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A Study on Length-Weight Relationships (LWR) and Growth Responses of Major Carps Exposed to Lead (Pb)

Saima Naz[*], Muhammad Javed and Amara Tahir
Department of Zoology and Fisheries, University of Agriculture, Faisalabad-38040, Pakistan.
*Corresponding author's E-mail: sam_ali_3@yahoo.com

Abstract
A group of *Catla catla, Labeo rohita* and *Cirrhina mrigala,* was exposed to sub-lethal concentrations of lead, separately, in glass aquaria whereas other another group was kept un-stressed as a control. The duration of trail was for 30 days. Both stressed and unstressed fish species were transferred to earthen ponds to rear, separately, for six months. After this exposure period, their growth performance in terms of weight, fork and total length were investigated at final harvest. Analysis of Variance and regression were applied to find out length and weight relationship of three fish species. Among three fish species *Cirrhina mrigala* gained maximum weight of 53.76± 7.39 g that varied significantly ($p<0.05$) from that of *Labeo rohita (*41.7± 9.48 g) but similar to the weight gains of *Catla catla* (52.65± 9.40 g). Fork and total length increments were observed significantly ($p<0.05$) higher in *Cirrhina mrigala* followed by that of *Catla catla* and *Labeo rohita*. Regarding length-weight relationship, all the three lead stressed fish species showed allometric growth. It was concluded that this investigation could stoutly be supportive to the researchers and policy makers for the preparation of very effectual sustainable management plans of fishery resources for the riverine systems.

1. Introduction

Water pollution is increasing gradually around industrial areas in Pakistan, which is the main environmental issue. Several pollutants like heavy metals, pesticides and fertilizers may affect the natural water systems. When heavy metals are discharged into the natural waters, they could affect the organisms due to their stable and persistent nature. Water pollutants affect the various biochemical and physiological responses of aquatic animals when they absorb different toxicants [1]. It has been recently found that coastal pollution has heavily influenced the environment in various developing countries. In an aquatic environment, the effects of heavy metals are severe due to their biological magnification in the food chain. The global heavy metal accumulation in natural waters is the most important environmental problem. High toxicity and hazardous materials, including heavy metals, are released by various industries [2], causing toxicity in the water systems. The accumulation of metals in an aquatic environment has direct influence on man and the ecosystem. Heavy metals play a significant role as chemical pollutants in fresh water and causing cytotoxic, mutagenic and carcinogenic effects on animals [3]. In fresh water, heavy metals are naturally present as trace element, but today their balance is being disturbed due to enhanced domestic, industrial and mining activities [4] and causing adverse effects on the diversity [5]. Several factors like salinity, pH and water hardness can play a main role in heavy metals uptake and accumulation in the living organism up to a level of lethal concentrations and cause ecological hazard [6]. Fishes are used to monitor the health of aquatic bodies [7] because of their tendency to accumulate heavy metals in their organs [8]. The fishes can accumulate heavy metals from their diet and water [4]. Heavy metals enter in the fish body through the gills, skin and food [9].

Lead occurs in the environment as a result of both anthropogenic and natural process, with coal burning, mining and smelting and also use in gasoline contributing most to lead contamination of freshwater environment. In the majority of eco-toxicological studies, prolonged effect of lead on freshwater fish have been evaluated [10]. These studies report a wide range of toxic effects induced by chronic exposure to increase lead concentration, including effects on pituitary function, oocyte growth and gonadsomatic index, neurological disorders and scoliosis [11]. As for other metals, large variation in sensitivity to lead exists [12] that, together with the limited number of reports of chronic lead exposure studies, limits our ability to establish safe and realistic environmental regulations. The purpose of the present study was to expand the information about sensitivity to chronic lead exposure in freshwater fish viz. *Catla catla, Labeo rohita* and *Cirrhina mrigala*

2. Materials and Method

Eight weeks old fingerlings of major carps viz. *Catla catla, Labeo rohita* and *Cirrhina mrigala* were acclimatized for 14 days in cemented tanks under laboratory conditions, at Fisheries Research Farms, University of Agriculture, Faisalabad, Pakistan. Two groups were assigned to each species of fish after acclimation. Prior to start the experiment, the average weights, fork and total lengths of both groups of fish were recorded. Out of two groups, one group was exposed to sub-lethal concentrations ($1/3^{rd}$ of LC_{50}) of 8.83, 10.00 and 16.67 mgL^{-1} of lead for *Catla catla*, *Labeo rohita* and *Cirrhina mrigala*, respectively [28], for 30 days while the other group was kept unstressed (control) in the glass aquaria. For the preparation of stock solution of 100 ppm (parts per million), the pure compound of lead ($PbCl_2$) was dissolved in distilled water. Three replications were made for all stress trials. Through capillary system, with the help of air pumps, continuous air was supplied to all the test media. Two times a day, fish were nourished to-satiation with the feed (35% digestible protein and 290 Kcalg^{-1} digestible energy) at 10:00 hours. The growth parameters viz. increase or decrease in average weight, fork and total lengths as well as feed intakes of three fish species were determined during the trial of 30 days. The water quality parameters viz. pH, temperature, electrical conductivity, dissolved oxygen, carbon-dioxide, chlorides, total ammonia, sodium, calcium, potassium, magnesium and total hardness of water were also examined at 09:00 hours daily by following the methods of [29].

2.1. Fish growth studies in ponds
Both treated and control fish were stocked in outdoor earthen ponds, separately, with the stocking density of 2.87 m^3 per fish [30] after 30 days sub- lethal metal stress trails. *Catla catla*, *Labeo rohita* and *Cirrhina mrigala* had interspecies ratio of 30, 50 and 20 percent, respectively. From the next day of stocking, all the earthen ponds were fertilized with poultry droppings on account of its nitrogen contents @ 0.16 g nitrogen / 100 g of fish weight daily. However, supplementary feed (35% digestible protein and 2.90 Kcalg^{-1} digestible energy) was also supplied to fish daily (six days a week) at the rate of 2% of fish biomass, when the water temperature exceeded 22 ºC. For growth studies, performance of test netting of fish as well as evaluation of growth parameters of fish viz. increase/decrease in wet weight, fork and total lengths as well as feed intake were also carried out on fortnightly basis for both treated and control fish for six months.

2.2. Statistical analysis of data
The data on the parameters of fish growth was subjected to statistical analysis by following [31] through microcomputer. MSTATC, MICROSTAT and STATISTICA packages were used to perform Analysis of Variance and Regression to observe and determine the evaluation of mean values for different parameters and significance of interactions.

3. Results
Three fish species viz. *Catla catla*, *Labeo rohita* and *Cirrhina mrigala* were reared under sub-lethal concentration of 8.33, 10.00 and 16.67 mgL^{-1}, respectively for 30 days.

3.1. Growth performances of fish during lead stress
Table 1 represented the average initial, final and increments of weight, fork and total length of *Catla catla*, *Labeo rohita* and *Cirrhina mrigala* during the sub-lethal exposure of lead for 30 days. *Catla catla* showed no increment in weight. However, average fork length showed increment of 0.97 mm but total length showed negative as -0.20 mm. Stressed *Labeo rohita* showed negative growth in terms of weight, fork and total lengths increments as -1.06 g,-7.00 and -7.40 mm, respectively. *Cirrhina mrigala* gained an average weight of 0.3 g and the fork length showed negative growth (-5.10 mm) while total length increased as 0.50 mm.

The mean values of water quality parameters viz. dissolved oxygen, temperature, chlorides, sodium, potassium, calcium, magnesium and total hardness were monitored on daily basis during stress period (Table 1).

3.2. Length-weight relationship
Length- weight relationship in fish is considered a parameter to predict and evaluate the degree of fish health and conditions conducive for its growth or otherwise. Fish weight may be considered as a function of the length. This relationship of length and weight follows approximately the cube law relationship expressed by the formula: $K = W/L^3$ in which "W" is symbol for weight (g) and "L" is the symbol for the length (mm).

Length-weight relationship (LWR) of *Catla catla*, *Labeo rohita* and *Cirrhina mrigala* were estimated and all of the treated and control fish species showed positive and statistically highly significant relationships between weight-fork length and weight-total length, as depicted by the value of R^2 (Coefficient of determination). Regarding length-weight relationships for lead stressed fish species, allometric growth was exhibited by all of them as the values of exponent b (regression coefficient) for *Catla catla*, *Labeo rohita* and *Cirrhina mrigala* were 3.445, 3.301 and 3.510 in case of weight-fork length equations and 2.865, 3.391 and 3.672 in weight-total length equations, respectively. However, in control fish species, *Labeo rohita* showed isometric growth as the values of exponent b in weight-fork length and weight-total length equations were 3.00 and 3.03, respectively, while other control fish species exhibited allometric growth as indicated by their regression coefficients (Table 2).

Discussion

Lead occurs in environment in a spacious range of physical and chemical forms that manipulate the behavior of fish negatively at concentration higher than normal. Most of the lead found in the environment is in the inorganic form [13] and it is the most stable ionic species present in the environment and consider as the form in which the maximum bioaccumulation of lead takes place in aquatic organisms. However, the toxicity of lead is based upon many factors including fish age, pH and hardness of the water [14]. In the present investigation, lead exposed *Catla catla* did not show any weight increment, while *Labeo rohita* showed negative increment in weight. [15] reported that the lead exposed *Cirrhina mrigala* showed significantly lower weight increment than control fish, however, reduced growth rate of rockfish (*Sebartes schlegeli*) due to Cu stress was reported by [16] as there was an inverse relationship between growth and Cu exposure. [17] determined the effect of sub-lethal concentrations of manganese exposed to fingerlings of *Catla catla, Labeo rohita* and *Cirrhina mrigala* for 30 days. Their results showed negative fish growth during the exposure period. [18] investigated the effect of copper on survival, growth and feed intake of *Cirrhina mrigala* for 60 days. Their results showed significantly reduced feed intake by fish at all the Cu treatments.

The Length-Weight relationship is an important tool to analyze fish populations. Its applications vary from simple estimates of an individual's weight to indication of fish body condition factor [19]. Knowledge on this relationship also helps to recognize energy investments for growth or reproduction as a natural cyclic phenomenon of natural populations [20]. Again, length weight relationship in fish is influenced by a number of other factors including gonad maturity, sex, diet, stomach fullness, health, and preservation techniques as well as season and habitat [21]. The length-weight relationship presented here may assist fish biologists to derive weight estimates for fishes that are measured but not weighed. This information is obligatory by most of models of stock assessment to estimate fishing mortality, population of cohorts and population of spawning stock as well as this investigation could strongly helpful to the researchers and policy makers for the preparation of very helpful and sustainable management plans of fishery resources of the riverine systems.

Growth of fish can be described as either allometric or isometric depending on the exponent b (regression coefficient) of the length-weight relationship which is normally between 2.0 and 4.0. The value b = 3.0 indicates that the fish grows symmetrically or isometrically while values other than 3.0 indicate allometric growth [22]. Length-weight relationships of five fish species were estimated by [23] in Epe Lagoon, Lagos. They calculated 95% confidence interval of length for about 76.19% of the fish species while length-weight relationships of 35 fish species were estimated by [24] from Badagry Creek, Lagos. The range of value of regression coefficient (2.670 – 3.672) obtained in this study is similar to the values (2.607-3.254) recorded by [24] who studied the length-weight relationships of 35 fish species from Badagry Creek, Lagos. It is also similar to the b (regression coefficient) values (2.790-3.210) obtained in [23] who studied the length-weight relationships of five fish species in Epe Lagoon, Lagos. The values of b for *Catla catla*, *Labeo rohita* and *Cirrhina mrigala* reported in this study for length weight relationships are different from those reported by [24] for Badagry Creek, which was directly connected to Ologe Lagoon. It is expected that this inconsistency is due to seasonal variability of the environment, food availability [24; 25], sampling size and the length interval within different areas [26] or habitat fitness [27].

Conclusion

Regarding growth performance of major carps, *Cirrhina mrigala* gained maximum weight that varied significantly ($p<0.05$) from that of *Labeo rohita* but non-significantly from the weight gains of *Catla catla*. Fork and total length increments were observed significantly ($p<0.05$) higher in *Cirrhina mrigala* followed by that of *Catla catla* and *Labeo rohita*. Regarding length-weight relationships for lead stressed fish species, allometric growth was exhibited by all of them while in case of unstressed fish species, only *Labeo rohita* showed isometric growth while other species exhibited allometric growth as indicated by their regression coefficients. This investigation could stoutly supportive to the researchers and policy makers for the preparation of very effectual sustainable management plans of fishery resources of the riverine systems.

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Table-1. Growth responses of *Catla catla, Labeo rohita* and *Cirrhina mrigala* during sub-lethal exposure of lead.

	Catla catla	*Labeo rohita*	*Cirrhina mrigala*
Exposure concentration (mgL^{-1})	8.33	10.00	16.67
Fish survival rate (%)	100	100	100
Initial average fish weight (g)	3.78±0.97	2.83±1.02	7.45±2.27
Final average fish weight (g)	3.78±1.19	1.77±0.60	7.76±0.83
Weight increment (g)	0.00 **c**	-1.60 **b**	0.31 **a**
Initial average fork length (mm)	67.33±6.47	60.00±6.38	87.50±13.08
Final average fork length (mm)	68.30±8.00	53.00±6.20	82.40±7.80
Fork length increment (mm)	0.97 **a**	-7.00 **c**	-5.10 **b**
Initial average total length (mm)	77.50±7.29	68.00±6.68	98.50±12.48
Final average total length (mm)	7.30±8.90	60.60±8.60	99.00±4.30
Total length increment (mm)	-0.20 **b**	-7.40 **c**	0.50 **a**
Feed intake (g)	2.70±0.15 **a**	4.62±0.24 **b**	3.61±0.21 **c**
<u>Physico-chemistry</u>			
Dissolved oxygen (mgL^{-1})	5.83±1.56	6.15±1.10	6.31±1.00
Temperature (^0C)	23.87±2.16	23.71±2.19	23.70±2.17
pH	8.28±1.03	8.28±0.26	8.22±0.22
Electrical conductivity ($mScm^{-1}$)	1.79±0.05	1.80±0.05	1.79±0.05
Total ammonia (mgL^{-1})	2.83±1.03	3.50±1.22	3.13±1.19
Carbon dioxide (mgL^{-1})	2.08±0.90	2.16±0.83	2.25±0.86
Chlorides (mgL^{-1})	236.81±7.50	240.45±7.22	243.18±6.43
Sodium (mgL^{-1})	360.00±32.25	367.27±24.12	370.90±38.32
Potassium (mgL^{-1})	7.90±0.53	8.00±0.77	8.90±0.70
Calcium (mgL^{-1})	52.46±15.53	53.92±15.96	42.63±15.75
Magnesium (mgL^{-1})	33.10±18.28	33.34±17.67	31.39±12.15
Total Hardness (mgL^{-1})	263.63±37.22	264.54±32.66	248.18±28.20

(Means with similar letter in a single row are statistically similar at $p<0.05$)

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Table-2. Length weight relationship in fish reared under semi-intensive cultural system.

Fish species	Regression Equation ($\log^{10} Y = a+b \log^{-10} X$)	r	R^2
Treated (Lead Stressed) fish			
Catla catla	Log weight = -5.26+3.445 log fork length	0.987	0.974
	S.E = 0.161**		
Labeo rohita	Log weight = -5.35+3.301 log fork length	0.991	0.982
	S.E = 0.126**		
Cirrhina mrigala	Log weight = -5.89+3.5101 log fork length	0.975	0.950
	S.E = 0.323**		
Catla catla	Log weight = -4.53+2.8651 log total length	0.885	0.731
	S.E = 0.501**		
Labeo rohita	Log weight = -5.69+3.391 log total length	0.992	0.984
	S.E = 0.121**		
Cirrhina mrigala	Log weight = -4.25+ 3.672 log total length	0.969	0.938
	S.E = 0.198**		
Control (Unstressed) fish			
Catla catla	Log weight = -5.04+3.182 log fork length	0.979	0.992
	S.E = 0.189**		
Labeo rohita	Log weight = -4.67+3.00 log fork length	0.992	0.997
	S.E = 0.107**		
Cirrhina mrigala	Log weight = -4.21+2.718 log fork length	0.986	0.995
	S.E = 0.130**		
Catla catla	Log weight = -5.35+3.231 log total length	0.984	0.994
	S.E = 0.168**		
Labeo rohita	Log weight = -4.98+3.039 log total length	0.991	0.996
	S.E = 0.116**		
Cirrhina mrigala	Log weight = -4.27+ 2.670 log total length	0.978	0.992
	S.E = 0.164**		

R^2 = Coefficient of determination
r = Regression coefficient
* = Significant at p<0.05
** = Significant at p<0.01
NS = Non-significant
S.E = Standard Error