



Growth Performance of *Labeo rohita* Under Chronic Dual Exposure of Water-Borne and Dietary Cobalt

Sidra Abbas* and Muhammad Javed

Department of Zoology and Fisheries, University of Agriculture,
Faisalabad-38040, Pakistan

ABSTRACT

In the present study the growth performance of three age groups viz., 60-, 90- and 120-day of *Labeo rohita* has been studied after exposure of fish to water-borne and dietary cobalt concentrations, for 12 weeks. It was found that wet weight of fish was significantly increased, whereas fork and total length did not show any significant change. The 120-day old fish group exhibited higher average wet weight, fork and total length compared with the 90- and 60-day old fish age groups. The three age groups showed higher feed intake than the stressed fish age groups. The feed conversion ratios of the three age groups during controlled conditions were minimum compared to the cobalt stressed media. It was observed that non-significant variations existed for condition factor during the investigation period of 12 weeks among the three age groups of fish. However, the condition factor of stressed fish was significantly lower (1.38 ± 0.45) than observed for un-stressed fish as 1.42 ± 0.39 .

Article Information

Received 31 January 2015
Revised 29 July 2015
Accepted 18 August 2015
Available online 1 January 2016

Authors' Contributions:

SA performed the experiment, analyzed the data and wrote the article. MJ supervised the work and helped in writing the article.

Key words

Growth performance, *Labeo rohita*, age groups, dual exposure, cobalt..

INTRODUCTION

Water is one of the most important natural resources. The quality of water is directly related with the human health (Kumar, 2004). Aquatic environments are susceptible to a wide variety of contaminants that are mainly released from effluents discharged from sewage treatment plants, industries, drainage from agricultural and urban zones and also from other anthropogenic activities (Lopez *et al.*, 2002; Karbassi *et al.*, 2006). That contaminants include organic and inorganic compounds as hazardous wastes, petroleum products, putrescible and combustible material, explosives, phenol, textile dyes and metals. Metals constitute a significant component of inorganic pollutants (Ghosh and Singh, 2005; Gad and Saad, 2008; Jadhav *et al.*, 2010). Due to the chemical stability, as compared to other aquatic organic pollutants, which can be biologically degraded completely, metals are non-biodegradable and hence present severe hazards to the aquatic flora and fauna (Wepener *et al.*, 2001; Begam, 2004; Ambreen *et al.*, 2015). Various metals are natural constituents of the freshwater ecosystems. Some of them are beneficial or even vital for sustaining life but others are toxic to aquatic organisms.

Heavy metals such as cobalt, zinc, lead, cadmium, arsenic, chromium and manganese are present at high concentrations affecting aquatic life, particularly fish

(Patil and Shrivastava, 2003). Heavy metals accumulate in fish produce a biomagnification effect throughout food chain (Khare and Singh, 2002). The fish sensitivity towards different metals decrease with the increase of fish age due to the tendency to accumulate heavy metals that effects on its tolerance limit (Giguere *et al.*, 2004).

The growth of fish is considered as a precise, reliable and sensitive indicator endpoint that is related to the sub-lethal (chronic) exposure of water-borne or dietary metals (Javed and Saeed, 2010). The growth rates in terms of body weight and length, intensity of feeding, feed conversion rates and condition factor are the primary variables that provide information about growth level of fish. However, the most important biological parameter of a fish species is its condition factor that provides information about health of fish and its community (Okgerman, 2005; Richter, 2007; Sarnowski and Jezierska, 2007; Bostock, 2010). Beyond the admissible limits, the uptake of metallic compounds by the fish is potentially toxic and their chronic (sub-lethal) exposures could result into decreased growth rates, and other physiological and behavioural disorders (De Boeck *et al.*, 1997; Vinodhini and Narayanan, 2008; Jadhav *et al.*, 2010). To estimate the chronic (sub-lethal) effects of metals on aquatic organisms fish are used as study model (Wong *et al.*, 2001; Javed and Saeed, 2010).

The metabolic behaviour, breeding, growth and survival of fish are severely affected as a result of heavy metal's exposure to the fish (Adhikari *et al.*, 2009). In nature, cobalt is not found in elemental form but exists in various forms as sulphide ores and in association with arsenic, copper, ferrous and nickel (Awofolu *et al.*,

* Corresponding author: sidraabbas2012@hotmail.com

0030-9923/2016/0001-0257 \$ 8.00/0

Copyright 2016 Zoological Society of Pakistan

2005). Cobalt (a component of vitamin B₁₂) has been recognized as basic element in the fish diet (Davis and Gatlin, 1991). In general, the cobalt concentration is low in fish, presumably due to its high predisposition to organic particles in the solution (Birds *et al.*, 1999). Hence, higher Co concentrations in the aquatic ecosystems may become toxic to the fish (Mukherjee and Kaviraj, 2009). The adverse chronic effects of cobalt intake may occur at concentration of 2.0 mgL⁻¹ (D.W.A.F., 1996). The toxic effects of cobalt on fish results from inhibition of crucial enzymes, displacement of divalent cations in the ion-center of metal-activated enzymes and by the generation of reactive oxygen species in cells resulting into oxidative damage to DNA, proteins and lipids (Simonsen *et al.*, 2012). The cobalt toxicity to fish includes loss of body weight and depressed appetite (Awofolu *et al.*, 2005).

Although, water-borne effects of cobalt on different fish species had been extensively studied in the past, but now-a-days fish are usually exposed simultaneously to water-borne as well as dietary cobalt. Therefore, keeping in view the potential of cobalt as an aquatic contaminant, causing adverse effects on the fish growth in their natural habitats, the present project was planned to investigate the growth performance of fish (*Labeo rohita*) under chronic dual exposure of water-borne and dietary cobalt.

MATERIALS AND METHODS

Experimental design

The growth trial of 12 weeks was conducted in the wet laboratory of Fisheries Research Farms, Department of Zoology and Fisheries, University of Agriculture, Faisalabad. The growth performance of *Labeo rohita* under chronic dual exposure of water-borne and dietary cobalt was investigated. Prior to the experiment, fingerlings of *Labeo rohita* of three age groups *viz.*, 60-, 90- and 120-day were kept under laboratory conditions in cemented tanks of 500-liter water capacity for two weeks, for acclimation. After this acclimation period, fish was transferred into glass aquaria of 70-liter water capacity for growth trials. Ten fish of known weights, fork and total lengths (7.85±0.69g, 81.31±0.34 and 92.05±0.54 mm) were placed in glass aquaria, with three replicates, containing 38.34 mg L⁻¹ of water-borne cobalt (1/3rd of LC₅₀). During 12 weeks of study period, fish were fed with diet (35% Digestible Protein and 2.90 Kcal g⁻¹ Digestible Energy) having sub-lethal (1/3rd of LD₅₀) cobalt concentration as 80.87 µg g⁻¹, to satiation, daily, at 0900 and 1700 h. The un-stressed (control) fish were fed with metal free diet during the whole experimental duration. The stock solution of desired metal

concentration was prepared by dissolving pure CoCl₂. 6H₂O in de-ionized water and feed was prepared by adding sub-lethal metal concentration in the diet. One-third of LC₅₀ (for water-borne) and LD₅₀ (for diet-borne) of cobalt as 38.34 mg L⁻¹ and 80.87 µg g⁻¹, respectively, were considered as sub-lethal levels for *Labeo rohita* as determined by Javed *et al.* (2008). Water were supplied with constant aeration through a pump with capillary system. Water pH (7.0), temperature (30°C), and total hardness (200 mg L⁻¹) were kept constant throughout the study period. Water pH and temperature were determined by using digital meters HANNA HI-8424 and HI-9146, respectively. Total hardness of the test media (water) was measured by following the methods of A.P.H.A. (1998). The wet weights, fork and total lengths of stressed and un-stressed (control) *Labeo rohita* were recorded weekly throughout the experimental period of 12 weeks. At the end of experiment, the average of increase or decrease for wet weights, fork and total lengths, feed intake, feed conversion ratios and condition factor at 60-, 90- and 120-day *Labeo rohita* (both stressed and un-stressed) were determined. Feed conversion ratio (FCR) and condition factor (K) were calculated by using formula (Carlander, 1970).

$$\text{FCR} = \text{Feed intake (g)} / \text{Weight gain (g)}$$

and

$$K = W \times 10^5 / L^3$$

where,

$$W = \text{Wet weight of fish (g)}$$

$$L = \text{Fork length (mm)}$$

Statistical analyses of data

The data obtained on various fish growth parameters were subjected to statistical analysis through Factorial Experiment (RCBD) by following Steel *et al.* (1996) and Tukey's/Student Newman-Keul test was used to compare mean values for various growth parameters. MSTATC and MICROSTAT packages of computer were used for these analyses.

RESULTS

Wet weight, fork and total length increments

Table I shows effect of sub-lethal dual exposure of water-borne (38.34 mg L⁻¹) and dietary (80.87 µg g⁻¹) cobalt concentrations for 12 weeks on wet weights, fork and total lengths, feed intake, feed conversion ratios and condition factor of *Labeo rohita* of three age groups *viz.*, 60-, 90- and 120-days. The initial and final average wet weights, fork and total lengths for 60-, 90- and 120-day age groups of metal stressed and un-stressed *Labeo rohita* are presented in Table I.

Table I. Initial and final average wet weights (g±SD), fork and total lengths (mm±SD) of *Labeo rohita* under sub-lethal dual (water-borne and dietary) cobalt exposure and control conditions.

	<i>Labeo rohita</i>					
	Stressed			Un-stressed		
	60-day	90-day	120-day	60-day	90-day	120-day
Initial average wet weight (g)	5.10±0.34	8.22±0.32	12.31±0.46	4.36±0.32	8.59±0.53	8.53±0.44
Final average wet weight (g)	9.19±0.40	13.04±0.48	21.32±1.45	14.18±0.28	16.40±0.53	16.43±0.47
Initial average fork length (mm)	73.00±4.00	81.39±1.51	95.03±1.02	73.61±2.65	82.42±2.06	82.41±1.43
Final average fork length (mm)	87.89±1.28	99.79±0.83	118.10±1.03	92.50±2.12	104.02±1.26	104.00±1.98
Initial average total length (mm)	82.64±0.87	92.25±1.86	107.74±6.00	82.24±2.48	93.71±2.53	93.71±2.32
Final average total length (mm)	97.54±2.24	111.35±3.92	132.85±2.71	102.82±1.94	116.40±5.26	116.41±2.03

Table II. Average increase in wet weights (g±SD), fork and total lengths (mm±SD) of *Labeo rohita*.

Weeks	Wet weight (g)	Fork length (mm)	Total length (mm)
1	0.11±0.03 e	0.76±0.15 d	0.60±0.08 d
2	0.18±0.04 e	0.98±0.21 cd	1.29±0.11 bcd
3	0.40±0.09 de	1.32±0.44 bcd	1.22±0.18 cd
4	0.47±0.12 cd	1.58±0.48 abc	1.77±0.42 abc
5	0.59±0.16 bcd	1.62±0.53 abc	1.51±0.39 bc
6	0.74±0.25 abc	1.63±0.50 abc	1.79±0.40 abc
7	0.69±0.18 abcd	1.86±0.62 ab	1.81±0.49 abc
8	0.76±0.21 abc	1.82±0.69 ab	2.06±0.68 abc
9	0.75±0.27 abc	1.79±0.25 ab	1.95±0.62 abc
10	0.73±0.14 abc	2.13±0.71 a	2.16±0.82 ab
11	0.98±0.28 a	1.98±0.82 ab	2.09±0.71 abc
12	0.86±0.29 ab	2.26±0.79 a	2.61±1.10 a
Stressed fish	0.50±0.11 b	1.57±0.39 a	1.64±0.43 a
Un-stressed fish	0.71±0.19 a	1.72±0.44 a	1.83±0.52 a

Means with the same letters in a column are statistically similar at $p < 0.05$.

	60-day	90-day	120-day	60-day	90-day	120-day	60-day	90-day	120-day
Stressed	0.34±0.09b	0.40±0.12a	0.75±0.17a	1.24±0.39a	1.53±0.45a	1.92±0.68a	1.24±0.41a	1.59±0.55a	2.09±1.12a
Un-stressed	0.82±0.10a	0.65±0.21a	0.66±0.27a	1.57±0.42a	1.80±0.63a	1.80±0.59a	1.72±0.69a	1.89±0.75a	1.89±0.71a
*Means±SD	0.58±0.22ab	0.53±0.16b	0.71±0.25a	1.41±0.52b	1.67±0.68b	1.86±0.81a	1.48±0.61b	1.74±0.69ab	1.99±0.87a

Means with the same letters in a single *row and column are statistically similar at $p < 0.05$.

+ Av., Average; Inc., Increase.

Table II shows analysis of variance on average increase in wet weight of *Labeo rohita* which shows that there existed highly significant ($p < 0.01$) differences among 12 weeks of the growth trial. The maximal average wet weight increase was observed during 11th week (0.98±0.28 g) of the experiment, while it was minimum during 1st (0.11±0.03 g) and 2nd (0.18±0.04 g) weeks of the experimental period with non-significant difference. During 12-week growth trial, highly significant ($p < 0.01$) variations were observed between stressed and un-stressed (control) fish. However, metal stressed fish exhibited lower average wet weight

increment as 0.50±0.11 g than the un-stressed fish which showed an average wet weight increase of 0.71±0.19 g. The 120-day old fish attained significantly ($p < 0.05$) higher average wet weight increase as 0.71±0.25 g than that of 90-day (0.53±0.16 g) and 60-day (0.58±0.22 g) old fish groups. All the three age groups of un-stressed fish showed significantly better growth in terms of average increase in wet weights than that of stressed fish except for 120-day *Labeo rohita*. The 60-, 90- and 120-day cobalt stressed fish showed average wet weight increments of 0.34±0.09, 0.40±0.12 and 0.75±0.17 g, respectively, whereas average increase in wet weight of

0.82±0.10, 0.65±0.21 and 0.66±0.27 g were shown by 60-, 90- and 120-day un-stressed *Labeo rohita*, respectively. Analysis of variance reveals that there existed statistically highly significant differences among the 12-weeks for the average increase in fork and total lengths of *Labeo rohita*. The maximum average increase in fork length (2.26±0.79 mm) was observed during the last week, followed by that observed during 10th week as 2.13±0.71 mm. However, during the 1st week of the experiment, the minimum average increase (0.76±0.15 mm) in the fork length of fish was observed. The maximum (2.61±1.10 mm) and minimum (0.60±0.08 mm) total length increments were observed during 12th and 1st weeks, respectively of the growth trial. Statistically non-significant differences were observed for average fork and total length increments between cobalt stressed and un-stressed *Labeo rohita*. The average increase in fork and total lengths of three age groups showed statistically significant variations at p<0.05. However, *Labeo rohita* of 120-day age group exhibited maximum fork and total length increments as 1.86±0.81 and 1.99±0.87 mm, respectively than observed for 90-day (1.67±0.68 and 1.74±0.69 mm) and 60-day (1.41±0.52 and 1.48±0.61 mm) age groups, respectively. Average increase in fork lengths of 60-, 90- and 120-day age groups of cobalt stressed *Labeo rohita* were recorded as 1.24±0.39, 1.53±0.45 and 1.92±0.68 mm, respectively, while for 60-, 90- and 120-day un-stressed fish, the same were found as 1.57±0.42, 1.80±0.63 and 1.80±0.59 mm, respectively. The cobalt stressed fish age groups of 60-, 90- and 120-day gained an average increase in total lengths of 1.24±0.41, 1.59±0.55 and 2.09±1.12 mm, respectively, while for the same age groups, average increase in total lengths were recorded as 1.72±0.69, 1.89±0.75 and 1.89±0.71 mm, respectively.

Feed intake, feed conversion ratio (FCR) and condition factor (K)

Table III presents analysis of variance on feed intake, feed conversion ratio (FCR) and condition factor (K) of *Labeo rohita*. Highly significant differences (p<0.01) were observed among 12 weeks of the experimental period for feed intake by the three age groups and between stressed and un-stressed fish. The maximum feed intake was observed as 1.57±0.62 g during 11th week of the experiment, followed by that observed during 8th week as 1.37±0.42 g. The feed intake of un-stressed fish was higher (1.14±0.42 g) as compared to the cobalt stressed fish which was observed as 0.95±0.31 g. However, 120-day *Labeo rohita* exhibited maximum feed intake of 1.25±0.62 g as compared to 90-day (1.01±0.46 g) and 60-day (0.87±0.44 g) age groups. Two age groups viz. 60- and 90-day of un-stressed fish

showed significantly more feed intake than the 60- and 90-day cobalt stressed fish. However, there existed statistically non-significant differences for feed intake between 120-day stressed and un-stressed *Labeo rohita*.

Analysis of variance on feed conversion ratio (FCR) shows highly significant differences among 12 weeks of the experimental duration. The feed conversion ratio was significantly better (2.25±0.41) during 1st week of the experiment, while it was minimum during 12th week of the experiment as 1.60±0.06. There existed highly significant variation for feed conversion ratios between stressed and un-stressed *Labeo rohita*. Both stressed and un-stressed fish exhibited the feed conversion ratios of 2.05±0.62 and 1.71±0.21, respectively. Statistically highly significant (p<0.01) differences were found for feed conversion ratios among the three age groups of fish. The feed conversion ratios for 60-, 90- and 120-day *Labeo rohita* were computed as 1.83±0.52, 2.02±0.31 and 1.78±0.40, respectively. However, the 90-day old stressed and un-stressed fish showed better feed conversion ratios than the other age groups. All the three age groups of un-stressed *Labeo rohita* showed statistically lesser feed conversion ratios than the metal stressed fish age groups.

Condition factor (K) is considered as the degree of fish well-beings. Analysis of variance on condition factor reveals that there existed statistically non-significant differences among the three age groups of *Labeo rohita*, during 12-week experimental period. However, highly significant (p<0.01) variations were found for the condition factor between the cobalt stressed and un-stressed fish. The condition factor of un-stressed fish was significantly higher as 1.42±0.39 than that of metal stressed fish, for which the same was computed as 1.38±0.45.

DISCUSSION

During the dual exposure of water-borne and dietary metal (cobalt) to *Labeo rohita* of three age groups viz. 60-, 90- and 120-day, it was observed that un-stressed fish exhibited maximum growth in terms of wet weight, fork and total length increments as compared to stressed *Labeo rohita*. Statistically highly significant (p<0.01) variations were found between stressed and un-stressed fish for average wet weight increments. These results are also in conformity with the findings of Shafiq *et al.* (2012). They reported statistically significant growth differences between the treated and control *Cirrhina mrigala* under combined (water-borne and dietary) exposure of nickel. The results of present investigation are agree with Kim and Kang (2004), according to whom, copper stress decreased the growth

Table III. Feed intake (g±SD), feed conversion ratio (FCR±SD) and condition factor (K±SD) of *Labeo rohita*.

Weeks	Feed intake (g)	Feed conversion ratio	Condition factor
1	0.22±0.08 f	2.25±0.41 a	1.38±0.02 a
2	0.37±0.07 ef	2.11±0.48 ab	1.36±0.02 a
3	0.70±0.11de	1.85±0.33 cd	1.36±0.01 a
4	0.90±0.18 cd	2.05±0.44 abc	1.36±0.04 a
5	1.09±0.21 bc	1.86±0.35 cd	1.37±0.10 a
6	1.34±0.23 ab	1.92±0.32 bcd	1.40±0.11 a
7	1.24±0.32 abc	1.86±0.29 cd	1.40±0.21 a
8	1.37±0.42 ab	1.82±0.25 cde	1.42±0.21 a
9	1.28±0.39 abc	1.78±0.17 de	1.43±0.34 a
10	1.20±0.31 abc	1.72±0.19 de	1.43±0.35 a
11	1.57±0.62 a	1.69±0.10 de	1.45±0.66 a
12	1.24±0.50 abc	1.60±0.06 e	1.44±0.67 a
Stressed fish	0.95±0.31 b	2.05±0.62 a	1.38±0.45 b
Un-stressed fish	1.14±0.42 a	1.71±0.21 b	1.42±0.39 a

Means with the same letters in a column are statistically similar at p<0.05.

	60-day	90-day	120-day	60-day	90-day	120-day	60-day	90-day	120-day
Stressed	0.65±0.09c	0.86±0.11bc	1.33±0.23a	2.13±0.08a	2.21±0.15a	1.80±0.11b	1.28±0.15a	1.42±0.21a	1.44±0.29a
Un-stressed	1.09±0.12ab	1.17±0.19a	1.16±0.21a	1.53±0.39c	1.82±0.31b	1.76±0.21b	1.38±0.19a	1.44±0.32a	1.43±0.34a
*Means±SD	0.87±0.44b	1.01±0.46b	1.25±0.62a	1.83±0.52b	2.02±0.31a	1.78±0.40b	1.33±0.19a	1.43±0.31a	1.44±0.37a

Means with the same letters in a single *row and column are statistically similar at p<0.05.

rate of *Sebastes schlegeli* (rockfish) and there was an indirect relationship between copper exposure and growth. Hansen *et al.* (2002) exposed *Oncorhynchus mykiss* to sub-lethal cadmium concentration at constant water pH (7.5), temperature (80 °C) and hardness (330 mg L⁻¹). They concluded that, as a result of sub-lethal exposure to fish, reduced growth was observed. Similar findings were reported by Javed (2006) who concluded that average increase in weights of *Labeo rohita*, *Catla catla* and *Cirrhina mrigala* varied significantly as a result of chronic exposure of manganese and nickel. The present results are also in accordance with Shaw and Handy (2001) who also found that dietary (200 mg Kg⁻¹ dry weight) copper exposure to *Oreochromis niloticus* (Nile tilapia) showed significantly lower body weight gains as compared to un-exposed (control) tilapia. During 12-week growth trial, the un-stressed fish exhibited higher average fork and total length increments as compared to the cobalt stressed fish with non-statistical differences between them. Similarly, Hussain *et al.* (2011) also observed that chronic exposure of iron to three fish species *viz.*, *Cirrhina mrigala*, *Catla catla* and *Labeo rohita* showed reduction in fork and total length gains, in comparison to control fish. Giguere *et al.* (2004) suggested that the control fish gained higher fork and total length increments as compared to the stressed fish.

Hayat *et al.* (2007) concluded that exposure of sub-lethal manganese concentrations to major carps result in lower but significant values of fork and total length increments than control fish. The cobalt stress to *Labeo rohita* during present investigation resulted into decreased metabolic rate, hence a reduction in growth parameters (wet weight, fork and total length) was observed. During the present investigation period of 12 weeks, it was observed that 120-day fish gained significantly higher wet weight, fork and total length increments than 60- and 90-day fish age groups. These results are well-supported by the findings of Ameer *et al.* (2013). They concluded that 120-day *Labeo rohita* and *Catla catla* exhibited higher growth patterns (in terms of increase in wet weights, fork and total lengths) than 60- and 90-day fish. It becomes evident that fish sensitivity toward metals decreased significantly with increasing age which is closely related to its metabolism (Abdullah and Javed, 2006; Ansari *et al.*, 2006).

During 12 weeks of study period of each growth trial, the un-stressed *Labeo rohita* exhibited significantly higher feed intake as compared to cobalt stressed fish. It was observed during present investigation that cobalt was toxic to fish as it could generate reactive oxygen species causing oxidative stress in fish. During stressful conditions, the feed intake of fish declines and eventually

reduction in growth performance of cobalt stressed *Labeo rohita* was evident. These results are also in agreement with the findings of Mohanty *et al.* (2009). They determined the effect of copper on feed intake, growth and survival of *Cirrhina mrigala* for 60 days and observed that at all given copper concentrations, the feed intake was significantly reduced in the fish. The sub-lethal (chronic) effects of both lead and zinc to the major carps were investigated by Javed (2012). He concluded that stressed fish exhibited significantly lower values of feed intake than the control fish. Pereira *et al.* (2001) reported significant effect of metals viz. zinc, copper, nickel, chromium, lead and cadmium on the feed intake and growth of *Cirrhina mrigala* when exposed chronically to the medium with these metals. James *et al.* (2003) reported that the feeding parameters were declined with the increasing copper concentration in *Xiphophorus helleri* (ornamental fish). The present results are not supported by Javed (2005) who reported that significantly higher values of feed intake were observed for the major carps under sub-lethal chronic zinc exposures. Statistically highly significant ($p < 0.01$) differences were recorded for feed conversion ratio (FCR) between treatments, during 12-week experimental duration. The results of the present investigation suggested that, as compared to the un-stressed fish, the cobalt stressed fish exhibited significantly higher feed conversion ratio. However, present results are in contradiction with the findings of Ali *et al.* (2002) who recorded significantly minimal feed conversion ratio in *Oreochromis niloticus* when exposed to various (0, 0.15, 0.3 and 0.5 ppm) sub-lethal copper concentrations. Similarly, Dai *et al.* (2008) determined the variations in feed conversion ratio of *Oreochromis niloticus* (tilapia) when exposed to different lead concentrations in diet for a period of 60 days. Their results are in contradiction to the present findings as no significant alterations in feed conversion ratio were observed between metal exposed as well as control fish. Collins *et al.* (2001) investigated the metabolism of copper in juveniles of *Oncorhynchus mykiss* (rainbow trout) and interactions between water-borne and dietary copper uptake. The fish were exposed to elevated dietary concentrations of copper that also revealed reduction in their feed conversion ratios. Similarly, Ameer *et al.* (2013) studied the mixed exposure of dietary and water-borne heavy metals (zinc, cadmium and copper) on the growth of *Catla catla* and *Labeo rohita*. They also concluded that regarding overall responses of control and treated fish, the control fish showed significantly better feed conversion ratio than that of treated one. Sherwood *et al.* (2000) studied the effects of zinc, cadmium and copper on yellow perch and concluded that escalated concentrations of these metals significantly altered the

feed conversion ratio and hence, growth rate of the fish. During present 12-week growth trials, it was found that the un-stressed fish exhibited significantly better condition factor than the fish exposed to water-borne and dietary cobalt. These results are in accordance with the findings of Shafiq *et al.* (2012). They also reported that the fish reared in control medium showed better condition factor as compared to fish treated with mixed sub-lethal water-borne and dietary nickel concentrations. Ali *et al.* (2002) also observed that the condition factor (K) of *Oreochromis niloticus* decreased significantly ($p < 0.05$) with water-borne copper exposure when compared with the un-exposed fish. Similarly, the condition factor derived from weight and length measurements has also often indicated lower condition in polluted fish relative to reference fish that may be due to the heavy metals present at escalated concentrations in the natural contaminated environment (Eastwood and Counture, 2002; Lovesque *et al.*, 2002). However, the present results are against to the findings of Ptashynski *et al.* (2002) who observed variations in the condition factor of adult *Coregonus clupeaformis* (lake whitefish) as a result of dietary nickel exposure and concluded that condition factor values were remained unaffected by the metal's exposure.

CONCLUSIONS

Water-borne and dietary cobalt stress to the *Labeo rohita* revealed a significant impact on its growth in terms of average wet weight, fork and total length increments as well as feed intake and condition factor as compared to the un-stressed fish, during present investigation. However, the feed conversion ratio of stressed fish was found higher as compared to the un-stressed fish. The 120-day fish age group exhibited more growth as compared to the 60- and 90-day *Labeo rohita*, which indicates that with increasing age the sensitivity towards metals toxicity decreased.

Conflict of interest statement

The authors have no conflict of interest to declare.

REFERENCES

- A.P.H.A., 1998. *Standard methods for the examination of water and wastewater*. (20th Ed.). American Public Health Association, New York, pp. 1193.
- Abdullah, S. and Javed, M., 2006. Studies on 96-hr LC₅₀ and lethal toxicity of metals to the fish *Cirrhina mrigala*. *Pak. J. agric. Sci.*, **43**: 180-185.
- Adhikari, S., Ghosh, L., Giri, B.S. and Ayyappan, S., 2009. Bioaccumulation of some heavy metals in tilapia fish relevant to their concentration in water and sediment of

- Wadi Hanifah, Saudi Arabia. *Afr. J. Biotechnol.*, **10**: 254-257.
- Ali, A., Al-Ogaily, S.M., Al-Asgah, N.A. and Gropp, J., 2002. Effect of sublethal concentrations of copper on the growth performance of *Oreochromis niloticus*. *J. appl. Ichthyol.*, **19**: 183-188.
- Ambreen, F., Javed, M. and Batool, U., 2015. Tissue specific heavy metals uptake in economically important fish, *Cyprinus carpio* at acute exposure of metal mixture. *Pakistan J. Zool.*, **47**: 399-407.
- Ameer, F., Javed, M., Hayat, S. and Abdullah, S., 2013. Growth responses of *Catla catla* and *Labeo rohita* under mixed exposure of dietary and water-borne heavy metals viz. Cu, Cd and Zn. *J. Anim. Pl. Sci.*, **23**: 1297-1304.
- Ansari, T.M., Saeed, M.A., Raza, A., Naeem, M. and Salam, A., 2006. Effect of body size on metal concentrations in wild *Puntius chola*. *Pak. J. anal. environ. Chem.*, **7**: 116-119.
- Awofolu, O.R., Mbolekwa, Z., Mtshemla, V. and Fatoki, O.S., 2005. Levels of trace metals in water and sediment from Tyume river and its effects on an irrigated farmland. *Water S. A.*, **31**: 378-389.
- Begam, G., 2004. Carbofuran insecticide induced biochemical alterations in liver and muscle tissues of the *Clarias batrachus* (Linn) and recovery response. *Aquat. Toxicol.*, **66**: 83-92.
- Birds, G.A., Mills, K.H. and Schwartz, W.J., 1999. Accumulation of Co-60 and Cs-134 in Lake Whitefish in a Canadian Shield lake. *Water Air Soil Pollut.*, **114**: 303-322.
- Bostock, J., 2010. Foresight project on global food and farming futures: The application of science and technology development in shaping current and future aquaculture production systems. *J. agric. Sci.*, 1-9. doi:10.1017/S0021859610001127.
- Carlander, D.K., 1970. *Handbook of freshwater fisheries biology*. The Iowa State University Press. Iowa, USA, pp. 293.
- Collins, K., Grosell, M., Higgs, D. and Wood, C.M., 2001. Copper metabolism in actively growing rainbow trout (*Oncorhynchus mykiss*) interactions between dietary and waterborne uptake. *J. exp. Biol.*, **205**: 279-290.
- D.W.A.F., 1996. *Department of water affairs and forestry. South African Water Quality Guidelines (1st Ed.) DWAF*, Pretoria, **8**: 11-15.
- Dai, W., Fu, L., Du, H., Jin, C. and Xu, Z., 2008. Changes in growth performance, metabolic enzyme activities, and content of Fe, Cu and Zn in liver and kidney of tilapia (*Oreochromis niloticus*) exposed to dietary Pb. *Biol. Trace Element Res.*, **128**: 176-183.
- Davis, A.D. and Gatlin, D.M., 1991. Dietary mineral requirements of fish and shrimp. In: *Proceedings of the aquaculture feed processing and nutrition workshop* (eds. D.M. Akiyama and K.H. Tan), American Soybean Association, Singapore, pp. 49-67.
- De Boeck, G., Vlaeminck, A. and Blust, R., 1997. Effects of sub-lethal copper exposure on accumulation, food consumption, growth, energy stores and nucleic acid contents in Common carp. *Arch. environ. Contam. Toxicol.*, **33**: 415-422.
- Eastwood, S. and Counture, P., 2002. Seasonal variations in condition and liver metal concentrations of yellow perch (*Perca flavescens*) from a metal contaminated environment. *Aquat. Toxicol.*, **58**: 43-56.
- Gad, N.S. and Saad, A.S., 2008. Effect of environmental pollution by phenol on some physiological parameters of *Oreochromis niloticus*. *Glob. Vet.*, **2**: 312-319.
- Ghosh, M. and Singh, S.P., 2005. Review on phytoremediation of heavy metals and utilization of its byproducts. *Appl. Ecol. environ. Res.*, **3**: 1-18.
- Giguere, A., Campbell, P.G.C., Hare, L., McDonald, D.G. and Rasmussen, J.B., 2004. Influence of lake chemistry and fish age on cadmium, copper and zinc concentrations in various organs of indigenous yellow perch (*Perca flavescens*). *Can. J. Fish. aquat. Sci.*, **61**: 1702-1716.
- Hansen, J.A., Lipton, J., Welsh, P.G., Morris, J., Cacula, D. and Suedkamp, M.J., 2002. Relationship between exposure duration, tissue residues, growth, and mortality in rainbow trout (*Oncorhynchus mykiss*) juveniles sub-chronically exposed to copper. *Aquat. Toxicol.*, **58**: 175-188.
- Hayat, S., Javed, M. and Razaq, S., 2007. Growth performance of metal stressed major carps viz., *Catla catla*, *Labeo rohita* and *Cirrhina mrigala* reared under semi-intensive culture system. *Pak. Vet. J.*, **27**: 8-12.
- Hussain, S.M., Javed, M., Javid, A., Javid, T. and Hussain, N., 2011. Growth responses of *Catla catla*, *Labeo rohita* and *Cirrhina mrigala* during chronic exposure of iron. *Pak. J. agric. Sci.*, **48**: 225-230.
- Jadhav, J.P., Kalyani, D.C., Telke, A.A., Phugare, S.S. and Govindwar, S.P., 2010. Evaluation of the efficacy of a bacterial consortium for the removal of color, reduction of heavy metals and toxicity from textile dye effluent. *Bioresour. Technol.*, **101**: 165-173.
- James, R., Sampath, K. and Edward, D.S., 2003. Copper toxicity on growth and reproductive potential in an ornamental fish, *Xiphophorus helleri*. *Asian Fish. Sci.*, **16**: 317-326.
- Javed, M., 2005. Growth responses of *Catla catla*, *Labeo rohita* and *Cirrhina mrigala* for bio-accumulation of zinc during chronic exposure. *Pak. J. biol. Sci.*, **8**: 1357-1360.
- Javed, M., 2006. Studies on growth responses of fish during chronic exposures of nickel and manganese. *Pak. J. biol. Sci.*, **9**: 318-322.
- Javed, M., 2012. Effects of zinc and lead toxicity on the growth and their bioaccumulation in fish. *Pak. Vet. J.*, **32**: 357-362.
- Javed, M., Abdullah, S. and Yaqub, S., 2008. *Studies on tissue-specific metal bioaccumulation patterns and differences in sensitivity of fish to water-borne and dietary metals*. First annual report of HEC project No. 803 (R&D), pp. 89.

- Javed, M. and Saeed, M.A., 2010. Growth and bioaccumulation of iron in the body organs of *Catla catla*, *Labeo rohita* and *Cirrhina mrigala* during chronic exposures. *Int. J. Agric. Biol.*, **12**: 881-886.
- Karbassi, R., Bayati, I. and Moattar, F., 2006. Origin and chemical partitioning of heavy metals in riverbed sediments. *Int. J. environ. Sci. Tech.*, **3**: 35-42.
- Khare, S. and Singh, S., 2002. Histopathological lesions induced by copper sulphate and lead nitrate in the gills of fresh water fish *Nandus nandus*. *J. Ecotoxicol. environ. Monit.*, **12**: 105-111.
- Kim, S.G. and Kang, J.C., 2004. Effect of dietary copper exposure on accumulation, growth and hematological parameters of the juvenile rockfish, *Sebastes schlegeli*. *Marine environ. Res.*, **58**: 65-82.
- Kumar, A., 2004. *Water pollution*. A.P.H Publishing Corporation, New Delhi, pp. 199.
- Lopez, A.M., Benedito, J.L., Miranda, M., Castillo, C., Hernandez, J. and Shore, R.F., 2002. Interactions between toxic and essential trace metals in *Catla catla* from a region with low levels of pollution. *Arch. environ. Contam. Toxicol.*, **42**: 165-172.
- Lovesque, H.M., Moon, T.W., Campbell, P.G.C. and Hontela, A., 2002. Seasonal variation in carbohydrate and lipid metabolism of yellow perch (*Perca flavescens*) chronically exposed to metals in the field. *Aquat. Toxicol.*, **60**: 257-267.
- Mohanty, M., Adhikari, S., Mohanty, P. and Sarangi, N., 2009. Role of waterborne copper on survival, growth and feed intake of Indian major carp, *Cirrhinus mrigala* Hamilton. *Bull. environ. Contam. Toxicol.*, **82**: 559-563.
- Mukherjee, S. and Kaviraj, A., 2009. Evaluation of growth and bioaccumulation of cobalt in different tissues of common carp, *Cyprinus carpio*, fed cobalt-supplemented diets. *Acta Ichthyol. Pisc.*, **39**: 87-93.
- Okgerman, H., 2005. Seasonal variations in the length-weight relationship and condition factor of rudd (*Scardinius erythrophthalmus* L.) in Sapanca Lake. *Int. J. zool. Res.*, **1**: 6-10.
- Patil, P.R. and Shrivastava, V.S., 2003. Metallic status of river Godavari. A Statistical Approach. *Indian J. environ. Prot.*, **23**: 650-61.
- Pereira, L., Countinho, C. and Rao, C.V., 2001. Toxic effects of sewage sludge on freshwater edible fish *Cirrhina mrigala*. *Bull. environ. Contam. Toxicol.*, **56**: 467-474.
- Ptashynski, M.D., Pedlar, R.M., Evans, R.E., Baron, C.L. and Klaverkamp, J.F., 2002. Toxicology of dietary nickel in lake whitefish (*Coregonus clupeaformis*). *Aquat. Toxicol.*, **58**: 229-247.
- Richter, T.J., 2007. Development and evaluation of standard weight equations for bridgelip sucker and large scale suckers. *N. Am. J. Fish. Manage.*, **27**: 936-939.
- Sarnowski, P. and Jezierska, B., 2007. A new coefficient for evaluation of condition of fish. *Electron. J. Ichthyol.*, **2**: 69-76.
- Shafiq, A., Abdullah, S., Javed, M., Hayat, S. and Batool, M., 2012. Effect of combined exposure of water-borne and dietary nickel on the growth performance of *Cirrhina mrigala*. *J. Anim. Pl. Sci.*, **22**: 425-430.
- Shaw, B.J. and Handy, R.D., 2001. Dietary copper exposure and recovery in Nile tilapia, *Oreochromis niloticus*. *Aquat. Toxicol.*, **76**: 111-121.
- Sherwood, G.D., Rasmussen, J.B., Rowan, D.J., Brodeur, J. and Hontela, A., 2000. Bioenergetic costs of heavy metal exposure in yellow perch (*Perca flavescens*): in situ estimates with a radiotracer (¹³⁷Cs) technique. *Can. J. Fish. aquat. Sci.*, **57**: 441-449.
- Simonsen, L.O., Harbak, H. and Bennekou, P., 2012. Cobalt metabolism and toxicology-A brief update. *Sci. Total Environ.*, **432**: 210-215.
- Steel, R.G.D., Torrie, J.H. and Dinkkey, D.A., 1996. *Principles and procedures of statistics* (3rd Ed.). McGraw Hill Book Co., Singapore, pp. 627.
- Vinodhini, R. and Narayanan, N., 2008. Bioaccumulation of heavy metals in organs of fresh water fish, *Cyprinus carpio* (Common carp). *Int. J. environ. Sci. Technol.*, **5**: 179-182.
- Wepener, V., Van-Vuren, J.H.J. and Du-Preez, H.H., 2001. Uptake and distribution of a copper, iron and zinc mixture in gill, liver and plasma of a freshwater teleost, *Tilapia sarrmanii*. *Water S. A.*, **27**: 99-108.
- Wong, C.K.C., Yeung, H.Y., Woo, P.S. and Wong, M.H., 2001. Specific expression of cytochrome P4501A1 gene in gill, intestine and liver of tilapia exposed to coastal sediments. *Aquat. Toxicol.*, **54**: 69-80.