

Heavy Metals (Lead, Cadmium and Nickel) Concentration in Different Organs of Three Commonly Consumed Fishes in Bangladesh

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Abstract

Pollution and food safety are concerning issues in recent years. For that, the present investigation was aimed to determine the levels of heavy metals (Lead, Cadmium and Nickel) in different organs of three fish (*Labeo rohita*, *Gibelion catla* and *Pangasius hypophthalmus*) and to compare the present value with Food and Agriculture Organization (FAO)/World Health Organization (WHO) acceptable limit. These commonly consumed fish were collected from Kawran Bazar fish market, Bangladesh and the concentrations of heavy metals were carried out using Atomic Absorption Spectrophotometer. The highest (48.33 ± 2.52 ppm) concentration of Lead (Pb) was found in the gill of Pangus fish and the lowest (23.33 ± 2.52 ppm) was found in the gonad of Rui fish. In case of Cadmium (Cd), the highest (0.96 ± 0.08 ppm) concentration was found in the gill of Rui fish whereas the lowest (0.02 ± 0.01 ppm) was found in the kidney of Pangus fish. The highest (6.63 ± 1.00 ppm) concentration of Nickel (Ni) was found in the gill of Katla fish and the lowest (0.10 ± 0.10 ppm) was found in the liver of the same species. The metal concentrations in the muscle of Rui, Katla and Pangus fish were in the decreasing order of $Pb > Ni > Cd$, $Pb > Cd > Ni$ and $Pb > Ni > Cd$ respectively. In the study, it was found that Pb has exceeded the limits in every case whereas Cd was within the limits except for a few and Ni was completely within the maximum allowable limits provided by FAO/WHO. However, all these concentrated metals in different parts of fish body could be concentrated into human body, if they are consumed and if so happen; there will be a massive health risk for us.

Keywords Heavy metal; Fishes; Organs; Concentration; Allowable limit

Introduction

Pollutions are increasing day by day and we have to be concerned about our future environment. The day we begin to control our environment we begin to destroy it consciously, subconsciously or unconsciously. Environmental pollution is the universal problem and most important pollutants are the heavy metals (HMs) in aquatic network because of their toxicity, accumulation and bio-magnification by aquatic organisms. Among the pollutants, HMs consist a major portion. The HM pollutions have dreadful effects on the environmental equilibrium and a variety of aquatic entities [1,2]. The pollutants like HMs gradually accumulate in food chain and cause the antagonistic effects, even death. Generally, accumulation relies on metal concentration, time of exposure, way of metal uptake, environmental conditions (water temperature, pH, hardness, salinity) and intrinsic factors (fish age, feeding habits) [3]. Commonly encountered HMs is chromium, cobalt, nickel, copper, zinc, arsenic, selenium, silver, cadmium, antimony, mercury, thallium and lead [4]. All metals can be toxic if levels are too high. HM is easily stored in fatty tissue and will bio-accumulate if the fish is exposed to further contamination.

Generally in fish, the toxic effect of heavy metal influences physiological functions, individual growth and mortality [5]. HMs enters plant, animal and human tissues via air inhalation, diet and manual handling. Water sources (groundwater, lakes, streams and rivers) can be polluted by HMs leaching from industrial and consumer waste; acid rain can exacerbate this process by releasing HMs trapped

in soils [6]. Absorption through skin contact, for example from contact with soil, is another potential source of heavy metal contamination [7]. Bioaccumulation of any metal above its threshold level invariably results in stress often leading to irreversible physiological conditions [8]. Fish occupy a major portion in our diet as well as provides 60% of animal protein. According to DoF [9], Rui, Katla as well as Pangus are mostly produced and consumed fish in Bangladesh. Contamination of freshwater fish with HMs is recognized as an austere environmental problem. The World Health Organization (WHO) as well as the Food and Agriculture Organization (FAO) of the United Nations state that monitoring eight elements in fish; Mercury (Hg), Cadmium (Cd), Lead (Pb), Arsenic (As), Copper (Cu), Zinc (Zn), Iron (Fe), Tin (Sn) is obligatory and monitoring of others is suggested [10]. The aim of the study were to investigate the presence of heavy metal contaminants like Lead (Pb), Cadmium (Cd) and Nickel (Ni) in three commonly consumed fish, determine the concentration of bio-accumulated HMs in liver, gill, kidney, gonad and muscle of fish and find out whether the studied fish have elevated concentrations of HMs in their tissues that could render them dangerous for human consumption, by comparing with the maximum permissible limits proposed by FAO and WHO.

Material and Methods

Sample collection and preparation

Mostly consumed three fish species (5-6 individuals of each species) namely, Rui (*Labeo rohita*; average weight 1.32 ± 0.02 kg and average length 41 ± 0.80 cm), Katla (*Gibelion catla*; average weight 1.5 ± 0.04 kg and average length 43 ± 0.75 cm) and Pangus (*Pangasius*

hypophthalmus, average weight 1.28 ± 0.02 kg and average length 38 ± 1.0 cm) were collected from one of the largest domestic fish market of Bangladesh, Kawran Bazar of Dhaka. Fish samples of uniform size were collected in order to avoid the possible error due to size differences. The specimens were placed immediately in poly-ethylene bags, put into isolated container of polystyrene icebox and then, brought to the Genetics and Fish Breeding laboratory on the same day for identification and dissection. After sample collection it was prepared for chemical analysis. Sample preparation included oven drying, grinding, screening and overnight digestion (12 hours), respectively. Each sample of 0.5 gram was taken and measured by electric balance, then digested by applying di-acid $\text{HNO}_3\text{-HClO}_4$ acid (2.5:1) [11,12] with the block digester (Model-VELP). Then, the digested samples were filtrate with Whatman No.1 and made a volume up to 50 ml with deionized water. This is how the digestion process was used for the determination of lead, cadmium and nickel.

Blank preparation

At each step of the digestion processes of the samples acid blanks (laboratory blank) were done using an identical procedure to ensure that the samples and chemicals used were not contaminated. They contained the same digestion reagents as the real samples with the same acid ratios but without fish sample. After digestion, acid blanks were treated as samples and diluted with the same factor. They were analysed by atomic absorption spectrophotometry before real samples and their values were subtracted to check the equipment to read only the exact values of HMs in real samples. Each set of digested samples had its own acid blank and was corrected by using its blank sample.

Chemical analysis

Sample analysis was done to determine the concentration of heavy metal (Ni, Pb, and Cd) in different fish organs. Analysis was done in Soil Science Laboratory, BSMRAU with the help of atomic absorption

spectrophotometer (AAS). The wavelength used for Pb, Cd and Ni analysis was 283.2 nm, 228.9 nm and 232 nm, respectively. Necessary information for each samples such as date of sample collection, time of sample collection, location etc., were recorded in the note book.

Calculation

The experiment was conducted in a dry weight basis. After getting the raw data from AAS it was multiplied with the dilution factor. As 0.5 g fish samples were taken and made volume up to 50 ml, so the dilution factor was 100.

Data analysis and Comparison

Descriptive statistics such as average and standard deviation values were calculated. Finally mean concentration of HMs found in different samples were compared to the recommended accepted value provided by FAO/WHO to check if the present value of HMs were within the limits or not.

Results and Discussion

Mean concentration of heavy metals in the organs of *Labeo rohita*, *Gibelion catla* and *Pangasius hypophthalmus* fish

Average HMs content in different organs of Rui fish were found to be in different levels (Table 1). Levels of Pb in Rui fish ranged from 2.33 ppm to 25.67 ppm. Concentrations of Cd ranged from 0.08 ppm to 0.96 ppm and concentrations of Ni ranged from 1.27 ppm to 4.80 ppm in the Rui fish. The metal bioaccumulation of Pb in the Rui fish has the decreasing order of liver>gill>kidney>muscle>gonad. The other view, Cd in the Rui fish has the decreasing order of gill>liver>muscle>kidney>gonad. In contrast of Ni in the Rui fish has the decreasing order of kidney>liver>muscle>gill>gonad.

Organs	Rui (ppm)			Katla (ppm)			Pangus (ppm)		
	Pb	Cd	Ni	Pb	Cd	Ni	Pb	Cd	Ni
Liver	25.67 ± 1.53	0.31 ± 0.02	3.77 ± 0.15	33.00 ± 3.61	0.14 ± 0.02	0.10 ± 0.10	42.33 ± 2.08	0.84 ± 0.04	1.70 ± 0.10
Gill	23.33 ± 2.52	0.96 ± 0.08	BDL*	32.33 ± 5.86	0.32 ± 0.00	6.63 ± 1.00	48.33 ± 2.52	0.37 ± 0.00	BDL
Kidney	17.67 ± 2.52	0.08 ± 0.00	4.80 ± 0.36	5.33 ± 0.58	0.42 ± 0.01	BDL	34.33 ± 0.58	0.02 ± 0.01	BDL
Muscle	7.67 ± 2.08	0.16 ± 0.01	1.27 ± 0.12	3.33 ± 1.53	0.49 ± 0.01	0.27 ± 0.15	10.33 ± 1.53	0.34 ± 0.01	1.49 ± 0.46
Gonad	2.33 ± 1.15	BDL	BDL	5.33 ± 1.15	0.71 ± 0.12	BDL	18.67 ± 1.53	BDL	BDL
FAO/WHO**	0.30	0.50	80	0.30	0.50	80	0.30	0.50	80

*Below Detection Limit.

** FAO/WHO, 1984; FAO/WHO, 2011.

Table 1: Heavy metals (Pb-Lead; Cd-Cadmium; Ni-Nickel) in the liver, gill, kidney, gonad and muscle of Rui (*Labeo rohita*), Katla (*Gibelion catla*) and Pangus (*Pangasius hypophthalmus*) fishes from Kawran Bazar fish market.

Pb was exceeded the recommended value provided by FAO/WHO whereas Cd and Ni were within the guidelines except for Cd in the gill of Rui fish. Ahmed et al. [13] also found higher Pb and Cd values that

exceeded the acceptable limit, which provided by Food and Agriculture Organization/World Health Organization.

Levels of Pb in Katla fish ranged from 3.33 ppm to 33.00 ppm. Concentrations of Cd ranged from 0.14 ppm to 0.71 ppm and

concentrations of Ni ranged from 0.10 ppm to 6.63 ppm in Katla fish (Table 1). The bioaccumulation of Pb in the Katla fish has the decreasing order of liver>gill> kidney>gonad>muscle. For the accumulation of Cd in the Katla fish has the decreasing order of gonad>muscle>kidney>gill>liver. Another, bioaccumulation of Ni in the Katla fish has the decreasing order of kidney>liver>muscle>gill>gonad. For Katla, Pb was exceeded the recommended value provided by FAO/WHO whereas Cd and Ni were within the guidelines except for Cd in the gonad. According to Ahmed et al. [14], fishes were highly exposed to those metals in gill, kidney and liver tissues.

Pb Levels in Pangus fish ranged from 10.33 ppm to 48.33 ppm. Concentrations of Cd ranged from 0.02 ppm to 0.84 ppm and concentrations of Ni ranged from 1.49 ppm to 1.70 ppm in Pangus fish (Table 1). The accumulation of Pb in the Pangus fish has the decreasing order of gill>liver>kidney>gonad>muscle. The metal bioaccumulation of Cd in the Pangus fish had the decreasing order of liver>gill>muscle>kidney>gonad. The other view, Ni in the Pangus fish has the decreasing order of liver>muscle>gill>kidney>gonad. The order of bioaccumulations of these metals might be as a result of the fact that different metals tend to accumulate differently in the tissues of different species of fish. Pb was exceeded the recommended value provided by FAO/WHO whereas Cd and Ni were within the guidelines except for Cd in the liver. Begum et al. (2013) [15] reported that the liver appeared to be the main heavy metal storage tissue, while the gonad had the lowest levels of analysed metals. This result is almost similar to our achievement. But only exception was occurred for Pb probably due to highly uptake of contaminated water through gills.

Comparison in the concentrations of HMs in different organs of different fishes

The HMs content in liver tissues among the three species of fish where Pb levels in the liver of three species ranged from 25.67 ppm to 42.33 ppm, Cd levels ranged from 0.14 to 0.84 ppm and Ni levels ranged from 0.10 to 3.77 ppm (Table 2). The metal concentrations in

the liver of Rui, Katla and Pangus fish were in the decreasing order of Pb>Ni>Cd, Pb>Cd>Ni and Pb>Ni>Cd respectively. The liver of Pangus accumulated significantly higher levels of Pb and Cd than other species; Ni found to be higher in the liver of Rui fish. In the liver of all the fish, Pb found to be exceeded the FAO/WHO limits whereas Cd (except for Pangus) and Ni found to be within the recommended levels. The liver plays an important role in accumulation and detoxification of HMs [16]. Exposure of fish to elevated levels of HMs induces the synthesis of metallothioneine proteins (MT), which are metal binding proteins [17,18]. Fishes are known to possess the metallothioneine proteins [19]. Metallothioneine proteins have high affinities for HMs and in doing so, concentrate and regulate these metals in the liver. Metallothioneine proteins bind and detoxify the metal ion [20]. On an average the liver tissue came second in terms of metals accumulation after gills.

The metal concentrations in the gill of Rui, Katla and Pangus fish were in the decreasing order of Pb>Cd>Ni, Pb>Ni>Cd and Pb>Cd>Ni respectively (Table 2). Pb levels ranged from 23.33 ppm to 48.33 ppm; Cd levels ranged from 0.32 ppm to 0.96 ppm and Ni found up to 6.63 ppm. Ni found in a concentration of 6.63 ppm only in the gill of Katla fish. Pb found to be exceeded the FAO/WHO limits whereas Cd and Ni found to be within the recommended levels except for Cd in the gill of Rui fish. Gill surfaces are the first target of water borne metals [21]. The microenvironment of the gill surface consists of an epithelial membrane which primarily contains phospholipids covered by a mucous layer [22]. According to Reid and McDonald [23], the gill surface is negatively charged and thus provides a potential site for gill-metal interaction for positively charged metal. Laboratory experiments have indicated that in fishes which take up HMs from water, the gills generally show higher concentration than in the digestive tract. On the other hand, fish accumulating HMs from food show elevated metal levels in the digestive tract as compared to the gills [24,25]. The gills of all the fish tend to accumulate significant high levels of heavy metal comparatively than other tissues. On an average the gill tissue came first in terms of metals accumulation.

Fish	Liver (ppm)			Gill (ppm)			Kidney (ppm)			Gonad (ppm)			Muscle (ppm)		
	Pb	Cd	Ni	Pb	Cd	Ni	Pb	Cd	Ni	Pb	Cd	Ni	Pb	Cd	Ni
Rui	25.67 ± 1.53	0.31 ± 0.02	3.77 ± 0.15	23.33 ± 2.52	0.96 ± 0.08	BDL	17.67 ± 2.52	0.08 ± 0.00	4.80 ± 0.36	2.33 ± 1.15	BDL	BDL	7.67 ± 2.08	0.16 ± 0.01	1.27 ± 0.12
Katla	33.00 ± 3.61	0.14 ± 0.02	0.10 ± 0.10	32.33 ± 5.86	0.32 ± 0.00	6.63 ± 1.00	5.33 ± 0.58	0.42 ± 0.01	BDL	5.33 ± 1.15	0.71 ± 0.12	BDL	3.33 ± 1.53	0.49 ± 0.01	0.27 ± 0.15
Pangus	42.33 ± 2.08	0.84 ± 0.04	1.70 ± 0.10	48.33 ± 2.52	0.37 ± 0.00	BDL	34.33 ± 0.58	0.02 ± 0.01	BDL	18.67 ± 1.53	BDL	BDL	10.33 ± 1.53	0.34 ± 0.01	1.49 ± 0.46
FAO/WHO	0.3	0.5	80	0.3	0.5	80	0.3	0.5	80	0.3	0.5	80	0.3	0.5	80

*Below Detection Limit.

Table 2: Comparison in the concentrations of heavy metals (Pb-Lead; Cd-Cadmium; Ni-Nickel) in liver, gill, kidney, gonad and muscle of different fish species of Kawran Bazar fish market.

The comparison in the concentrations of HMs in the kidney samples among three species of fish where Pb levels ranged from 5.33 ppm to 34.33 ppm; Cd levels ranged from 0.02 ppm to 0.42 ppm and Ni found up to 4.80 ppm (Table 2). The metal concentrations in the kidney of Rui, Katla and Pangus fish were in the decreasing order of Pb>Ni>Cd, Pb>Cd>Ni and Pb>Cd>Ni respectively. Ni found in a

concentration of 4.80 ppm only in the Kidney of Rui. Pb found to be exceeded the FAO/WHO limits whereas Cd and Ni found to be within the recommended levels. Higher temperatures promote accumulation of cadmium especially in the most burdened organs; kidneys and liver [26]. In teleost's, the kidney, together with the gills and intestine, are responsible for excretion and the maintenance of the homeostasis of

the body fluids [27,28] and besides producing urine, act as an excretory route for the metabolites of a variety of xenobiotic to which the fish may be exposed [29]. The effects of pollutants on fish kidneys have been studied in some species and the severity of damage seen depends on the sensitivity of the species to the substances released into the environment [30,31].

In the gonad samples, the comparison in the concentrations of HMs among three species of fish where Pb levels ranged from 2.33 ppm to 18.67 ppm; Cd found up to 0.71 ppm and Ni were below the detection level (Table 2). The highest concentration of Pb (18.67 ppm) was found in the gonad of Pangus fish and the lowest (2.33 ppm) was found in the gonad of Rui fish. Cd found in a concentration of 0.71 ppm only in the gonad of Katla fish. Pb found to be exceeded the FAO/WHO limits whereas Cd and Ni found to be within the recommended levels. As gonad is an important organ for reproduction, basically without gonad and gonadal function it is quite impossible to think of a next generation [32]. Heavy metal concentration in fish gonad have comparatively found somehow lower than other organs of fish, which might have happened because of some probable mechanism to avoid the malfunctioning of gonad due to toxic HMs. But the presence of some HMs found in gonad would be a risk for the fish reproduction.

Among the three species of fish, the variation of HMs concentration in the muscle samples were as presented where Pb levels ranged from 3.33 to 10.33 ppm; Cd levels ranged from 0.16 ppm to 0.49 ppm and Ni levels ranged from 0.27 ppm to 1.49 ppm (Table 2). The metal concentrations in the muscle of Rui, Katla and Pangus fish were in the decreasing order of Pb>Ni>Cd, Pb>Cd>Ni and Pb>Ni>Cd respectively. Pb found to be exceeded the FAO/WHO limits whereas Cd and Ni found to be within the recommended levels. In average HMs concentrations have found to be lower in fish muscle than other organs and this might happened because of that might be a reflection of the low levels of metal binding proteins in the muscle [33].

Proximate heavy metals uptake in human body

As the most edible part of fish is its muscle, so, it was found that after consuming these fish with above concentration, an average person could accumulate Pb in an amount of 147.97 mg, 64.33 mg and 199.43 mg for Rui, Katla and Pangus fish, respectively in a year (Figure 1). Cd accumulation could be 3.03 mg, 9.50 mg, and 6.63 mg for Rui, Katla and Pangus fish, respectively in a year. Ni accumulation could be 24.45 mg, 5.15 mg, and 28.76 mg for Rui, Katla and Pangus fish, respectively in a year. Significantly higher levels of Pb than Cd and Ni could be accumulated in human body. In Bangladesh, 60% of animal protein comes from fish and on an average, about 19.30 kg fish is consumed by a person in a year [34]. Consumption of these metal concentrated fish could lead to severe health hazards including lung cancer, anaemia, nausea, vomiting, kidney malfunctions, and cardiovascular diseases as well as mental disorder [35].

Bioaccumulations of the HMs, it might be as a result of the fact that different metals tend to accumulate differently in the tissues of different species of fish. Besides, different concentration of HMs in different fish species might be a result of different ecological needs, metabolism and feeding patterns [33]. As fish lives primarily in water they concentrate a large volume of HMs in its body by bioaccumulation and bio-magnification [36]. Gonads, liver, kidney and gills are target organs for metals accumulation since they are metabolically active accumulating metals sometimes in high levels [37].

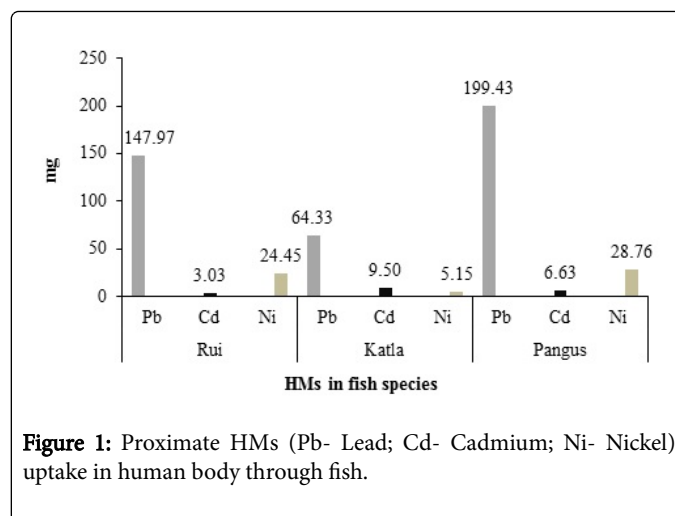


Figure 1: Proximate HMs (Pb- Lead; Cd- Cadmium; Ni- Nickel) uptake in human body through fish.

The low concentrations of metals in the muscle of fish species may reflect the low levels of binding proteins in the muscle [33]. In the present study, it was found that Pb has exceeded the FAO/WHO limits in every case whereas Cd was within the limits except a few and Ni was completely within the limits. However, all these concentrated metals in different parts of fish body could be concentrated into human body, if they are consumed. In Bangladesh, water sources are getting more polluted day by day and thus these HMs from polluted water bodies are getting more concentrated in those fish living in that areas. There is another way of concentrating HMs in fish body through the feed they are reared with and for farmed fish; it is our lacking that we don't care about those HMs from artificial feed getting introduced into fish body parts. If all these bad practices live long and if so happened in this way there will be a lot of reason to worry about as well as there will be a great threat for human health. The results found in the present study are just a warning for us and our future generation.

Conclusion

In aquatic water body high concentration of metals become gradually accumulated on the sediments and in due course get transferred to fish. The study clearly indicated significant accumulation of HMs in the organs of fishes. Consumption of contaminated fish should be monitored to avoid the adverse effects brought by HMs. Pb was found to be dominated in all of the studied metals and it has exceeded the recommended value provided by FAO and WHO in every case whereas Cd was within the limits except a few and Ni was completely within the limits for the three studied fish species in different organs. The carnivorous species (*P. hypophthalmus*) showed the highest metal concentrations in most cases, confirming the hypothesis that carnivorous species are prone to incorporate more HMs than fish in other trophic levels. Health risk analysis of HMs in the edible parts of the fish indicated toxic for human consumption based on FAO/WHO acceptable limit. This has confirmed the observations of other similar studies across the country, concerning the gradual build-up of toxic pollutants in our aquatic environment and it is important to note that the presence of trace heavy metal pollutants in diets could create serious health problems ranging from neuro-, nephro-, carcino- to immunological disorders, if ingested over a long period of time.

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