-120	*Labeo rohita*	10.90±0.19	12.20±0.82	1.03a	1.78a	1.72b	1.04a	59.88b	171.15a
	90-120	*Cirrhina mrigala*	10.49±0.33	10.40±0.40	1.05a	1.38b	1.60b	1.11a	65.62a	124.32b
II	121-150	*Catla catla*	10.42±0.78	11.91±0.39	0.54b	0.42c	1.90a	1.15a	28.42b	36.52c
	121-150	*Labeo rohita*	13.29±0.32	14.16±0.39	0.68a	0.95b	1.85a	1.09a	36.76a	87.15b
	121-150	*Cirrhina mrigala*	13.29±0.78	13.18±0.40	0.68a	1.82a	1.89a	1.14a	35.98a	159.65a
III	151-180	*Catla catla*	16.72±1.16	16.00±1.37	-2.72c	0.90a	2.30a	0.62a	-118.26c	145.16a
	151-180	*Labeo rohita*	16.00±1.29	18.13±1.28	-1.72b	0.55c	2.06a	0.59a	-83.49b	93.22c
	151-180	*Cirrhina mrigala*	18.49±1.48	20.16±1.25	-1.52a	0.84b	2.16a	0.61a	-70.37a	137.70b
IV	181-210	*Catla catla*	23.66±6.48	21.00±3.86	-1.94c	1.42b	4.02a	2.01a	-48.26c	70.65c
	181-210	*Labeo rohita*	27.76±8.15	18.78±3.61	-1.59b	2.18a	3.81b	2.00a	-41.73b	109.00a
	181-210	*Cirrhina mrigala*	17.95±2.23	33.07±8.52	-1.07a	2.08a	3.90a	2.01a	-27.43a	103.48b
V	211-240	*Catla catla*	22.62±2.58	14.25±3.77	-1.09a	0.75c	4.62a	2.50a	-23.59b	30.00c
	211-240	*Labeo rohita*	34.22±14.70	20.67±2.52	2.10b	0.83b	4.55a	2.48a	46.15a	33.47b
	211-240	*Cirrhina mrigala*	40.71±19.30	25.75±8.81	-0.92a	1.90a	4.58a	2.49a	-20.09b	76.30a
VI	241-270	*Catla catla*	24.55±6.55	20.58±3.44	0.34c	1.10a	2.27c	2.21a	14.98c	49.77a
	241-270	*Labeo rohita*	35.93±13.24	18.22±2.32	0.48b	0.64b	2.42b	2.19a	19.83b	29.22b
	241-270	*Cirrhina mrigala*	33.29±10.87	40.02±11.92	0.68a	0.57b	2.61a	2.18a	26.05a	26.15b
VII	271-300	*Catla catla*	27.38±13.70	9.84±3.39	-2.61c	2.32a	2.62a	1.84a	-99.62c	126.09a
	271-300	*Labeo rohita*	21.26±2.10	25.51±6.29	0.52a	1.28b	3.09c	1.86a	16.83a	68.82b
	271-300	*Cirrhina mrigala*	53.35±13.70	22.80±3.82	-1.36b	1.24b	3.25b	1.88a	-41.85b	65.96b
VIII	301-330	*Catla catla*	46.16±38.17	30.52±5.05	2.47c	1.84c	3.34c	2.32a	73.95b	79.31c
	301-330	*Labeo rohita*	21.21±3.02	24.86±5.70	3.61a	3.66a	3.89b	2.14a	92.80a	171.03a
	301-330	*Cirrhina mrigala*	69.39±50.85	50.27±6.67	2.87b	3.30b	4.18a	2.30a	68.66c	143.48b
IX	331-360	*Catla catla*	27.15±3.94	28.86±3.51	0.97c	1.83c	3.39c	2.32a	28.61c	78.88c
	331-360	*Labeo rohita*	36.75±24.44	22.19±1.04	3.63a	2.67b	3.65b	2.30a	99.45a	116.09b
	331-360	*Cirrhina mrigala*	43.30±9.07	55.02±22.07	1.94b	4.33a	3.81a	2.32a	50.92b	186.64a
		Catla catla			-0.37±0.80c	1.30±0.61c	2.97±0.93a	1.78±0.68a	-12.54±4.87c	80.81±11.86c
		Labeo rohita			0.97±0.93a	1.62±1.06b	3.00±1.03a	1.74±0.67a	27.39±9.38a	97.68±9.26b
		Cirrhina mrigala			0.26±0.56b	1.94±1.20a	3.11±1.08a	1.78±0.66a	9.72±5.74b	113.74±8.57a

Means with similar letters in a single column are statistically non-significant at P<0.05.

gills, showed positive correlation with fish age while these accumulations in gills and skin showed significantly direct correlation with metallic ion concentration. Fish kidney and muscle showed significantly higher ability to concentrate Pb which was positively dependent upon the extent of metallic toxicity of water (Table 5).

DISCUSSION

Trace metals play a significant role in all facet of animal's life. However, their excessive amounts in water would become toxic and cause deleterious effects on fish (Jabeen and Javed, 2011). The exposure of both Zn and Pb, at sub-lethal concentrations, to all age groups of fish caused significantly higher feed intakes than the control fish. However, FCE of fish decreased significantly with increasing metallic ions toxicity (Ali *et al*., 2003). Three fish species responded similarly for their ability to take feed under both Zn and Pb exposures. Weight increments, feed intake and FCE of fish varied significantly among nine age groups. Different fish species showed variable sensitivity to various metals based on species specificity and age (Rauf *et al*., 2009; Stoskus *et al*., 1999). The stress of Zn and Pb caused significant influence on fish appetite also (DeBoeck *et al*., 1997). However, acclimation and species specificity towards FCE affected the growth of all the three fish species significantly. Metals can enter the fish through gills, food particles or/and skin. After ingestion, metals would start accumulating in the body organs depending upon the intensity of their uptake (Vinodhini and Narayanan, 2008). Significant decrease in weights of *Labeo rohita*, *Catla catla* and *Cirrhina mrigala* due to manganese exposure has been reported by Hayat *et al*. (2007). McGeer *et al*. (2000) reported significant interactions among feed intake, growth, oxygen consumption and ionic regulation in rainbow trout due to chronic exposures of Cd, Cu and Zn. This study also reveals metallic ion concentration specific escalation of both Zn and Pb in the body organs of all the three fish species during chronic exposures. However, sensitivity of fish to both Zn and Pb decreased significantly with fish age (Abdullah and Javed, 2006) and weight (size) which is closely linked with fish metabolism (Ansari *et al*., 2006). Fish organs exhibited significantly variable tendencies for the uptake and amassing of both Zn and Pb. Fish Liver and kidney showed significantly higher while that of muscle and scales with lower tendency to accumulate both Zn and Pb. Significantly higher concentrations of metals in the liver and gills while least in the muscles of *Oreochromis niloticus*, *Mugil cephalus* and *Anguilla anguilla* were reported by Yilmaz (2009). *Labeo rohita* showed

Table 2: Fish growth under chronic exposure of Pb

Age group	Days	Fish Species	Initial Average Weight (g)		Average Weight Increment (g)		Feed Intake (g)		*FCE (%)	
			Treated	Control	Treated	Control	Treated	Control	Treated	Control
	90-120	*Catla catla*	10.53±0.74	9.78±2.21	0.48c	1.12c	2.35	1.01	20.42b	110.89c
I	90-120	*Labeo rohita*	10.79±0.80	12.20±0.82	1.15a	1.78a	2.07	1.04	55.55a	171.15a
	90-120	*Cirrhina mrigala*	10.94±0.55	10.40±0.40	0.93b	1.38b	1.73	1.11	53.76a	124.32b
	121-150	*Catla catla*	11.90±1.13	11.91±0.39	0.43b	0.42c	1.76	1.15	24.43b	36.52c
II	121-150	*Labeo rohita*	13.56±0.43	14.16±0.39	0.43b	0.95b	1.85	1.09	23.24b	87.15b
	121-150	*Cirrhina mrigala*	12.36±0.39	13.18±0.40	0.60a	1.82a	1.85	1.14	32.43a	159.65a
	151-180	*Catla catla*	15.46±1.48	16.00±1.37	-1.01a	0.90a	3.17	0.62	-31.86a	145.16a
III	151-180	*Labeo rohita*	17.96±2.18	18.13±1.28	-1.71b	0.55c	1.94	0.59	-88.14b	93.22b
	151-180	*Cirrhina mrigala*	16.91±2.96	20.16±1.25	-1.78b	0.84b	1.47	0.61	-121.09c	137.70a
	181-210	*Catla catla*	28.29±7.49	21.00±3.86	-1.57c	1.42b	3.54	2.01	-44.35c	70.65b
IV	181-210	*Labeo rohita*	22.96±4.18	18.78±3.61	-1.15b	2.18a	3.54	2.00	-32.48b	109.00a
	181-210	*Cirrhina mrigala*	18.58±3.54	33.07±8.52	-0.91a	2.08a	3.87	2.01	-23.51a	103.48a
	211-240	*Catla catla*	26.48±2.50	14.25±3.77	-1.15b	0.75b	4.62	2.50	-24.89b	30.00b
V	211-240	*Labeo rohita*	26.25±6.34	20.67±2.52	0.75a	0.83b	4.19	2.48	17.90a	33.47b
	211-240	*Cirrhina mrigala*	55.94±6.79	25.75±8.81	-1.59c	1.90a	4.73	2.49	-33.61c	76.30a
	241-270	*Catla catla*	14.91±8.48	20.58±3.44	1.89a	1.10a	4.39	2.21	43.05a	49.77a
VI	241-270	*Labeo rohita*	26.82±10.39	18.22±2.32	1.19b	0.64b	4.32	2.19	27.55b	29.22b
	241-270	*Cirrhina mrigala*	34.14±13.21	40.02±11.92	0.61c	0.57b	2.59	2.18	23.55b	26.15b
	271-300	*Catla catla*	14.37±6.67	9.84±3.39	-1.21b	2.32a	2.62	1.84	-46.18b	126.09a
VII	271-300	*Labeo rohita*	38.13±7.48	25.51±6.29	0.89a	1.28b	2.87	1.86	31.01a	68.82b
	271-300	*Cirrhina mrigala*	34.43±11.03	22.80±3.82	-1.11b	1.24b	2.62	1.88	-42.37b	65.96b
	301-330	*Catla catla*	28.34±5.88	30.52±5.05	1.20a	1.84b	3.38	2.32	35.50b	79.31c
VIII	301-330	*Labeo rohita*	20.72±11.15	24.86±5.70	2.36a	3.66a	3.86	2.14	61.14a	171.03a
	301-330	*Cirrhina mrigala*	53.65±11.94	50.27±6.67	0.90b	3.30a	3.09	2.30	29.13c	143.48b
	331-360	*Catla catla*	22.73±12.06	28.86±3.51	1.60a	1.83c	3.48	2.32	45.98a	78.88c
IX	331-360	*Labeo rohita*	21.44±2.35	22.19±1.04	0.98b	2.67b	3.57	2.30	27.45b	116.09b
	331-360	*Cirrhina mrigala*	32.47±2.32	55.02±22.07	0.69c	4.33a	3.19	2.32	21.63b	186.64a
		Catla catla			0.07±0.33b	1.30±0.61b	3.26±0.92a	1.78±0.68a	2.46±1.60b	80.81±9.86c
		Labeo rohita			0.54±0.25a	1.62±1.06a	3.13±0.98a	1.74±0.67a	13.69±4.50a	97.68±5.26b
		Cirrhina mrigala			- 0.18±0.14c	1.94±1.20a	2.09±1.06a	1.78±0.66a	-6.68±2.26c	113.75±10.57a

FCE= Feed intake/Increase in weight x 100; Means with similar letters in a single column are statistically non-significant at P<0.05.

Table 3: Accumulation of Zn (μg g^{-1}) in fish body organs

	Metal Concentrations (μg g^{-1})					
Fish Age (Days)	Gills	Kidney	Liver	Skin	Muscle	Scales
90-day	329.38±3.25a	657.28±5.63c	897.08±6.32a	186.44±6.30b	53.14±6.37c	61.98±4.39c
120-day	319.28±5.26b	665.22±7.12c	644.75±5.89c	188.59±7.29b	39.24±5.32e	69.21±8.56b
150-day	265.29±4.26c	822.76±6.35a	691.81±4.26c	260.09±5.86a	54.36±4.19c	89.49±7.45a
180-day	248.00±2.98d	767.96±8.35b	640.19±7.21c	120.53±6.38c	42.59±7.19d	53.12±5.36d
210-day	176.25±8.36f	662.00±4.26c	731.88±3.95b	278.87±9.46a	40.23±8.24d	62.14±6.35c
240-day	148.25±7.21g	502.25±5.92e	932.28±5.86a	260.35±8.35a	74.31±6.38a	73.62±7.29b
270-day	146.23±8.25g	656.58±4.32c	728.83±7.36b	230.84±7.26a	62.33±5.96b	84.98±8.21a
300-day	192.78±9.31e	653.30±7.12c	352.21±6.54d	189.74±5.38b	43.53±7.10d	52.85±6.87d
330-day	112.08±7.14h	565.88±5.29d	320.50±4.29e	289.34±3.98a	45.66±8.24d	84.03±5.26a
Treated Fish						
Catla catla	174.95±3.21c	611.69±2.15b	751.16±4.08b	251.56±3.98a	54.80±3.26b	63.21±2.69b
Labeo rohita	221.95±2.59a	680.42±3.16a	780.58±3.12a	179.75±4.11c	61.32±4.11a	60.53±3.31b
Cirrhina mrigala	198.94±3.12b	632.29±3.57b	715.78±2.51c	236.95±2.35b	63.01±2.63a	67.73±4.99a
*Means ± SD	198.61±23.50d	641.46±35.27 b	749.17±32.44a	222.75±37.95c	59.71±4.33e	63.82±3.63e
Control Fish						
Catla catla	68.25±4.01a	40.11±3.21a	80.85±3.59a	18.19±3.32a	28.55±3.65a	10.11±2.59b
Labeo rohita	63.11±3.56a	38.16±4.12a	78.66±4.13a	20.14±2.65a	30.19±4.11a	13.54±1.25a
Cirrhina mrigala	68.18±3.10a	36.25±3.58a	79.21±3.87a	24.25±3.16a	30.75±3.21a	11.02±2.35b
*Means ± SD	66.51±2.94b	38.17±1.93c	79.57±1.14a	20.86±3.09e	29.83±1.14d	11.56±1.78f

Means with similar letters in a single column and *row for overall means are statistically non-significant at P<0.05.

significantly higher tendency to accumulate Zn, followed by that of *Cirrhina mrigala* and *Catla catla* while Pb accumulation followed the order: *Catla catla* > *Labeo rohita* > *Cirrhina mrigala* with significant differences.

Chronic exposure of both Zn and Pb caused amassing of metals in fish body that varied significantly with age resulting in a positive correlation between exposed metallic ions and fish age possibly due to restricted capacity of fish to amass metals (Azmat and Javed, 2011). This shows an exponential role of exposure period when the storage ability of kidney and liver would become limited up to a certain extent that stimulates fish muscle to

Table 4: Pb accumulations (μg g^{-1}) in fish body organs

	Metal Concentrations (μg g^{-1})					
Fish Age	Gills	Kidney	Liver	Skin	Muscle	Scales
90-day	98.36±5.23a	269.02±5.42c	284.81±6.35a	50.98±7.41c	54.02±8.21b	46.10±6.35c
120-day	100.91±4.98a	218.52±6.32d	220.56±4.25b	57.25±8.16c	80.20±6.32a	52.34±5.29b
150-day	85.46±6.35b	286.12±7.12c	217.71±7.12b	75.27±7.01a	47.45±4.26c	50.68±6.86b
180-day	40.10±6.15e	205.93±5.26d	269.91±8.59a	65.92±6.85b	28.11±7.53e	52.84±7.49b
210-day	98.45±7.25a	174.71±4.55e	229.36±5.69b	56.89±5.65c	53.26±6.58b	60.72±8.12a
240-day	106.61±6.85a	251.21±4.95c	231.97±7.85b	53.49±4.29c	42.14±5.26c	40.95±8.36d
270-day	55.81±3.98c	368.39±6.35ab	174.68±6.39c	44.72±5.65dc	41.97±6.29c	41.37±7.28c
300-day	45.93±4.59d	349.02±7.12b	193.81±8.54bc	64.95±6.82b	36.31±7.42d	39.56±5.62c
330-day	43.45±6.84d	430.62±8.05a	138.94±7.52d	50.10±8.24c	35.29±8.21d	42.50±5.19c
Treated Fish						
Catla catla	80.59±2.36b	281.45±3.95a	227.75±4.26a	54.63±4.12a	51.02±4.23a	51.26±3.01a
Labeo rohita	84.41±3.31a	258.59±4.16b	199.94±4.19b	58.26±3.52a	47.00±3.59a	43.62±2.95b
Cirrhina mrigala	78.02±2.56b	294.80±3.21a	196.90±3.25b	47.96±2.54b	42.56±3.58b	42.82±2.56b
*Means ± SD	81.01±3.22c	278.28±18.31a	208.19±17.00b	53.61±5.22d	46.86±4.23e	45.90±4.66e
Control Fish						
Catla catla	12.16±2.35a	35.21±2.56a	65.21±4.38a	14.16±4.16a	13.13±4.19a	10.95±3.21a
Labeo rohita	12.25±4.32a	34.84±3.45a	66.38±2.69a	15.28±3.59a	14.35±3.56a	9.91±2.54a
Cirrhina mrigala	11.66±3.56a	32.05±3.25a	62.15±3.12a	13.14±3.25a	11.65±4.21b	10.01±2.59a
*Means ± SD	11.69±0.89d	34.03±1.73b	64.58±2.18a	14.19±1.07c	13.04±1.35c	7.29±4.62e

Means with similar letters in a single column and *row for overall means are statistically non-significant at P<0.05.

Table 5: Metals bioaccumulation trends in fish body organs

	Metal Stress	Correlation Coefficients					
		Gills	Kidney	Liver	Skin	Muscles	Scales
Age Based Metal Accumulation in	Zn stressed fish	0.6571	0.5866	0.8325	0.5216	0.5333	0.2167
	Pb stressed fish	0.8721	0.6657	0.9217	0.5427	0.6622	0.3424
Concentration Based Metal Accumulation in	Zn stressed fish	0.6636	0.8251	0.8722	0.6222	0.6721	0.5117
	Pb stressed fish	0.7211	0.7257	0.6222	0.5717	0.8759	0.3577
	Control fish	0.4257	0.4001	0.1051	0.2511	0.3389	0.4014

Critical value (1-tail) ± 0.4803

amass metals (Cinier *et al*., 1997). Fish gills generally had the highest metal concentrations, due to their intimate contact with the environment and its importance as an effecter of ionic and osmotic regulations. The chronic stress caused liver to store significantly higher Zn and Pb, as organs of detoxification, than that of other organs (Nussey *et al*., 2000). Pb is known to bioaccumulate in the tissues of fish (Dallas and Day, 1993). The uptake of aqueous Pb (Pb^{2+}) across the gills into the blood stream is the primary mode of uptake in fish (Coetzee, 1996). Zn accumulation in all the three fish species followed the order: liver>kidney>skin>gills>scales=muscle while Pb was: kidney > liver > gills > skin > muscle = scales. Significantly lower accumulation of Pb in the muscle of *Catla catla* has also been reported by Palaniappan and Karthikeyan (2009) that followed the order: kidney > liver > gill > brain > muscle.

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