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RESEARCH

Implications of Heavy Metal Accumulation in Fish Feed, Water, Sediment, and Different Fish Species in a Polyculture System

Allah Bachayo Rajar[1] · Zainab Malik[2] · Javed Ahmed Ujan[3,4] · Khalid Hussian Rind[5] · Raqeeb Ullah[6] · Saira Naz[7,8] · Mujeeb Ullah[9] · Muhammad Zahid[9] · Khalid Khan[9] · Khayyam Khayyam[9] · Salim S. Al-Rejaie[10] · Mohamed Mohany[10]

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Abstract

Heavy metal bioaccumulation in organisms is primarily a result of dietary uptake. The current study examines the concentrations of heavy metals (Pb, Cd, Cr, and Cu) in fish feed, water, sediment, and three fish species (*Catla catla*, *Labeo rohita*, and *Cyprinus carpio*) from different feeding zones in a polyculture pond system. Furthermore, associated human health risks were also evaluated. The fish samples ($n = 25$ for each species) were collected from 10 different fish ponds in the Kohat district, Pakistan. Heavy metals were determined using an atomic absorption spectrometer. Results revealed higher concentrations of heavy metals in sediment, followed by water. However, the concentration of heavy metals in fish feed was lower than the standard limits. In the case of fish, the bottom feeder (*C. carpio*) notably exhibited higher ($P < 0.05$) levels of heavy metals than the column feeder (*L. rohita*) and surface feeder (*C. catla*) fish. Moreover, in the liver of all fish species, the bioaccumulation of heavy metals was higher, followed by the gills. Principal component analysis (PCA) demonstrated a strong correlation of heavy metals in *C. carpio* gills, flesh, feed, and pond water, while the heavy metals in the liver correlated with the detected metals in sediment. The human health risk analysis shows that bottom feeder fish had higher estimated daily intake (EDI), target hazard quotient (THQ), and hazard index (HI) values (> 1). Consequently, the exposed population may experience adverse health effects. The findings of this study suggest that the bottom feeder (*C. carpio*) bioaccumulates a higher concentration of heavy metals than column (*L. rohita*) and surface feeder (*C. catla*) in the polyculture system.

Keywords Bioaccumulation · Heavy metals · Bottom feeder, Fish feed · Column feeder · Surface feeder

✉ Saira Naz
saira.zoologist1991@gmail.com

1 Muhammad Medical College Mirpurkhas/Ibn-e-Sina University, Mirpurkhas 69012, Sindh, Pakistan

2 Department of Zoology, Wildlife and Fisheries, University of Agriculture Faisalabad, Faisalabad 38040, Punjab, Pakistan

3 Department of Zoology, Shah Abdul Latif University Khairpur, Khairpur 66020, Sindh, Pakistan

4 Department of Animal Sciences, University of Florida, Gainesville, FL 32608, USA

5 Department of Molecular Biology and Genetics, Shaheed Benazir Bhutto University, Shaheed Benazirabad 67450, Sindh, Pakistan

6 Department of Zoology, Bacha Khan University Charsadda, Charsadda 24540, Khyber Pakhtunkhwa, Pakistan

7 Centre for Research on Fish Nutrition and Environmental Ecology of the Ministry of Agriculture, Shanghai Ocean University, Shanghai 201306, China

8 Department of Zoology, University of Lahore, Sargodha 40100, Punjab, Pakistan

9 Department of Zoology, Islamia College University Peshawar, Peshawar 25120, Khyber Pakhtunkhwa, Pakistan

10 Department of Pharmacology and Toxicology, College of Pharmacy, King Saud University, P.O. Box 55760, 11451 Riyadh, Saudi Arabia

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Introduction

Aquaculture is a major source of animal protein [1]. Countries with overpopulation problems, such as Pakistan, need to produce more protein from fish [2]. Thus, it is imperative to maximize the yield of all resources that are usable for food production. Increased aquaculture facilities and the expansion of cultured fish ponds can facilitate these efforts [3].

The rapid development of the industrial sector has led to a significant amount of heavy metal pollution in the air, water, sediment, and feed, which constitute major environmental concerns globally [4, 5]. Furthermore, heavy metal pollutants may enter the ecosystem through anthropogenic activities, such as the disposal of sewage sludge and the application of inorganic fertilizers and pesticides [6]. In recent decades, there has been a growing concern about the contamination of freshwater and commercial feeds with several pollutants like heavy metals because they not only pose a threat to public water supplies but also harm aquatic life [7, 8]. As the level of heavy metal contamination increases, the growth of aquatic organisms is reduced, and the ecological balance of the aquatic environment is disrupted [9].

Aquatic ecosystems are widely evaluated using fish because pollutants accumulate in the food chain and cause adverse effects and loss [10]. Studies on diverse fishes have revealed that heavy metals alter their physiology, tissue, and blood biochemical parameters [11, 12]. Several studies have been conducted on the toxic effects of heavy metals, including the bioaccumulation of these metals [13, 14].

The habitats of aquatic organisms and their feeding zones determine the extent of heavy metal accumulation in them [15]. Soil organic carbon, total nitrogen, and total phosphorus play interconnected roles in water pollution and the transfer of heavy metals [11, 16]. Their dynamics are influenced by various environmental factors and human activities [9, 17]. High metal concentrations in the environment enhance the accumulation of metals in aquatic organisms [15]. The food chain can contribute to transferring heavy metals to organisms of high trophic levels, which may threaten human health [9, 18]. Metal-contaminated fish can adversely affect human health, and the toxic effects of metal-contaminated fish can extend to other organs or systems, making them even more dangerous for humans [19].

Fish are particularly vulnerable to pollution in the aquatic environment because they feed and live in an environment polluted by contaminants [20]. Various metals (cadmium, chromium, lead, nickel, copper, mercury, zinc, and arsenic) accumulate in fish bodies, depending on the species, location, temperature, and size [21, 22]. In response to exposures, such as diet or exposure to a higher metal level in their surrounding environment, their tissues and organs accumulate metals [23]. The quality of fish feed is a crucial aspect of aquaculture practice. Aquaculture systems that are intensive or semi-intensive require nutritionally balanced fish feeds [24]. A well-balanced commercial feed is essential for successful aquaculture production [25]. Different types of fish feed and water sources are used to produce fish. Certain heavy metals are a significant source of pollution from these sources [26]. Studies have been limited to evaluating heavy metal levels in fish species based on their feeding zones in polyculture systems. In the current study, we hypothesized that heavy metals bioaccumulation in fish depends on different feeding zones and habits. Therefore, we measured the contents of heavy metals (Pb, Cd, Cr, and Cu) in water, fish feed, sediment, and tissues of fish species (*C. carpio*, *L. rohita*, *and C. catla*) fed in different zones in a polyculture system. Then, the concentration and bioaccumulation of heavy metals in the different fish tissues were compared, and the human health risks were assessed.

Materials and Methods

Study Site

The study site was located in Kohat district in the southern region of the province (Khyber Pakhtunkhwa), Pakistan (Fig. 1). The total area of the district is 2973 km^2 with geographical coordinates 33.4973° N, 71.5249° E. In Pakistan, it ranks 35th in terms of population and fourth in Khyber Pakhtunkhwa. It is estimated that the district has a population of more than 562,644 residents, according to the Khyber Pakhtunkhwa survey report. Three major water reservoirs (Tanda, Gandiali, and Kandat) can be found in the area. The climate at the site is hot and semi-arid. May to September are the hottest months in Kohat and the surrounding area. The site has a harsh and cold winter. Guava is the most popular fruit in the country and a major crop of the site. Rainfall occurs throughout the year in the area. Monsoon rains occur from May to October. In August, the area receives the most rain, averaging 114 mm. There are three main crops grown in the district: wheat (*Triticum aestivum*), maize (*Zea mays*), and barley (*Hordeum vulgare*); the area is also known for the production of several vegetables. Tube wells are used to irrigate most agricultural lands. Warm-water fish farming is a common practice on the site. Mostly, the fish are cultured in earthen ponds in polyculture form. The commonly cultured fish species are carp (major Indian carp), Nile tilapia, and catfish.

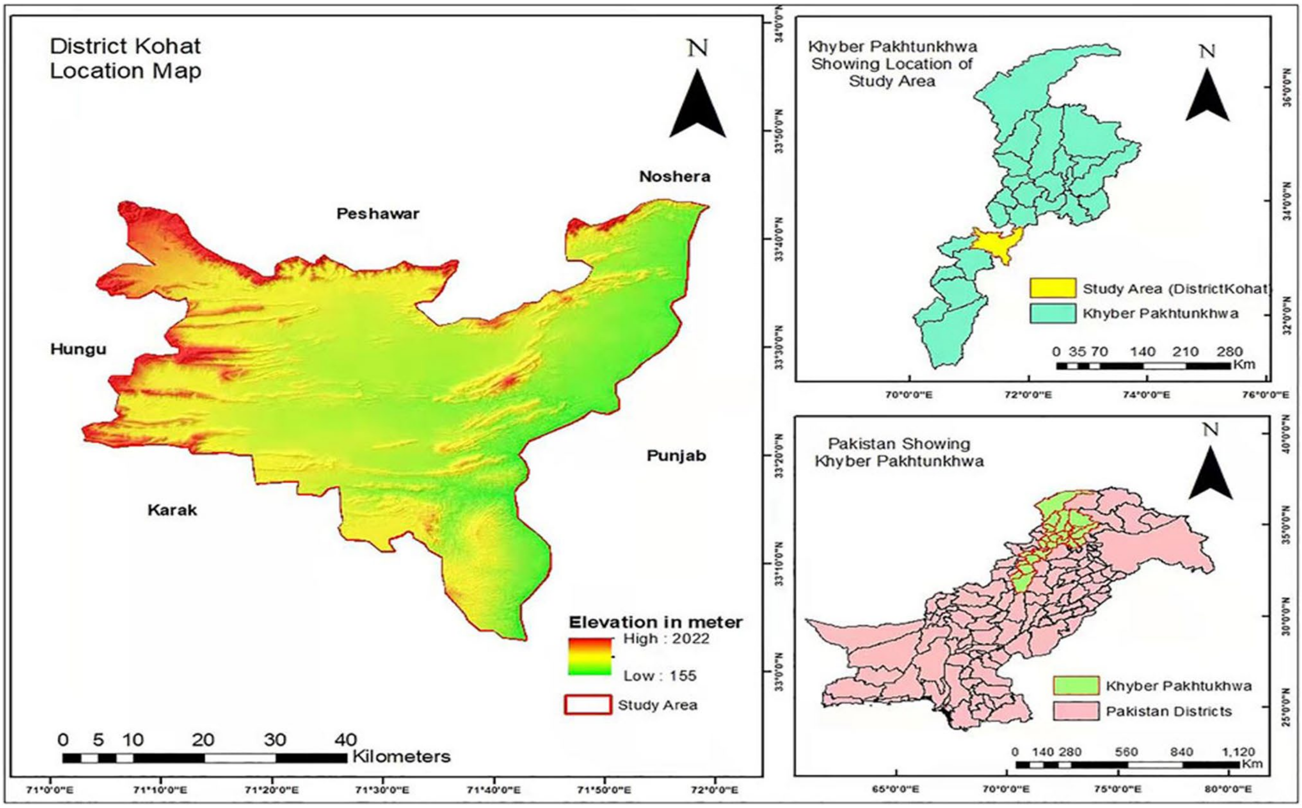

Fig. 1 Modified map of District Kohat Khyber Pakhtunkhwa, Pakistan (adopted from Tasleem et al. [20])

Sample Collection and Preparation

All the fish (*C. catla*, *L. rohita*, and *C. carpio*) samples ($n = 25$ for each species) were collected from 10 different earthen ponds in district Kohat where the fishes were cultured in polyculture form having different feeding zones (*C. catla*: surface feeder, *L. rohita*: column feeder, and *C. carpio*: bottom feeder). Fish species were caught by using a cast net. The weight and length of sampled fish were measured using an electronic weighing balance (model name: Newtech) and measuring tape made up of fiberglass. The mean weight (g) and length (cm) of *C. catla* were approximately 423.13 g and 19.89 cm, respectively. For *L. rohita*, these values were approximately 430.09 g and 20.03 cm, while for *C. carpio*, they were around 433.17 g and 20.02 cm, respectively. After that, all the samples were correctly labeled and transported to the laboratory in ice-filled containers and stored at − 20 °C for further analysis.

The next morning, the samples were thawed and washed with distilled water. Using a sharp knife, the scales were carefully removed from the fish and paper towels were used to dry the samples. A plastic tray was used to separate the tissue and organ samples for the dissection. The edible parts (flesh) from each sample were separated, and a sterilized stainless-steel knife was used to slice them into smaller pieces. Following the method of Habib et al. [15], the gills and liver were dissected. They were subsequently dried in a plastic petri dish at 80 °C to achieve constant weight, then cooled in a desiccator before being ground in a porcelain mortar and pestle [24].

Fish feed samples from Supreme Company Lahore, Pakistan, were used to feed the fish species. About 0.5 kg of feed samples were collected from fish farmers in clean plastic bags. The samples were ground using a grinding machine and stored in airtight containers to avoid contamination. They were subsequently used for proximate [27] and heavy

Table 1 Proximate composition of fish feed used for the fish species reared in a polyculture system

Parameters	Supreme feed (%)
Crude protein	28.06
Crude fat	13.14
Crude fiber	6.42
Moisture	7.93
Ash	7.06
Nitrogen free extract	45.32

NFE = 100 − (%crude protein + %crude fat + %ash + %fber) metals analyses [15]. Table 1 presents the proximate composition of fish feed.

Fish pond water samples were collected in clean (already autoclaved) glass-made bottles (1000 ml) from each pond in triplicates at 15–30 cm underwater. Water samples were filtered with a 0.45-µm membrane and then treated with 2% nitric acid before analysis [20].

For sediment samples, a stainless-steel grab sampler was used to collect sediment samples in clean polyethylene bags, which were then freeze-dried for 60 h. After grounding in a mortar, the sediments were sieved through a 0.125-mm mesh screen.

Water Quality Parameters

During sampling, water temperature (°C), dissolved oxygen (mg/l), pH (log H +), and total dissolved solids (ppm) were measured on the spot. Water temperature was measured using digital thermometer "model = HI-991002," dissolved oxygen (DO) through DO-meter "HI98198-Hanna," pH by using a pH meter "Beckman model-72," and total dissolved solids (TDS) was examined through a TDS meter "HM-COM-80". Other parameters, including ammonia (NH_3), nitrite (NO^-_2), and nitrate (NO^-_3), were observed through the Freshwater Master Test Kit "API, USA" from the sampled water. The details of water quality parameters are presented in Table 2.

Heavy Metals Analysis

For heavy metals (Pb, Cd, Cr, and Cu) analysis, 0.5 g of each sample, such as fish feed, sediment, fish tissues, and 50 ml water, was digested with three concentrated acids (nitric acid: HNO_3, sulfuric acid: H_2SO_4, and perchloric acid: $HClO_4$ (37%); Sigma–Aldrich) at a temperature of 80 °C [20]. Once the sample had been digested, it was cooled to room temperature and filtered using Whatman's filter paper of 0.45-µm pore size. Then, the sample was diluted with deionized water to a volume of 50 ml before further analysis. Estimated concentrations of heavy metals were determined. Flame conditions for atomic absorption have been optimized in order to achieve a higher absorption rate and a linear response. Blank, working standard, and actual biological samples were directly aspirated into the flame and with the help of atomic absorption spectrometer "model AA240FS, Varian Atomic Absorption Spectrophotometer," absorption was recorded. HNO_3 was added to 100 ml of filtered water and digested at 100 °C for 10 min. After cooling the samples to room temperature, they were diluted and filtered using the Whatman-42 filter paper. HNO_3 (0.01 N) was used to prepare 50 ml of the filtrate; samples were then analyzed. To ensure that the process was safe and effective, good quality chemicals and reagents were used during the process, as well as thorough cleaning and deionization of the glassware.

Bioaccumulation Factor

Based on the equation below, we calculated the bioaccumulation factor (BAF):

$$\mathrm{BAF} = \frac{\text{Concentration of heavy metal in the fish}}{\text{Concentration of heavy metal in the water}}$$

Estimated Daily Intake

In order to estimate the possibility of metal poisoning in humans, the estimated daily intake (EDI) of metals from fish muscles is used. Based on the following formula, an estimated daily intake (EDI) can be determined [28].

$$\mathrm{EDI}\left(\frac{\mathrm{mg}}{\mathrm{kg}}\text{body weight of}\frac{\text{consumer}}{\text{day}}\right) = \frac{(\text{Concentration of metal } x \text{ Weight of fish}}{\text{Body weight of consumer}}$$

where metal concentrations in fish muscles were converted into dry weight using an average concentration factor of 4.8 [29]. Pakistani men consume, on average, 70 g of fish daily, while their average weight was taken as 60 kg [30, 31].

Table 2 Water quality parameters of fish pond

Parameters	Mean ± SD
Temperature (°C)	28.3 ± 1.13
DO (mg/L)	5.8 ± 0.23
pH (-log H^+)	7.7 ± 0.14
TDS (ppm)	156.6 ± 8.52
Ammonia (ppm)	0.01 ± 0.005
Nitrite (ppm)	0.006 ± 0.002
Nitrate (ppm)	0.04 ± 0.006

Human Health Risk Assessment

To assess the risk associated with the consumption of metal-loaded fish, the estimated daily intake (EDI) was calculated. It is possible to measure the potential health risks associated with consuming heavy metal-contaminated food through the use of a target hazard quotient (THQ) and hazard index (HI) [32]. Calculations were made using the following formula.

$$THQ = EDI/RfD$$

Statistical Analysis

A one-way ANOVA was performed in GraphPad Prism (version 10.1.1) to separate the means (significance level: $P < 0.05$) of heavy metals. In addition, Duncan's multiple range test was used to determine whether there were significant differences between the detected heavy metals in fish species. Furthermore, to assess the association between the analyzed metals, principal component analysis (PCA) was applied.

Results

Heavy Metal Concentration inFish Feed, Water, and Sediment

The concentration of heavy metals in fish feed, pond water, and sediment is shown in Figs. 2, 3, and 4, respectively. According to the results, the Pb, Cu, and Cd concentrations were higher in sediment than in feed and pond water. The Cd level was almost similar in feed and pond water. The Cr level in feed was below the detection limit, while a slightly higher concentration was observed in the sediment than in pond water. In most cases, the mean concentration of heavy metals was greater in sediment, followed by water. However, the least amount was recorded in fish feed. The concentration of heavy metals in fish feed was lower than the FAO limit. However, the Pb and Cd levels were above the standard limit in water. In the case of sediment, the Cd, Cr, and Cu concentrations were recorded above the permissible limit (Table 3).

Comparative Analysis of Heavy Metals Concentration in Body Tissues of Fishes

The comparison of the heavy metals in the gills, liver, and flesh of different fish species based on their feeding zones is illustrated in Figs. 5, 6, and 7. After data analysis, it was found that the level of Pb and Cu concentration in gills, liver, and flesh were significantly higher ($P < 0.05$) in *C. carpio* (bottom feeder). The amount of Cd in body tissues of *C. catla* (surface feeder) and *L. rohita* (column feeder)

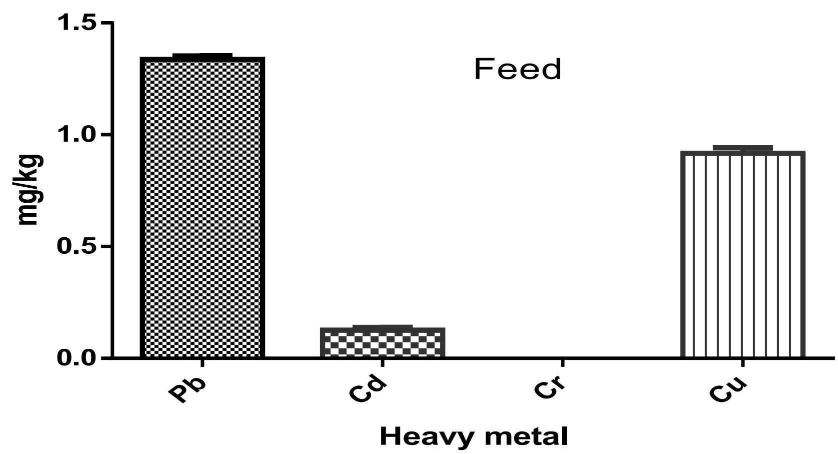

Fig. 2 Heavy metals concentrations in the fish feed used for the feeding of *C. carpio*, *L. rohita*, and *C. catla* in a polyculture system

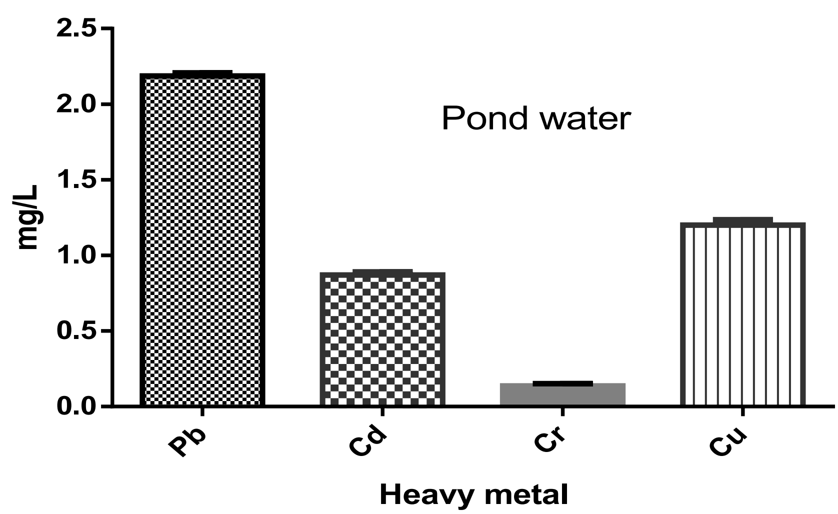

Fig. 3 Heavy metals concentrations in pond water of the polyculture system

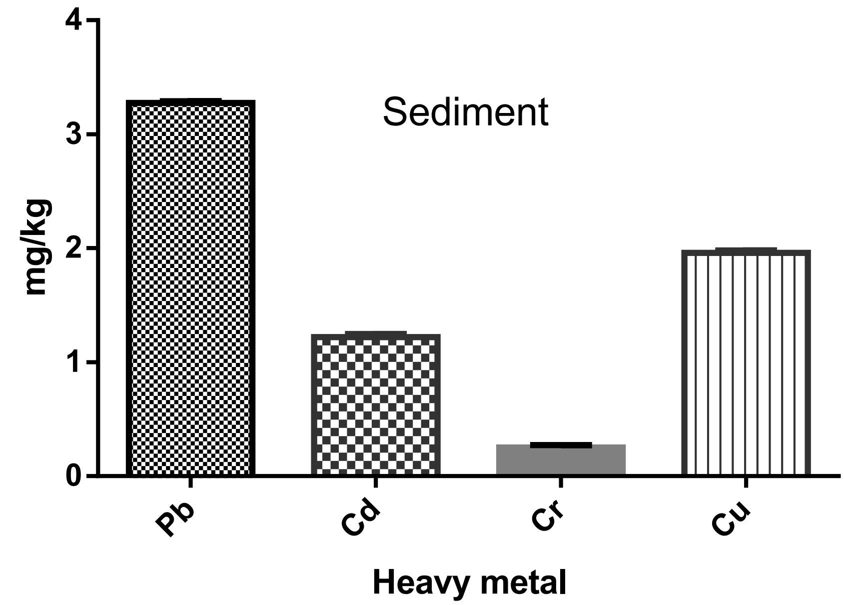

Fig. 4 Heavy metals concentrations in the sediment of the polyculture ponds

Table 3 Standard limit for heavy metals in fish feed, water, sediment, and fish

Heavy metals	Feed (mg/kg) [FAO, 33]	Water (mg/L) [WHO 34, 35]	Sediment (mg/kg) [USEPA [36]	Fish (mg/kg) [WHO, 35]
Pb	2.00	0.010	5	2
Cd	1.00	0.003	0.6	1
Cr	0.05	0.1	0.1	0.05–0.15
Cu	10.00	1.3	0.2	30

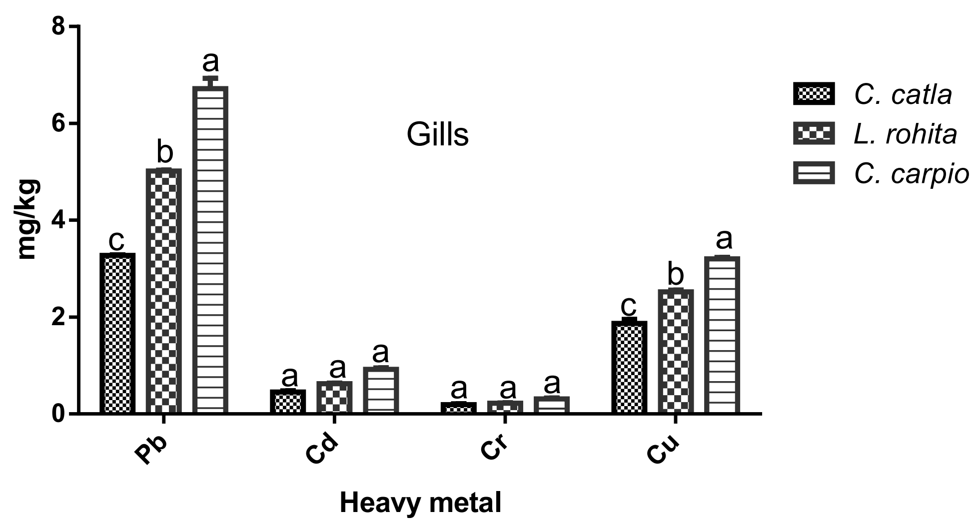

Fig. 5 Heavy metals concentrations in the gills of *C. carpio*, *L. rohita*, and *C. catla* reared in a polyculture system

revealed no significant difference ($P > 0.05$), while notably higher ($P < 0.05$) in the case of *C. carpio*. The concentration of Cr in fish species of different feeding zones showed no significant difference ($P > 0.05$). The mean concentration of heavy metals recorded was significantly higher ($P < 0.05$) in the liver, followed by gills in fish species. However, the least amount was found in the flesh of fishes. Overall, the bottom feeder fish showed significantly higher ($P < 0.05$) concentrations of heavy metals in their body tissues than the other fish species. The Pb and Cr levels in gills, liver, and flesh were higher in *L. rohita* and *C. carpio* while lower in *C. catla* flesh than the standard limit of

WHO. The amount of Cd was higher in the liver of all fish species compared to the WHO standard. However, the Cu level was lower than the standard limit (Table 3).

Bioaccumulation of Heavy Metals

Table 4 presents the bioaccumulation factor of heavy metals for different fish species. The results showed that the higher bioaccumulation of heavy metals was recorded in the liver, followed by gills in all experimental fish species of different feeding zones. However, the lower bioaccumulation factor was found in fish flesh. Comparatively, the bioaccumulation of metals was higher in bottom feeder fish (*C. carpio*) followed by column feeder (*L. rohita*), while lower bioaccumulation was found in surface feeder fish (*C. catla*).

PCA Analysis for Heavy Metals

According to the PCA analysis for surface feeder (*C. catla*), the heavy metals observed in fish flesh and feed are strongly

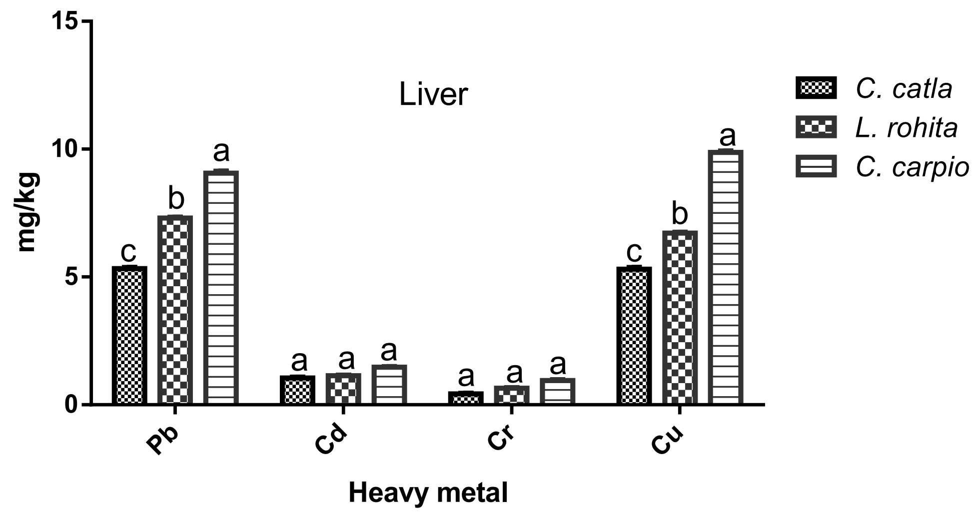

Fig. 6 Heavy metals concentrations in the liver of *C. carpio*, *L. rohita*, and *C. catla* reared in a polyculture system

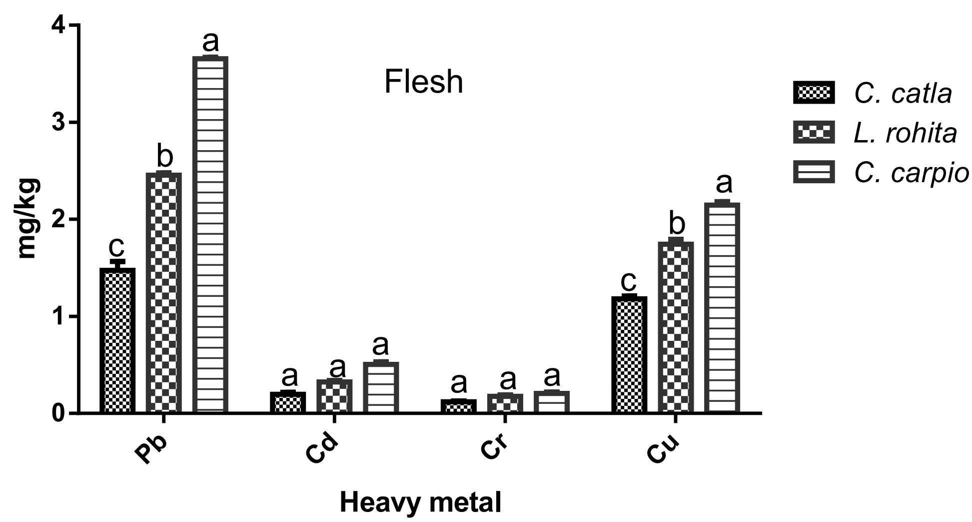

Fig. 7 Heavy metals concentrations in the flesh of *C. carpio*, *L. rohita*, and *C. catla* reared in a polyculture system

Table 4 Bioaccumulation factor of heavy metals (mg/kg) in different tissues of *C. carpio*, *L. rohita*, and *C. catla* in a polyculture system

Heavy metal	*Catla catla*			*Labeo rohita*			*Cyprinus carpio*		
	Gills	Liver	Flesh	Gills	Liver	Flesh	Gills	Liver	Flesh
Pb	1.49	2.43	0.67	2.29	3.34	1.12	3.07	4.14	1.67
Cd	0.52	1.20	0.22	0.71	1.31	0.37	1.06	1.68	0.58
Cr	1.41	3.14	0.87	1.60	4.75	1.29	2.26	6.97	1.51
Cu	1.55	4.42	0.98	2.1	5.6	1.45	2.66	8.23	1.78

correlated, which means that they are positively correlated. On the other hand, the heavy metals in the liver are closer to the sediment and gills. Furthermore, the Cd level is not significantly correlated to other heavy metals, as the vector direction indicates (Fig. 8).

Similar results for the column feeder (*L. rohita*) were found in which the feed, flesh, gills, and pond water correlate (Fig. 9).

In the case of the bottom feeder (*C. carpio*), the detected heavy metals in fish feed, flesh, gills, and pond water are correlated, as shown in Fig. 10. The heavy metals Cr, Pb, and Cu are also strongly associated with each other, while Cd is slightly positively correlated.

Health Risk Analysis

The association of heavy metals with human health risks from the flesh of different fish species is depicted in Table 5. The results revealed that for all the heavy metals investigated (Pb, Cd, Cr, and Cu), the values of EDI and THQ were higher in the flesh of *C. carpio* than in *L. rohita* and *C. catla* reared in polyculture form. Fish consumption by children and adults was associated with a higher risk of exposure to harmful elements when the EDI value was high. Furthermore, an elevated HI value greater than 1 indicates a greater threat to human health. In our study, the HI value for the *C. carpio* was higher than one compared to other fish species.

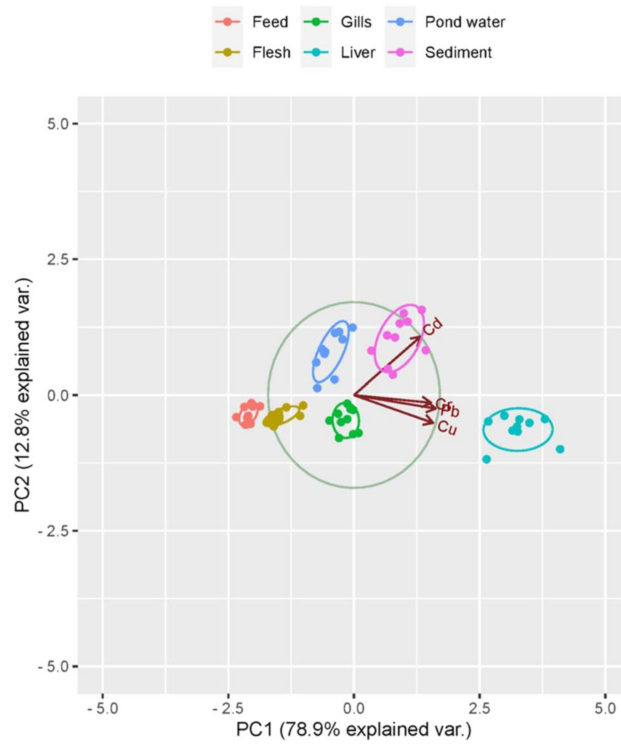

Fig. 9 PCA analysis for *L. rohita*

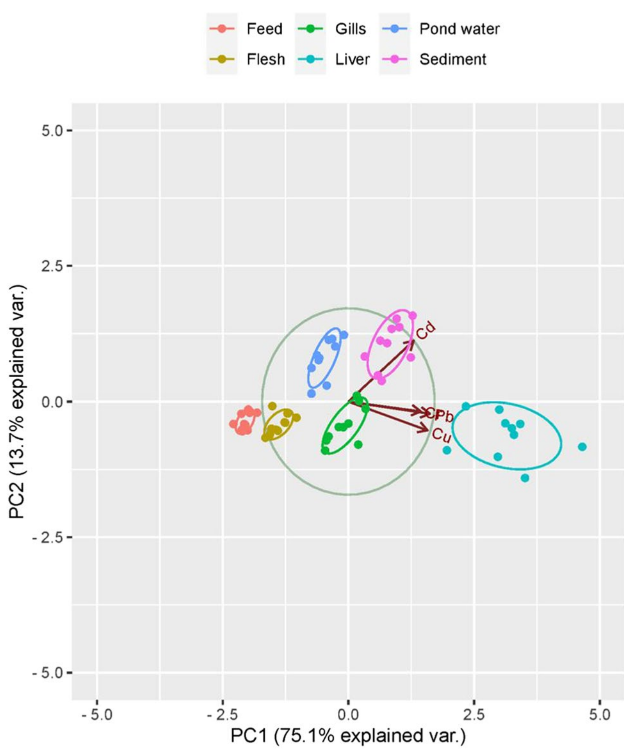

Fig. 8 PCA analysis for *C. catla*

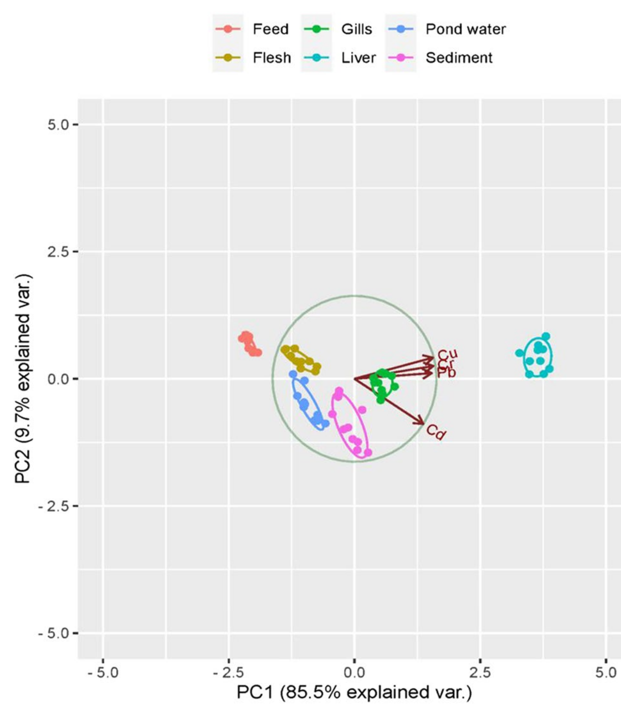

Fig. 10 PCA analysis for *C. carpio*

Table 5 Health risk estimation of heavy metals from the muscles of *C. carpio*, *L. rohita*, and *C. catla* in a polyculture system

Fish species	Heavy metals	EDI mg kg^{-1} day^{-1}	THQ	HI
Catla catla	Pb	0.43	0.97	0.75
	Cd	0.05	0.52	
	Cr	0.26	0.26	
	Cu	1.22	0.24	
Labeo rohita	Pb	0.73	1.52	0.93
	Cd	0.07	0.82	
	Cr	0.24	0.24	
	Cu	1.52	0.32	
Cyprinus carpio	Pb	0.81	1.23	1.04
	Cd	0.07	0.76	
	Cr	0.33	0.26	
	Cu	1.57	0.35	

EDI, estimated daily intake; *THQ*, target hazard quotient; *HI*, hazard index

According to the current research, bottom feeder fish such as *C. carpio* pose a higher risk to consumers than column and surface feeders. The value of HI was the least recorded in surface feeder fish.

Discussion

Various factors influence fish's heavy metal bioaccumulation, such as feeding habits, water temperature, pH, salinity, metal interaction, sediment, feed, sex, and age [15, 37]. However, little literature exists that compares heavy metal bioaccumulation in fish species with different feeding zones in a similar habitat. Due to this, our study investigated and compared the heavy metal levels in three fish species (*C. catla*, *L. rohita*, and *C. carpio*) reared in a polyculture system fed in different feeding zones. Furthermore, this study analyzed the heavy metals in fish feed, water, sediment, and related human health risks.

Results revealed that bottom feeder fish, *C. carpio*, tend to have higher concentrations of heavy metals in their gills, liver, and flesh compared to fish species in different feeding zones. This suggests that bottom feeders are likely more exposed to polluted sediments and absorb more heavy metals from earthen pond sediments. The sediments play a crucial role in the uptake of heavy metals by fishes, and the foraging behavior and swimming of bottom-dwelling fishes may contribute to a transfer of heavy metals from the sediments to the fish [38].

Other studies also revealed similar findings, such as Kumar et al. [39] found that the bottom feeder fish (*Channa striata*) has the highest concentration of heavy metals than the column (*L. rohita*) and surface feeder fish (*C. catla*). Jiang et al. [40] reported that the bioaccumulation of heavy metals (Pb, Cd, Zn, and Cu) in demersal fishes was higher compared to pelagic and benthopelagic fishes in Dongting Lake, China, which is similar to this study. Asante et al. [34] observed notably significantly higher levels of heavy metals (Mn, Cs, Co, Cr, Se, and As) in demersal fishes. Similar findings were also reported by Jitar et al. [35] about the higher amount of heavy metals bioaccumulation in demersal fishes from the Black Sea. Heavy metals are absorbed from sediment by demersal fish as the primary environment is closely related to them. Furthermore, heavy metals are transferred to fish via both dietary and dissolved routes during predation and swimming [41, 42]. Inconsistencies in heavy metal concentrations in surface and bottom feeder fish may be due to geographic factors, living environment factors, and the specific toxicity of the metal [43].

High concentrations of heavy metals (Pb, Cd, and Cr) observed in the liver, exceeding WHO [44, 45] standards, may be linked to natural protein binding, including metallothioneins [46]. Moreover, in the liver, metals are stored, detoxified, and re-distributed [47]. Metabolic function may be affected when heavy metals are concentrated in the liver. In addition, fish liver is considered an important indicator of water pollution [48]. Other studies also show a higher concentration of heavy metals in fish liver [14]. In this study, the higher concentrations of heavy metals recorded in the gills than in the liver may be because metals are mostly transferred in water through the gill pores, which have a vast surface area that permits the metals to spread more quickly [15, 49]. Several investigations have shown that the high quantity of heavy metals in fish gills is mainly concentrated in the water [50, 51]. High levels of lead (Pb) in fish tissues can potentially harm human health if ingested through food. Pb-contaminated fish can negatively affect a person's liver, brain, neurological, kidneys, and reproductive systems [36]. When fish are exposed to toxicants, bioaccumulation is the primary mechanism for Pb-induced toxicity [52]. However, the means by which the chemicals accumulate differ, depending on the environmental conditions, for example, freshwater or seawater, as well as the route of exposure (dietary or waterborne) of the chemicals [53]. The concentration of Pb in the water and fish was higher than the WHO limit but lower in the flesh of the surface feeder.

A study by Okocha and Adedeji [54] reported a higher concentration of Cd in fish liver and gills, which is according to the current research. Fish exposure to Cd for a longer duration slows bone development, induces tumors, and elevates blood pressure [11, 55]. According to Fazio et al. [11], the growth and hematological parameters of fish (*Mystus seenghala*) were lowered by the sub-lethal effect of Cd. The Cd recorded in this study was higher in fish species' liver, water, and sediment than the WHO [44] and USEPA [56] limits. The Cr level in sediment, gills, liver, and flesh of column and bottom feeder fish was higher than the USEPA and WHO standards. The presence of high levels of Cr in fish can alter blood parameters as well as damage to the liver and kidneys [57]. Overconsumption of Cr has been associated with serious human health risks [33].

The current study findings revealed a lower level of heavy metals in fish feed than the standard limit [44]. This is consistent with the findings of other studies, such as Habib et al. [13, 15], who reported the lower level of heavy metals (Cd, Cr, Pb, Cu, and Zn) in commercial feed used for the culturing of *Oreochromis niloticus* and *Cyprinus carpio* in biofloc technology. Other studies from Bangladesh also revealed lower levels of heavy metals (Cr, Cu, Zn, Pb, and Cu) in different commercial feeds [25].

In the current sediment study, heavy metals concentration was higher than in water and feed. Moreover, compared to the permissible limits [56, 58], the Cd, Cr, and Cu amounts were higher. This is according to the results reported by Weber et al. [59], who recorded higher concentrations of heavy metals (Cd, Cr, Zn, Fe, Cu, Mn, As, and AI) in sediment followed by water in the Brazilian River. The sediment was the primary source of metal pollution in this polyculture pond. Thus, the bottom feeder fish accumulate more metals in their liver than the column and surface feeder. The suspended sediments act as adsorbents, thus reducing the concentration of pollutants in the water column [37].

The reference dose is the maximum acceptable daily consumption of a particular metal with no harmful effects on health [60]. The human health risk assessment showed that the reference dose for bottom feeder fish was higher, indicating a potentially greater risk to human health associated with their consumption. Thus, there is a chance of health risks in children and adults consuming bottom-dwelling fish. Similar results were reported by Kumar et al. [39], who revealed a higher HI value (> 1) for the bottom feeder than the surface feeder. The current study is contrary to the results of Wu et al. [61], who revealed a lower reference dose for heavy metals in pelagic, benthopelagic, and demersal fish. When exposed to high metal concentrations, fish can absorb and accumulate the metals through their gills and skin or ingest contaminated water and food [15, 62]. As a result, the metal concentration in fish tissues might be many times greater than that of the surrounding medium [24].

Conclusion

This study suggests that the concentration of heavy metals in fish feed, water, and sediment samples from polyculture ponds varies. They were found to be higher for water (Pb and Cd) and sediment (Cd, Cr, and Cu) and lower for fish feed than the permissible limit proposed by several agencies. A higher concentration was found in sediment than in water and feed. Thus, the sediments are the major sink of heavy metals and play a significant role in the uptake of heavy metals by bottom-feeding fish. This bottom feeder fish might be useful as bioindicators to trace metal contamination. Long-term consumption of fish, particularly the bottom feeders from this region, is not advisable, as evidenced by risk assessments in the form of THQ and HI. This may cause adverse health consequences to humans.

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Data Availability The datasets generated during and/or analyzed during the current study are available from the corresponding author upon reasonable request.

Declarations

Ethics Approval The authors followed all the valid national rules for the use and care of animals, and the study was conducted after the ethical committee approval of the Department of Zoology, University of Lahore, Pakistan.

Consent for Publication Not applicable.

Competing Interests The authors declare no competing interests.

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