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Assessment of heavy metals bioaccumulation in alien fish species, *Cyprinus carpio* from the Gomti river, India

Ashish Tiwari[1] and Amitabh Chandra Dwivedi[2]

[1]Bharghava Laboratory, Department of Botany, University of Allahabad, Allahabad, India
[2]Regional Centre, Central Inland Fisheries Research Institute (ICAR), 24 Panna Lal Road Allahabad, India

ABSTRACT

The paper deals the assessment of bioaccumulation of four heavy metals (Cu, Cr, Pb and Zn) in Cyprinus carpio fish species and water from the Gomti river at Lucknow, India during 2011-2012. The heavy metals (Cu, Cr, Pb and Zn) were assayed using Atomic Absorption Spectrophotometer and the results were given as μg/g dry weight. The data obtain after water analysis reflected that the order of occurrence of heavy metals to be Zn > Pb > Cr > Cu > Cd. The analysis of heavy metals in muscle of C. carpio was measured in order of Zn > Cr > Pb> Cu, in case of gill Pb >Cr> Zn > Cu and in case of liver Pb > Cr > Cu > Zn. The bioaccumulation factor of the heavy metals was computed as in muscle Cu (24.5), Cr (35), Pb (22), Zn (20.66), in gill Cu (34), Cr (41.66), Pb (89.33) and Zn (9.25), whereas in case of liver Cu (34.5), Cr (338.33), Pb (36.8), Zn (28.16). The presence of heavy metal in our environment has been of great concern because of their toxicity when their concentrations are more than the permissible level.

Keywords: Bioaccumulation, *Cyprinus carpio,* Gomti river, Muscle, Gill and Liver.

INTRODUCTION

Fishes are major part of the human diet due to rich protein content, low saturated fat and sufficient Omega-3 fatty acid which are known to support good health. *C. Carpio* is an exotic fish species for India. It is commercially exploited with (14.20%) in the Ganga river (largest river in India) at Allahabad [1].

The pollution of the aquatic environment with heavy metal has become a worldwide problem, because they are indestructible and most of them have toxic effects on organism [2]. Fishes have been widely used as bio-indicator of pollution by metals. Heavy metals from manmade pollution sources are continually released into aquatic systems, and they are a serious threat because of their toxicity, long persistence, bioaccumulation and biomagnifications in the food chain [3]. The natural aquatic systems have been extensively be contaminated with heavy metals released from domestic, industrial and other manmade activities [4]. Among fishes are the inhabitants that cannot escape from the detrimental effects of these pollutants [5]. There are two main routes of heavy metals exposure [6] the primary route of intake of these chemicals in fish species is via gills or transport of dissolved contamination water across biological membranes and ionic exchange etc. 2) the secondary route is through ingestion of food or sediment particles with subsequent transport across the gut. Gastrointestinal route is the most important route of uptake [7].
Heavy metals are known to distort the structural or biological functions of biomolecules [8]. Since metals act as endocrine disruptors, they can interfere with metabolism, synthesis and transport of hormones or receptors [9, 10]. The bioaccumulation of trace elements in living organisms and biomagnifications in them describes the processes

and pathways of these (possible) pollutants from one tropic level to another, exhibiting the higher bioaccumulation ability in the organisms concerned. Increasing concentration through the food chain caused higher retention time of toxic substances than that of the other normal food components. Therefore, various fish species are ideally used as bio-indicators of metal contamination [11]. The objective of the present study was to determine the levels of certain heavy metals in Gomti river water (at Lucknow) and muscle, gill and liver of *Cyprinus carpio*.

MATERIALS AND METHODS

Sampling procedures

Present study was conducted in the Gomti river at Lucknow, India. The Gomti river is a left bank tributary of the Ganga river. A consecutive seasonal (summer, monsoon and winter) work was carried out for the collection of water and fish samples in the year 2011- 2012. Water samples were collected from the Gomti river at Naya ghat, Lucknow (Latitude 26^0 52' 22.58'' and Longitude 80^0 54' 58.40'') in 500 ml polyethylene bottles fitted with polyethylene caps. The collected samples were immediately acidified with 10 ml of 6N HNO3 on the sampling spot. Fish samples were collected in the 11.00 to 12.00 am with the help of local fishermen. In case of fish samples, size of fishes varied from 10 to 30 cm. Fishes were transported to the laboratory in the ice box on same day. All the samples were brought to the laboratory and were stored at 4ºC in the refrigerator for further analysis.

Preparation of samples for analysis of heavy metals in water and fish

For estimation of heavy metals in water, 500 ml of water sample (pre acidified) was filtered and subjected to nitric acid digestion. For digestion, 5 ml of 6N HNO_3 was added to 100 ml of the sample followed by heating to slow boil till the digestion was completed which was indicated by a light coloured clear solution. The digested samples were used for metal estimation using Atomic Absorption Spectrophotometer (MODEL No. SL 173, ELICKO, India) as per [12]. The heavy metals were estimated i.e. Copper (Cu), Chromium (Cr), Lead Pb) and Zinc (Zn).

The tissues were homogenized and approximately 10gm of the homogenate then digested as follows 1.0 gm of the powdered samples (muscle, liver and gill) were placed in a 100 ml round bottom flask with ground glass joint and mineralized under reflux using a mixture of 6 ml HNO_3, 2.0 ml HClO and 4 ml H_2O_2 . The digestion is done with help of water bath .The digestion procedure lasted for about 6 h to obtain a clear solution. The digests were prepared in triplicate and carefully transferred with their respective washing into a 25 ml volumetric flask and diluted to volume. The digests were then analyzed for Cu, Cr, Pb and Zn, using an atomic absorption spectrophotometer (MODEL No. SL 173, ELICKO, India) with aqueous calibration standards prepared from the stock standard solutions of the respective elements as was reported by [13].

Bioaccumulation Factor (BAF)

Bioaccumulation Factor is defined as the ratio of the concentration of a specific heavy metal in the organism to the concentration of the metal in the water [14]. The bioaccumulation of all four heavy metals in different samples of the Gomati river was calculated as follows:

RESULTS AND DISCUSSION

Evaluation of physico-chemical properties of water from the Gomti river

The maximum temperature was recorded in the summer and minimum in winter season. pH, Cl, BOD, Cu, Cr, Zn and Pb were observed maximum in summer season (Table 1). SO_4^{--}, Nitrate, DO and COD were recorded maximum in the monsoon season. DO, COD, SO_4^{--} and NO_2^{--} values of selected site showed that water was highly contaminated with sewage and industrial effluents which coming from the city/industry in to the river directly. The recorded ph values were ranging from 7.9 to 8.4 indicated that the present water samples were slightly alkaline in nature in summer season. Other parameters were varied from (2.5- 4.5 mg/l), (10-15 mg/l), (32-46 mg/l), (48-91 mg/l), (1.4-1.8mg/l) and (8-26 mg/l) in case of DO, BOD, COD, So^{-4}, No^{2-} and Cl^-, respectively.

All observed values shown that concentration of heavy metal higher in summer season because low volume of water in the river. Lower concentrations of heavy metal were recorded in monsoon season due to higher volume of water in the river. In present study, heavy metals concentration in water (river Gomti) varied from season to season. Average concentrations were measured of Cu 0.02 mg/l, Cr 0.03mg/l, Pb 0.03 mg/l and Zn 0.07 mg/l. Ranged of Cu from (0 to 0.05 mg/l), Cr (0 to 0.06 mg/l), Cd (0-0.01 mg/l), Pb (0.02 to 0.07 mg/l) and Zn 0 to 0.18 mg/l (Table 1)

Heavy metals concentration in different organs of *C. carpio*

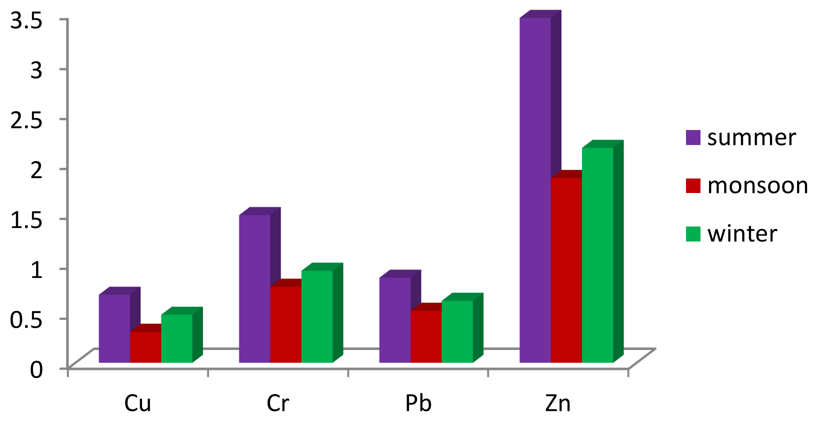
Considerable variations in the heavy metals were observed in all the samples, with minimum accumulation during monsoon season and maximum in summer season in all fish samples and organs (muscle, gill, liver). The order of heavy metals accumulation were found to be in muscle Zn > Cr > Pb > Cu, in gill Pb > Cr > Zn > Cu and in liver Pb

> Cr > Cu > Zn. In case of muscle, Cu, Cr, Pb and Zn were varied from (0.31 to 0.68 µg/g), (0.76 to 1.48 µg/g), (0.52 to 0.85 µg/g) and (1.85 to 3.45 µg/g), respectively (Fig. 1). The average value of Zn was recorded maximum in muscle with 2.48 ±0.85 µg/g. In case of gill, heavy metal accumulation were ranged from Cu (0.53-0.88 µg/g), Cr (0.82-1.92 µg/g), Pb (1.72-3.98 µg/g) and Zn (0.81-1.58 µg/g) (Fig. 2). The average value of Pb was recorded highest in gill with 2.68 ±1.16 µg/g. In case of liver Cu (5.21-8.25 µg/g), Cr (7.05-12.71 µg/g), Pb (9.13- 14.05 µg/g) and Zn (2.15- 4.95 µg/g,) were observed (Fig. 3). The average value of Pb was observed maximum in liver with 11.07 ±2.61 µg/g. The liver accumulates relatively higher amount of metals. The higher accumulation in liver may alter the levels of various biochemical parameters in liver.

Table :1 Physico-chemical properties of water from the Gomti river

Pa Parameters	Year 2011-2012				
	Summer	Monsoon	Winter	Average ± Sd	Permissible limit WHO 1993/2006 [39, 40]
Temperature (C^0)	28	22	16	22.00 ± 7.11	--
pH	8.4	7.9	8.1	8.13 ± 0.25	6.5-8.5 WHO 1993
SO^{--}_4 mg/l)	55	91	48	63.00 ± 24.26	500 WHO 1993
Cl^- (mg/l)	26	8	22	18.67 ± 9.45	250 WHO 1993
Nitrate (mg/l)	1.5	1.8	1.4	1.56 ± 0.20	50 WHO 2006
DO (mg/l)	4.5	2.5	5	4 ± 1.04	NA
BOD mg/l)	12	15	10	12.33 ± 2.51	NA
COD/(mg/	32	35	28	38.33 ± 7.09	NA
Cu (mg/l)	0.05	0	0.02	0.02 ± 0.01	2 WHO 2006
Cr (mg/l)	0.06	0	0.03	0.03 ± 0.02	0.05 WHO 2006
Cd(mg/l)	0.02	BDL	0.01	0.01 ± 0.00	0.003 WHO 2006
Zn (mg/l)	0.18	0	0.10	0.09 ± 0.06	3 WHO 2006
Pb (mg/l)	0.05	0.02	0.03	0.03 ± 0.01	0.01 WHO 2006

Fig. 1 Accumulation of heavy metals in muscle of *Cyprinus carpio*



Cu metal distribution pattern was different from muscle, gill and liver. Cu concentration was reported maximum in liver (8.25 µg/g) in summer season and minimum for muscle (0.31 µg/g) in monsoon season. Average concentration of Cu was measured 0.49±0.18 µg/g in muscle, 0.68±0.18µg/g in gill and 6.90±1.56µg/g in liver. Cr concentration was observed highest 12.71 µg/g in liver in summer season and minimum for muscle 0.76 µg/g in monsoon season. Average concentration of Cr was recorded (1.05±0.37 µg/g) in muscle, (1.25±0.58 µg/g) in gill and (10.15±2.86 µg/g) in liver. Bottom dwelling fish *C. carpio* is found exhibit higher concentration.

The maximum accumulation of Pb was recorded 14.05µg/g in liver in summer season and minimum 0.52 µg/g in muscle in monsoon season. Average concentration of Pb was observed 0.66±0.37 µg/g in muscle, 2.68±1.16 µg/g in gill and 11.07±2.61 µg/g in liver. The statistical analysis of Pb in gill and liver and water showed higher mean values above the detection limits. Pb maximum accumulate in liver as compared to gill and muscles. The maximum accumulation of Zn was observed 4.95µg/g in liver in summer season and minimum for gill 0.81 µg/g in monsoon season. The result of present study was found that average concentration of Zn (2.48±0.85 µg/g) in muscle, (1.11±0.41 µg/g) in gill, (3.38±1.34 µg/g) in liver. Zn accumulation was observed maximum in liver as compared to muscle and gill.

Fig. 2 Accumulation of heavy metals in gill of *Cyprinus carpio*

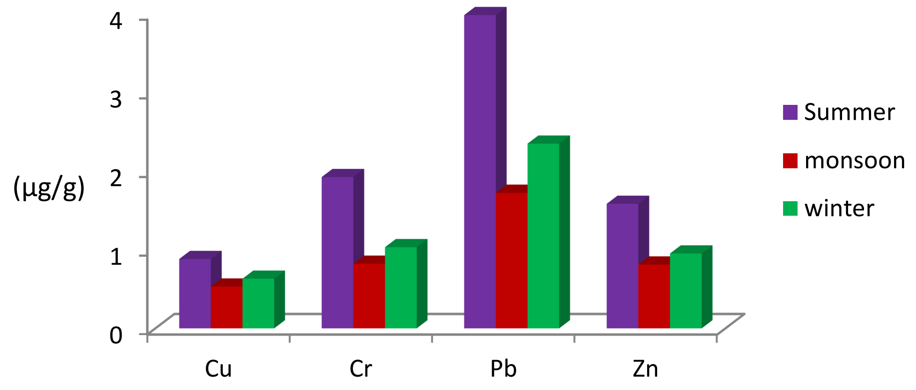
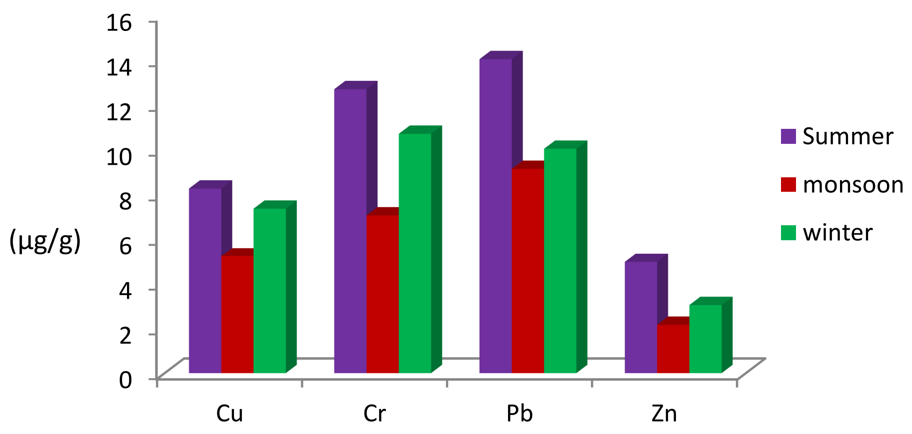


Fig. 3 Accumulation of heavy metals in liver of *Cyprinus carpio*



The results of our present study show that there is no threat of any hazard at present except Pb, because it crosses permissible limit of WHO. However, their increasing accumulating tendency in water, muscles, gill and liver of the fish species indicates that a constant monitoring of this river is needed before the level cross its threshold and become toxic to the aquatic animals and their predators including humans.

Assessment of heavy metals through bioaccumulation factor (BAF) in *C. carpio*

The bioaccumulation factor of the heavy metals was computed as in muscle Cu (24.5), Cr (35), Pb (22), Zn (20.66), in gills Cu (34), Cr (41.66), Pb (89.33) and Zn (9.25), whereas in liver Cu (34.5), Cr (338.33), Pb (36.8), Zn (28.16). The BAF of all metals were maximum reported in liver.

During rainy season rivers are heavily flooded and the drainage system in drastically affected which result in mixing of polluted and unpolluted waters. This leads to decrease in heavy metal concentration whereas increase in the concentration of metals during summer season could be due to drought and decrease in water level [15]. [16] stated that the changes in the physico-chemical parameter of water such as temperature, Ph, BOD, COD, presence of chelating materials, salinity, rainfall, climatic changes etc, all effect the presence of heavy metals in the aquatic environment. The heavy metals accumulated predominantly in gill, liver, intestine and kidney [17, 18].

Target organs, such as liver and gill have tendency to accumulate heavy metals in high values. [19] reported that the Cu ranged in muscle (6-42 µg/g) in *C. Carpio* from the tank at Tumar (India). [20] reported that Cu in muscle (255 ± 303 µg/g), in gill (159± 44µg/g) and in liver (390±13.5µg/g) of *C. carpio*. [21] reported Cu median 0.217, mean 0.237. [22] stated that the accumulation of bioactive metals as like Copper and Zinc were mainly controlled by the fish through various metabolic processes and the level of accumulation usually independent of ambient concentrations. [19] observed higher concentration of Pb in muscle (0.8 µg/g) of *C. carpio* from the tank at Tumar (India). [23] reported that Pb in muscle (1.93 ± 0.79 µg/g), in gill (2.89 ± 1.64 µg/g) and in liver (7.91 ± 3.08 µg/g) in *C carpio* from the Gomti river at Jaunpur, India. [21] reported that Pb in muscle median 0.217 and mean 0.237. [24] reported that the bio-accumulation of Pb+2 in fish body and oxygen requirement of fish increased with concomitant increase in metal concentration. Fish have been reported to be very much sensitive to Pb, its uptake, increases with increasing concentration in the environment [25].

In the present study muscle accumulated the least metals burdens as compared to the other organs. [19] reported that Cr ranged in muscle (0.004 to 2 µg/g) *C. Carpio* from the tank at Tumar (India). [26] reported that Cr average concentration 1.02µg/kg in muscle, 4.56µg/kg in gill and 2.45µg/kg in liver. The liver is an active site of metal metabolism and plays a protective role by acting as a storage site [27]. Muscle is the major tissue of interest under routine monitoring of environmental contamination with metals. In the present study muscle accumulated very minute compared to other organs which is in accordance with the findings of [28] and [29]. [30] reported that Zn in muscle (30.31 ± 4.16 mg/g) and in Gill (27.85 ± 3.93 mg/k). Zn Accumulation in fish results in their mortality, growth retardation and destroys gill epithelium causing hypopxiya ([31, 32]. High concentration of Pb and Cr in the fish tissue such as liver, muscles and gills of common carp (*C. carpio*) has been reported specially in area close to industries ([33, 34].

Most laboratory research on the bioaccumulation of heavy metals suggests that no single mechanism is responsible for metal uptake in aquatic systems. The accumulation of a particular metal depends to a large degree on the presence of the metal in the water column. In the present study, however, it was interesting to note that although average total concentration (mg /l) of Zn in the water sample was highest, its BAF value in C. Carpio muscle placed it in the third position, first in gill and liver among the heavy metals. Pb accumulated first position in water and muscle and fourth in gill and liver. This means that some heavy metal accumulation in fish could depend not only on the concentration of the metal in the water column but also on the abiotic features of the environment (eg., pH, Redox potential alkalinity and salinity), and the duration of exposure [35, 36, 37, 38].

The BAF for different heavy metals from water to fish is a key component of human exposure to the metals via the food chain. The highest BAF value in present study was for Cr. Overall, the BAF values for Cu, Cr, Pb and Zn were all significant, supporting the finding that the accumulation of Pb and Zn is comparatively more, while that of Cu and Cr were less in fish.

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