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Health Risk Assessment of Heavy Metal Intake of Common Fishes Available in the Brahmaputra River of Bangladesh

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Authors' contributions

This work was carried out in collaboration among all authors. Author HMZ designed the study, supervise the work, performed the statistical analysis and corrected the final draft of the manuscript. Author MSAE collected samples and performed analysis. Author QFQ helped to design the study and corrected the final draft of the manuscript. Author SM helped in manuscript preparation. All authors read and approved the final manuscript.

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ABSTRACT

Fifteen (15) fish species were collected from three (3) locations of the Brahmaputra River to determine heavy metals content in fish flesh, and to assess potential human health risk through consumption of those fishes. The contents of heavy metals in aqueous extract of different fish species were determined by an atomic absorption spectrophotometer (AAS) at the Department of Agricultural Chemistry, Bangladesh Agricultural University, during the period from March to June, 2018. The concentrations of Mn, Zn, Cu, Pb, Cd and Cr in different fish species ranged from 41.70- 376.62, 38.98-279.08, 3.19-47.91, 2.20-3.73, 0.19-1.20 and 0.59-2.40 mg kg⁻¹, respectively. The study results revealed that the edible portion of fish species of the river contained reasonably higher amount of Mn, Zn, Pb, Cr and Cu compared to some other previous studies carried out elsewhere with freshwater fishes. Among the fish species, *mola* (*Amblypharyngodon mola*) showed the highest metal pollution index (20.12) followed by *chela* (*Salmophasia bacaila*) (17.72), *chanda* (*Chanda nama*) (16.78) and *chingri* (*Macrobrachium sp*.) (15.83), while *baim* (*Macrognathus*

aculeatus) exhibited the lowest (6.79). The calculated daily metal intake for Pb, Mn and Cd for both male and female were higher than that of upper tolerable intake level, which indicates consumption of common fishes of this river, is not safe. Similarly, target hazard quotient (THQ) values for Mn surpassed 5.0 for both male and female, and the same value for Pb also surpassed 5.0 for female indicate that the exposed population are unsafe. The study results also revealed that all individual THQ values of other metals for both male and female surpassed 1, which means exposed population are in level of concern interval. In Bangladesh freshwater river fishes are more popular than farm fishes. However, the study results recommend to aware people about heavy metal contents before consuming fish species of a polluted river system.

Keywords: Heavy metal; health hazard; river fish species; Bangladesh.

1. INTRODUCTION

The contamination of freshwater ecosystems with a wide range of contaminants including heavy metals has become a great concern in all over the world during the last few decades. Rivers and canals are the most important ones among the different leading pathways for transporting contaminants or pollutants from one place to another. Heavy metals enter into these aquatic systems mainly through natural inputs such as weathering and erosion of rocks and anthropogenic sources including urban, industrial and agricultural activities, terrestrial runoff and sewage disposal [1-6]. Contamination of aquatic ecosystems with heavy metals has received much attention due to their toxicity, abundance and persistence in the environment and subsequent accumulation in aquatic habitats [7].

Most of the rivers of Bangladesh being polluted fast and the reasons are connected with various factors. There is a progressive increase in industrial as well as urban wastes and effluents
due to the rapid industrialization and the rapid industrialization and urbanization. Such waste products and effluents are discharged into the rivers without any kinds of treatment, which have a significant contribution to the heavy metal content of river water and sediments [8-13]. These heavy metals and other toxic substances, which often lead to decrease in number of fish species in rivers and canals of Bangladesh. On the other hand, the available fish species may contain higher amount of these toxic metals, which often leads to contamination of food chain.

Heavy metals accumulate by fish in polluted aquatic environment are varies due to ecological conditions, metabolisms and contamination level of water, sediments and foods. Hence, the Food and Agricultural Organization (FAO), World Health Organization (WHO), US Environmental Protection Agency (USEPA), and other regulatory bodies of various countries have established the maximum permissible limits/ concentrations of heavy metals in food stuffs [14]. However, for most of the people, the main route of exposure to heavy metals is through diet, accounting for >90% compared to other ways of exposure such as inhalation and dermal contact [15]. It is worth mentioning that although heavy metals can change their chemical form, they cannot be degraded or destroyed. Therefore, the risk assessment of these metals through dietary intake is an important issue [16-19].

The Brahmaputra River is a large trans-Himalayan river, which is originating in the southern Tibet (China). Geologically, the river is the youngest of the major rivers of the world and unique in many respects. The river runs for a length of 2880 km through the parts of China, Bhutan, India and Bangladesh [20]. The Brahmaputra is one among the major rivers in Bangladesh. Like others, the river system also receives untreated wastewater, sewage and effluents from municipalities and industries. However, it provides a variety of different fish species to the people of the country. Recently, a total of 67 finfish species including 63 indigenous and 4 exotic/alien species have been recorded from the Brahmaputra River belonging to 46 genera, 24 families and 8 orders [21]. The available fish species may accumulate heavy metals in their tissues through absorption and human can be exposed to heavy metals risk through consumption of those fishes because the river fishes are most popular among the all income group's people of Bangladesh. Considering the above fact, this study was carried out to measure heavy metal contents in edible parts of available fish species of the Brahmaputra River and to assess potential health risk for adult male and female through consumption of those fishes.

2. MATERIALS AND METHODS

2.1 Collection of Fish Samples

There were 3 (three) locations viz. *Bhabakhali bazar*, Bangladesh Agricultural University (BAU) campus and Mymensingh town of the Brahmaputra River from where fish samples were collected. Out of 15 fish species purchased, 2 were collected from all locations and the rest 13 were collected from 2 locations, thus total 32 fish samples (2×3+13×2=32) were used for the present experiment. The details of sampling locations and fish species are presented in Fig. 1 and Table 1, respectively. A reasonable amount (500 g to 1.0 kg) of fish samples were purchased directly from the aforementioned locations of the river system, and requisite amount of samples were brought to the laboratory of the Department of Agricultural Chemistry, BAU, Mymensingh and processed for subsequent experiment.

2.2 Processing of Fish Samples

After collection, scales of fish samples were removed and cleaned first. Then the fish samples were separated into edible (mainly flesh, but it also includes bones and heads of small fishes) and non-edible part (heads, bones, scales, intestines, fins, gills and gill covers). After separation, edible part of fish samples were sun dried for 2 days, and then the samples were oven dried at $50-60^{\circ}$ C for another 2-3 days until a constant weight was obtained. After drying, the samples were ground well with the help of mortar and pestle, and then the ground samples were preserved in polythene bags with appropriate marking for further chemical analyses.

Fig. 1. Map shows the sampling sites of collected fish samples from the Brahmaputra River of Bangladesh (after Eti et al. [22])

SI. No.	Bengali name	English name	Scientific name	Name of locations
1.	Punti	Ticto barb	Puntius ticto	BB, BAUC and MT
2.	Tengra	Striped dwarf catfish	Mystus vittatus	BAUC and MT
3.	Bele	Tank goby	Glossogobius giuris	BB, BAUC and MT
4.	Bangna	Reba	Labeo ariza	BB and BAUC
5.	Baim	Zig-zag eel	Macrognathus aculeatus BAUC and MT	
6.	Shingi	Stinging catfish	Heteropneustes fossilis BAUC and MT	
7.	Kalibaush	Orange-fin labeo	Labeo calbasu	BB and MT
8.	Mola	Mola carplet	Amblypharyngodon molaBB and BAUC	
9.	Chela	Silver razor-belly minnow	Salmophasia bacaila	BB and MT
10.	Kaikka	Garfish	Xenentodon cancila	BB and MT
11.	Meni	Gangetic leaf-fish	Nandus nandus	BB and MT
12.	Chanda	Elongate glassy perchlet	Chanda nama	BB and MT
13.	Chingri	Prawns or shrimps	Macrobrachium sp.	BB and MT
14.	Guizza or Guizza ayre	Giant-river catfish	Sperata seenghala	BB and MT
15.	Hiralo or Murari --	Carplet	Aspidoparia murar $B4110 - B411 - \cdots - 11T - 11 - \cdots$	BAUC and MT

Table 1. Details of fish samples collected from the Brahmaputra River of Bangladesh

BB= Bhabakhali bazar; BAUC= BAU campus; MT= Mymensingh town

2.3 Preparation of Extract of Fish Samples

Ground fish samples were used to prepare extract for the determination of different heavy metals. Extract was prepared by wet oxidation method using di-acid mixture [23]. In this method, 0.5 g of finely ground samples were taken into a 250 mL conical flask and 5 mL of di-acid mixture $(HNO₃:HClO₄ = 2:1)$ was added to it. Then the flask was placed on an electric hot plate for heating at 180-200ºC temperature until the solid particles disappeared and white fumes were evolved from the flask. Then, it was cooled at room temperature, washed with distilled water and filtered into 100 mL volumetric flask through filter paper (Whatman No. 1). For quality control purpose, a blank extract was also prepared by taking same reagent without sample. Finally, the volume was made up to the mark with distilled water and preserved for the determination of different heavy metals in the fish samples.

2.4 Determination of Heavy Metals

Determination of different heavy metals (Cu, Pb, Cr, Cd, Zn and Mn) in prepared fish extracts were done by an atomic absorption spectrophotometer (AAS) (SHIMADZU, AA-7000; Japan). At first the AAS was calibrated followed by the manufacturer's recommendation. Then the extract was run directly in AAS for the determination of heavy metal in the samples. Hollow cathode lamp of Cu, Pb, Cr, Cd, Zn and Mn was employed as light source at wavelengths of 324.8, 283.3, 357.9, 228.8, 213.9 and 279.5 nm, respectively for the determination of each metal.

2.5 Estimation of Daily Metal Intakes (DMI)

To assess the human health risk associated with heavy metal contamination in the available fish species of the Brahmaputra River, the daily intake of metal was calculated with the following formula-

 $DMI = (IR \times C)/BW$

Where, IR is the fish ingestion rate (mg person $⁻¹$ </sup> day⁻¹), C is the individual metal concentration in edible parts of fish samples (mg kg^{-1} , fresh weight), BW is the body weight assuming 70 kg for adult male and 50 kg for adult female in the present study [24].

2.6 Metal Pollution Index (MPI)

To examine the overall heavy metal concentrations of fishes of the Brahmaputra River, the metal pollution index (MPI) was computed. This index was obtained by calculating the geometrical mean of concentrations of all the metals present in the fish samples [25].

MPI (mg kg⁻¹) = (Cf₁ × Cf₂ × ……. × Cf_n)^{1/n}

Where, Cf_n = Concentration of metal n in the sample.

2.7 Target Hazard Quotients (THQ)

THQ is calculated by the general formula established by the US EPA as follows-

$$
THQ = (E_F \times F_D \times DMI) / (RfD \times W \times T)
$$

Where, E_F is exposure frequency; F_D is the exposure duration; DMI is the daily metal ingestion (mg person⁻¹ day⁻¹) and RfD is the oral reference dose (mg kg^{-1} day⁻¹; W is the average body weight (kg) and T is the average exposure time for noncarcinogens (365 days year⁻¹ \times number of exposure years).

3. RESULTS AND DISCUSSION

3.1 Heavy Metal Contents in Different Fish Species

3.1.1 Copper (Cu)

Copper is an essential trace element for plants and animals. The human body only contains about 150 mg of this vital mineral. The established recommended dietary allowance (RDA) for Cu in normal healthy adults is 2 mg day $^{-1}$ [26]. The concentration of Cu in different fish samples collected from the Brahmaputra River varied from 3.19 to 47.91 μ g g⁻¹ (Fig. 2) with a mean value of 11.22 μ g g⁻¹. The highest amount of Cu (47.91 μ g g⁻¹) was obtained from *Chingiri*, which might be contribution of exoskeleton of this fish. The Cu mass in the

exoskeleton increased throughout the first 80 days of the growing cycle of shrimp [27]. The authors also stated that there was an inverse correlation between Cu concentrations in muscle and exoskeleton, suggesting an exchange of Cu from muscle to the exoskeleton. Concentration of copper in fish species collected from Chanoga, Okovango Delta of Botswana varied between 0.02-0.21 mg 100 g^{-1} [28], which was much smaller than the present study results. According to US EPA Cu is highly toxic in aquatic environments and has effects in fish, invertebrates, and amphibians, with all three groups equally sensitive to chronic toxicity [29]. However, it is evident from Fig. 2 that 3 fish species contained higher and the other 3 contained almost same amount of Cu as described by FAO standard for fish and fishery products in Australia [30].

3.1.2 Lead (Pb)

Lead is an extremely common metal in our daily life. Water, paint, electric storage batteries, insecticides, and gasoline are considering some common sources of Pb. It is rapidly absorbed into the bloodstream of human body through inhalation, ingestion, or by skin contact [31]. The amount of Pb in different fish samples collected from the Brahmaputra River varied from 2.20 to 3.73 μ g g⁻¹ (Fig. 3) with an average value of 3.01 µg g⁻¹. The maximum concentration of Pb was obtained from the fish species, *Mola*

Fig. 2. Copper content (µg g−1 **) in different fish species collected from the Brahmaputra River of Bangladesh. Each value is the mean for three replicates, and vertical bars indicate the standard errors**

followed by *Tengra*, *Chingri*, *Chanda* and *Meni*. Concentrations of Pb in two commercial fish species of four Turkish seas were 4.48 and 5.20 μ g g⁻¹ (wet wt.) in red mullet and whiting, respectively [32] and their result was slightly higher than the contents obtained by the present study. In fact, there are no any known health benefits or biological role of lead for the human body. On the contrary, lead has adverse effects that deleterious to the human body. It can affect almost every organ and system in the human body [33]. Although there is no safe level of exposure to lead has been found, chronic toxicity of it is much more common and occurs at blood levels of about 40-60 μ g dL⁻¹ [31]. All fish species obtained from the Brahmaputra River contained higher amount of Pb as described by FAO standard for fish and fishery products in Australia [30] (Fig. 2).

3.1.3 Chromium (Cr)

Chemically, trivalent Cr is non-toxic and necessary for humans (trace amounts), while the hexavalent form is toxic. In 2001, dietary reference intakes for chromium were established and an adequate intake of chromium is 35 mg day⁻¹ for adult males and 25 mg day⁻¹ for adult females [34]. The amount of Cr in different fish samples collected from the Brahmaputra River ranged from 0.59 to 2.40 μ g g⁻¹ (Fig. 4) with an average value of 1.10 μ g g⁻¹. Cr concentrations in muscle and liver tissues were higher in red mullet collected from Mersin Bay than that of all other stations of Turkey [32] and also the permissible limit (1.0 µg g^{-1} (wet wt.) of FAO [30]. But the average concentration of Cr obtained by the present study was higher than the permissible limit of FAO. However, out of 15 fish species, 7 species contained higher amount of Cr than the standard for fish and fishery products (Fig. 4). 50% lethal concentration value of chromium for the potassium dichromate salt is 41.75 mg L-1 in *Channa punctatus* (Bloch) [35] while the value for Cr is reported to be 39.40 mg L-1 in case of *Labeo rohita* [36].

3.1.4 Cadmium (Cd)

Cadmium is a heavy metal with a high toxicity. It is toxic at very low exposure levels and has acute and chronic effects on human health and environment. The average Cd concentration in different fish species of the Brahmaputra River was 0.70 μ g g⁻¹ and the range varied from 0.19 to 1.20 μ g g⁻¹ (Fig. 5). The Cd concentration in fishes was in the sequence of *Kalibaus* > *mola* > *tengra* = *bangna* = *shingi* = *kaika* > *meni* > *punti*

> *chanda > bele* = *guizza ayre* = *hiralo* > baim = chingri > *chela*. The fish species contained the highest amount of Cd while the lowest was found in *Chela*. Cadmium accumulates in the human body and especially in the kidneys. According to the current knowledge kidney damage (renal tubular damage) is probably the critical health effect of Cd [37]. However, the World Health Organization has established a provisional tolerable weekly intake of cadmium and the value is 7 μ g kg⁻¹ body weight [38]. The International Agency for Research on Cancer (IARC) classified cadmium in "Class 1", which means the agent is carcinogenic to humans [39]. Concentrations of Cd in red mullet and whiting fish species varied from 0.01-0.40 and 0.01-0.24 µg g⁻¹, respectively [32]. The standard limit of Cd in fish and fishery products as described by FAO is 0.2 μ g g⁻¹ in Australia [30]. The Cd concentration in only 3 fish species obtained from the Brahmaputra River was within this standard limit and the limit exceeded in the rest 12 species (Fig. 5).

3.1.5 Zinc (Zn)

Zinc concentration in different fish samples collected from the Brahmaputra River ranged from 38.98 to 279.08 μ g g⁻¹ (Fig. 6) and the mean value was 101.97 μ g g⁻¹. The highest amount of Zn was obtained in the fish species *Chela*, followed by *Mola*, *Chanda* and *Chigri*. Relatively lower content of Zn (0.64-0.81 mg 100 g-1) was reported for farmed fishes in China [40]; 0.13 -2.5 mg 100 g⁻¹ for market fishes in French [41]; 0.57-1.3 mg 100 g^{-1} for lake fishes in Turkey [42]. The established RDA for Zn is 8.0 mg day⁻¹ for women and 11.0 mg day⁻¹ for men [43]. In the present study, average concentration of Zn in edible part of different fish samples collected from the Brahmaputra River was higher compared to all these reports published earlier. It is also evident from Fig. 6 that all fish species except *Shingi* (*Heteropneustes fossilis*) contained higher amount of Zn than the standard limit for fish and fish products of New South Wales and South Australia [30]. Zn is the second metal present in the human body (about 2.5 g), after Fe (about 4.0 g). It is a component of many metallo-enzymes, important for gene expression and cellular growth [44]. It is an essential trace element that functions as a cofactor for certain enzymes involved in metabolism and cell growth, which is found in nearly 300 specific enzymes. It is particularly important for healthy skin and for a healthy immune system and resistance to infection [45].

Fig. 3. Lead content (µg g−1 **) in different fish species collected from the Brahmaputra River of Bangladesh. Each value is the mean for three replicates, and vertical bars indicate the standard errors**

Fig. 4. Chromium content (µg g−1 **) in different fish species collected from the Brahmaputra River of Bangladesh. Each value is the mean for three replicates, and vertical bars indicate the standard errors**

Fig. 5. Cadmium content (µg g−1 **) in different fish species collected from the Brahmaputra River of Bangladesh. Each value is the mean for three replicates, and vertical bars indicate the standard errors**

Fig. 6. Zinc content (µg g−1 **) in different fish species collected from the Brahmaputra River of Bangladesh. Each value is the mean for three replicates, and vertical bars indicate the standard errors**

3.1.6 Manganese (Mn)

Manganese is a trace mineral that is present in tiny amounts in the human body. It is one of the most important nutrients for human health. The average human body contains about 12.00 mg of Mn and about 43% of it is found in the skeletal system [46]. Mn helps the body to form connective tissue, bones, blood-clotting factors and sex hormones [47]. Mn concentration in different fish samples ranged from 41.70 to 376.62 μ g g⁻¹ (Fig. 7) with a mean value of 128.24 μ g g⁻¹. The highest amount of Mn was obtained in the fish *Mola*, followed by *Bele, Hiralo* and *Chanda*. These results of Mn content in the fish samples of the Brahmaputra River were much higher than the results reported in Turkey (0.08 to $7.33 \mu g g^{-1}$) [32]. The Mn content in all samples were also higher than the permissible limit of food set by FAO/WHO (540 µg 100 g food) [48] and the RDA value for Mn (2.3 mg dayfor adult males and 1.8 mg day $^{-1}$ for adult females) [34]. Concentrations of manganese found in tissues of marine and freshwater fish tend to range from <0.2 to 19.0 μ g g⁻¹ dry weight [49]. If we consider the upper limit of this range as standard, Mn content in all fish samples of the Brahmaputra River exceeded this limit (Fig. 7).

3.2 Assessment of Metal Pollution Index (MPI)

The metal pollution index (MPI) was used to compare the total metals accumulation level in various tissues of different fish species. In spite of indisputable importance of established chemical, biochemical and biological methods, MPI might be included in complex freshwater monitoring programmes since it could produce some additional information on metal bioavailability, bioconcentration and metal input into the environment [50]. The MPI is a reliable and precise method for monitoring metal pollution in food samples. The MPIs of individual fish samples collected from the Brahmaputra River are presented in Fig. 8. Among the fish samples used in the present study, *mola* showed the highest MPI (20.12) followed by *chela* (17.72), *chanda* (16.78) and *chingri* (15.83), but the lowest MPI was obtained for the *baim* (6.79) fish (Fig. 8). MPI in fishes collected from Taihu lake in China ranged from 0.2-2.5 [51], which was very much smaller than the present investigation. On the other hand, calculated MPI showed that the total metal load in fish organs followed the sequence as: kidney $>$ liver $>$ gill $>$ muscle, which gives a better idea about the target organs for metal accumulation in fish [52].

Fig. 7. Manganese content (µg g−1 **) in different fish species collected from the Brahmaputra River of Bangladesh. Each value is the mean for three replicates, and vertical bars indicate the standard errors**

Fig. 8. Metal pollution index (MPI) for edible part of different fish species collected from the Brahmaputra River of Bangladesh

3.3 Estimation of Daily Metal Intakes (DMI)

To evaluate the health risk associated with heavy metal through consumption of available common fishes in the Brahmaputra River, the daily intake of metals was calculated. There are several possible pathways of exposure of metals to humans, but the food chain is the most common. The daily intake of metals was calculated according to the average fish consumption for both adults male and female. According to HIES fish ingestion rate in Bangladesh is 62.58 g person $^{-1}$ day⁻¹ [53], which is used to calculate DMI. The daily metal intakes estimate of Mn, Zn, Cu, Cr, Cd and Pb from edible part of fish samples were calculated by multiplying the daily intake by the metal concentrations determined in this study. The DMI were compared with the upper tolerable daily intakes for each metal. It is evident from Table 2 that daily metal intakes for Pb, Mn and Cd for both male and female were higher than that of upper tolerable intake level, which indicates serious adverse effects have been associated with intake of common fishes of the Brahmaputra River.

3.4 Target Hazard Quotient (THQ)

Target hazard quotient is a complex parameter used for the estimation of potential health risks associated with long term exposure to chemical pollutants. The THQ <1 means the exposed population is assumed to be safe; $1 < THQ < 5$ means that the exposed population is in a level of concern interval, and THQ > 5 means exposed population is unsafe [59-60]. THQ was measured considering DMI of people, average body weight (male: 70 kg and female: 50 kg) and average life expectancy (male: 70.6 and female: 73.1) [24]. The calculated values of THQ due to consumption of edible part of common fishes available in the Brahmaputra River for investigated metals are presented in Fig. 9. THQ values for Mn surpassed 5.0 for both male and female, and the same value for Pb also surpassed 5.0 for female indicates that the exposed populations are unsafe. It is also worth mentioning from the results that all individual THQ values of other metals for both male and female surpassed 1 (except Cu for men), which means exposed populations are in level of concern interval.

Fig. 9. Target hazard quotient (THQ) for both male and female due to consumption of common fishes available in the Brahmaputra River of Bangladesh

4. CONCLUSION

This study results revealed that the Brahmaputra River has a diversity of different fish species, which are very popular to the local people of the country. But, higher amount of heavy metals particularly Pb, Mn and Cd are accumulated in edible part of obtained fish species, which make them unsafe for human consumption. However, more representative data can be obtained if we could collect samples from the whole river system along with large scale of fishes. Finally, present study result concluded that human health risk due to consumption of available fish species of the Brahmaputra River is not safe as regards to Mn and Pb. Therefore, it is important to monitor river water and sediment quality because continued discharged of waste water, sewage and other garbage to the river system is likely to increase the magnitude of different heavy metals

and metalloids which may accumulate in river fishes and finally contaminate human food chain. Thus, people and concern authorities should come forward to prevent pollution and save aquatic environment of the river.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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