ANALYSES OF CONTAMINATION OF THE GANGA RIVER WATER AND ITS IMPACT ON THEIR FISH IN UTTAR PRADESH REGION

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SUMMARY

The Ganges is India's largest (2,525 km) river, arises in the western Himalayas, flows south, and empties into the Bay of Bengal. The main townships at the Bank of Ganga at Uttarakhand and Uttar Pradesh (UP) are Rishikesh, Haridwar, Kanpur, Allahabad, Varanasi, and Mirzapur. It is one of the most sacred rivers and is worshipped as the goddess *Ganga*. It serves as the lifeline for a population of over 600 million people. Its water was treated as 'amrit'. Unfortunately, it has become the world's 5th most polluted river (Hamner et al., 2006). The primary sources of the Ganga River pollution are effluents coming from municipal sewage, industrial discharges, oil refineries, tanneries, textiles, paints, and agricultural runoffs. As a result, the Ganga River water (GRW) has been extensively contaminated with several xenobiotics. The Ganga River's metal pollution is a great public concern because it could reach millions of people from the aquatic ecosystem via drinking, domestic, ritual and agricultural and aquatic animal food utilization. However, the amount and diversity of metals in the Ganga River depend directly on the increasing population living on its bank, urbanization, industrialization, and topological and seasonal changes. It has been reported that the pollution load on the Ganga River is maximum (76%) in UP. Since, in UP, the Ganga River traverses a maximum distance (~ 1000 km) than the rest of the Indian states. Therefore, massive amounts of metal discharges via industrial and sewage effluents could harm the hydrochemistry and biological integrity of receiving GRW in UP. Hence, efforts were made in this presentation to analyze the contamination of the GRW (mostly metal contaminants) in the UP region and their detrimental effects on the fish tissues. Fish at the higher trophic level in the aquatic ecosystem suffers severely due to the high toxic load of metals in water. Metal

bioaccumulation in their tissues could cause a considerable decline in their catches from polluted rivers regarding poundage and numbers. Simultaneously the taste of this fish has also deteriorated markedly.

In current work, the GRW sampling was performed during January 2019-2020 and March 2021 to May 2022 from three sites in each of the four selected cities of UP, namely Bithoor (S 1), Ganga Barrage (S 2), and Jajmau (S 3) in Kanpur, Rasulabad (S 4), Teliarganj (S 5) and Shastri Bridge (S 6) in Prayagraj, Bhagwanpur (S 7), Assi Ghat (S 8) and Manikarnika Ghat (S 9) in Varanasi, and Pakka Pul (S 10), Pakka Ghat (S 11) and Bariya Ghat (S 12) in Mirzapur covering about 370 km stretch of the Ganga River in order to invest its physicochemical properties and metal contaminants. Three locations in each city were selected based on a prior survey to understand the majority of polluting sources directly contributing to river pollution at these places. One additional site, namely Katarijiyora near Ganga Barrage, Kanpur, where the sewage effluent gets discharged directly into the main river, was also considered for the examination of the GRW quality to test the claim that sewage effluent liberation in GRW is one of the primary contributing sources of metal pollution of GRW in Kanpur, UP. The physicochemical properties like temperature, pH, electrical conductivity (EC), total acidity (TAc), total alkalinity (TA), dissolved oxygen (DO), biological oxygen demand (BOD), total hardness (TH), and total chloride were analyzed following the standard methods for the examination of water and wastewaters (APHA/AWWA/WPCF, 1998) while the concentrations of eleven metals (Al, As, Pb, Cr, Cd, Zn, Cu, Ni, Co, B, and Be) were analyzed in the GRW samples collected from all twelve sites in UP using Inductively coupled plasma-mass spectrometry (Agilent 7900 ICP-MS) from the Central Research Facility (CRF), Indian Institute of Technology (IIT), Delhi. It was observed that the pH of the GRW changed gradually from slightly to strongly alkaline downstream from Kanpur to Mirzapur. The temperature and pH of the GRW samples ranged between 22.20 - 30.63°C and 7.73 - 8.77, respectively, at all the studied sites. The amounts (mg/L) of DO, BOD, TC, TH, TA, TAc, TDS, and EC (µS cm^{-1}) of the GRW samples at all the sites of respective cities ranged between 3.87-10.93, 1.47 - 7.33, 9.33 - 39.99, 32.67 - 97.33, 8.00 - 19.33, 14.67 -38.67, 103.33 - 229.33, and 209.67 - 471.67, respectively. The values of TC, TH, TA, TAc, and TDS were below their permissible levels recommended by BIS (2012). Similarly, the mean contents (mg/L) of DO and BOD of the GRW samples at every site were noted to be as per the recommended guidelines of BIS (2012) except for S 9 (Varanasi). At S 9, Varanasi, the concentrations of DO and BOD were 3.87 and 7.33, respectively, which were found not in compliance with the standards prescribed by BIS (2012). In addition, the EC values of the river water samples at most of the studied locations concerning the cities were noticed to be significantly ($p < 0.05$) higher than their permitted limits (BIS, 2012), excluding two levels sites, namely S 1 (228.00) and S 2 (209.67) at Kanpur. In addition, the GRW samples at Katarijiyora, Kanpur, UP also had low DO and high alkalinity. The elevated EC values at most sampling places indicate contamination of the Ganga River with higher numbers of dissolved charged chemicals that might decline species richness and abundance of fish from the Ganga River at these locations (Zhang et al., 2019). Though, values of pH, DO, and EC of GRW at these locations were within the recommended range of 6 - 8.5, DO > 5 mg/L, and EC < 2000 µS cm^{-1} proposed by CPCB (2020) for suitability for bathing and irrigation purposes, still contamination of the GRW samples with various toxic metals contradict with their suitability for application. At Kanpur, GRW samples were most polluted at S 2 compared to both S 1 and S 3 sites because concentrations of Pb, Al, Ni, Cd, As, Cr, Co, and Zn were 13300%, 8885%, 1760%, 666.67%, 383%, 218%, 215% and 48.87% higher than their BIS (2012) standards value respectively. At S 1, the concentration of only As was 56% above BIS (2012) permissible limits, while at S 3, two metals: Ni and As, were 660% and 54% more than their BIS (2012) levels, respectively. According to the HPI analysis, the GRW was critically polluted at S 2 in Kanpur mainly by Pb, Cd and Al. The HPI values were 7398.96 > 100 standard HPI at S 3 and unfit for any use compared to S 1 (HPI-100.71) and S 3 (HPI-58.34), which could be utilized for domestic and agricultural purposes. Further, at Prayagraj, the GRW samples were polluted mainly with Al and Cr. At S 4, S 5, and S 6 sites of Prayagraj Al and Cr were respectively 1470%, and 753%, 1582.5% and 3580%, 550% and 4730% greater than their recommended safe limits (BIS, 2012) while at S 6, in addition to Al and Cr, Ni and Pb were also 60% and 90% higher than their BIS (2012) values respectively. Rest all other elements were within the prescribed (BIS, 2012) limits. Further, analysis of individual HPI levels of metals in the GRW samples at Prayagraj revealed that S 6 (378.503) was the most polluted site, followed by S 4 (330.293) and S5 (133.898) mainly due to high levels of Cr in the GRW. At Varanasi, in addition to Al and Cr, concentrations of Pb in the GRW samples were significantly higher than their BIS (2012) recommended values at all three sites. The quantities of Pb, Cr, and Al at S 7 were 9232.14%, 5020%, and 685%, respectively; at S 8, 6992.74%, 3420%, and 680%, respectively and at S 9, 2371.68%, 900% and 150.15%, respectively higher than their BIS (2012) levels. At S 7 and S 8 sites, Ni amounts were also found to be 165% and 40% greater than their recommended BIS (2012) limits, respectively. Like

Prayagraj, the HPI analysis revealed that the GRW at Varanasi city were also highly polluted with Pb, Cr, Al, and Ni. Further, at Mirzapur also, Pb and Cr were mainly contributing to the GRW at all sites. At S 10, S 11, and S 12 (Mirzapur), Cr amounts were respectively 2016.82%, 4581.92%, and 7079.60% greater than their BIS (2012) limits while Pb were 2691.11%, 5404.35% and 3999.33%. Further, Al levels were 366.71% and 180.07% greater than their BIS (2012) limits at only S 11 and S 12, respectively. Moreover, the HPI values of metals indicate that the GRW at S 11 was more polluted than S 10 and S 12 sites at Mirzapur in UP due to Cr and Pb. Moreover, the GRW at katarijiyora, Kanpur, were also contaminated with Pb. The metal amounts in the GRW at katarijiyora, Kanpur, followed the trend of Pb > Cu > Zn > Fe > As > Co > Cr > Cd. Thus, it could be concluded that among eleven metals, the GRW was severely polluted with Pb, Cr, Al and Ni (comparatively less than the other three metals) in UP. To evaluate the toxicity of metals in humans, certain health risk assessment indices were calculated. Their values for humans via dermal exposure revealed no health issues due to contamination of the GRW with heavy metals like Pb, Cr, Al and Ni in UP. However, at S 2 (Kanpur), $CDI_{ingestion}$ values of Al (1.52) and Zn (1.31) and $HQ_{ingestion}$ of Pb ad As in the GRW were beyond desired limits of 1 in both age groups thus, indicate ingestion of GRW at S 2 is harmful to human health. In addition, $CR_{ingestion}$ values for most metals like Cd, Cr, Co, As, Ni, and Pb in the GRW at Kanpur for the child and adult exhibit higher values than the Carcinogenic Risk (CR) safe range. Moreover, only at S 4 (Prayagraj) $HQ_{ingestion}$ values of the As were greater than one in children and adults. In the child, its values were 2.73, 1.01, and 4.44 at S 4, S 5, and S 6, while in the adult, its values were 2.52 and 4.11 at S 4 and S 6, respectively. In addition, the $CR_{ingestion}$ values of Cr, Cd, and As for both the child and adult were noted to be greater than the CR recommended range at all three sites of Prayagraj. Along downstream at Varanasi, $HQ_{ingestion}$ values of two metals such as Pb and As, were higher than one for both children and adults at S 7 and S 8, respectively. At the same time, at Mirzapur, $HQ_{ingestion}$ values of Pb in the GRW exceeded the safe limit of one for both the population group (child and adult). In contrast, $CR_{ingestion}$ values of Cr, Cd, Co, Ni, and As were higher than their CR safe range for both Varanasi and Mirzapur for both children and adults. Hence, the present findings also illustrate the toxic health impact of metal pollution of the GRW at different studied places in UP on both children and adults. Furthermore, higher levels of $HQ_{ingestion}$ and $CR_{ingestion}$ in children indicate that children are more vulnerable to various health risks than adults. Thus, the present study suggests that detoxification of the GRW in UP should be prioritized along with improvements in its physicochemical parameters to improve the quality of Ganga water to save flora and fauna, including its fish population in UP.

Increased metal concentration in the river could cause higher bioaccumulation of metals in the fish than in water. It thus could make them unsuitable for consumption & loss of their nutritional value. Therefore, bioaccumulation analysis of metals in fish tissues along the different stretches of the Ganga River in the UP region was also performed. Bioaccumulation of metals (Be, B, Al, Cr, Co, Ni, Cu, Zn, As, Cd and Pb were analyzed in the various tissues (muscles, liver, kidneys, and gills) of fishes of the Ganges River, namely *Labeo rohita*, *Rita rita*, *Pangasius pangasius* and *Ctenopharyngodon idella* caught from the GRW at different sampling sites of UP. It was observed that bioaccumulation of the metals varied with different fish species (*L. rohita, R.rita, P. pangasius and C. idella*), their tissues (liver, kidneys, muscles, and gills), and catching sites S 1, S 2, S 3 in Kanpur, S 4, S 5, S 6 in Prayagraj, S 7, S 8, S 9, in Varanasi, and S 10, S 11 and S 12 in Mirzapur at UP. Further, no permissible limits were found for Be, B, Al and Co in fish and fishery products. Therefore, their suitability for human consumption were not further discussed. The bioaccumulation of Be in all fish tissues were less than the rest of the examined metals in fish tissues at S 4 and S 6 sites of Prayagraj; however, at S 5, it was higher in the muscles of *R.rita* as well as in muscles and liver of *P. pangasius* . At Varanasi and Mirzapur however, Be deposition in all the tissues of fish were lower compared to rest of the three cities of UP. Similarly, B accumulation is also considerable in the muscles of fish. At Kanpur, B deposition was higher in the muscles of *R. rita* (1.48), *L. rohita* (17.00), of *P. pangasius* (0.98) compared to the remaining fish tissues. In contrast, at Prayagraj, B deposited highest in the gills (0.728) of *L. rohita* at S 5 and in the muscles of *P. pangasius* (1.35) at S6. At Varanasi, B highest deposition was noticed in the liver of *R. rita* (0.073) at S 7, and at S 8 in the kidneys of P. pangasius (0.736) while at S 9, it was the kidneys of *L. rohita* (0.95). Further, at Mirzapur, the highest B deposition was noted in the gills of *R. rita* (0.90) at S 10, in the gills of *P. pangasius* (1.026) at S 11, while least B accumulation was noticed in fish tissues at S 12. In contrast to Be and B, accumulation of Co was greater in the liver, kidneys and gills of fish compared to muscles at each of three sites of four cities in UP. Compared to WHO (1985) safe limits of Ni (0.5 mg/kg) in fish, Ni accumulated beyond their safety levels in the liver of *P. pangasius* (2.44) at S 1 and the gills (9.22) as well as in the liver (0.95) of *L. rohita* at S 2 and S 3, respectively while high levels of Ni were observed in the liver of *L. rohita* at S 5 and S 6 at Prayagraj, respectively. In contrast, at Varanasi, the accumulation of Ni in all fish tissues was below the toxic levels compared to WHO

(1985). At Mirzapur, however, Ni deposited highest in tissues of *L. rohita* and *R. rita* than WHO (1985) permitted limits. Additionally, in comparison to safe levels of Cu (10 mg/kg) in fish given by FAO (1983), Cu accumulation in every tissue of all studied fish caught from the GRW in UP was much below their permissible limits allowed in fish except *R. rita* caught from the GRW at Katarijiyora, Kanpur, where Cu deposited above the safe levels in their muscles and gills (12.000 and 12.476, respectively). Similarly, Zn deposition in each fish tissue at all sites of four cities was observed to be much below their harmful levels of 50 mg/kg prescribed by FAO (1983). Further, except for muscles of *R. rita* (3.00) at S 1 (Kanpur), Al deposition was recorded only in the liver, kidneys and gills of all three fish at each studied city in UP. Similarly, accumulation of Cr was also noticeable in the gills, liver, and kidneys of *L. rohita*, *R. rita* and *P. pangasius* at all sites of four cities in UP. However, Cr deposition was beyond their safe levels {1 mg/kg, FAO (1983)} in most of the tissues of fish caught from the GRW at Kanpur, Prayagraj, Varanasi and Mirzapur in UP. In addition to Cr, the As accumulation in muscles of *P. pangasius* at S 3 (Kanpur) and S 6 (Prayagraj) were above the permitted levels of 0.1 mg/kg recommended by FAO (1991), while its deposition in the gills, liver and kidneys of all fish at each site were also noteworthy in terms of their probable potency to cause a human health risk. Meanwhile, except Pb, among all remaining studied elements, Cd accumulation was noticed highest in kidneys (65.51) of *P. pangasius* caught from Kanpur sites. Also, it was interesting to note that Cd deposited above the safe levels of 0.05 mg/kg prescribed by FAO (1991) in most of the tissues of *R. rita* and *P. pangasius* caught from the GRW at sites of UP, but its accumulation in tissues of *L. rohita* were lower at Varanasi and Mirzapur than Kanpur and Prayagraj. However, at Katarijiyora, Kanpur, and Prayagraj, Cd in muscles was found beyond the allowed levels in *C.*

idella and *R. rita*, respectively. Therefore, the results revealed that metal contamination of fish varied with the water qualities in particular cities suffered differently from the drainage of various effluents. The Pb accumulation in fish tissues is highly toxic for fish and humans because of their neurotoxic effects. In the present work, among all metals, Pb was deposited in maximum amounts in all tissues of fish caught from the GRW from all sites of Kanpur. It was found highly deposited in the liver of *P. pangasius* (90.88) at S 2, followed by the gills of *R. rita* (22.38) at S 1 and the kidneys of *L. rohita* (20.56) at S 3. Furthermore, Pb accumulation in the muscles of *R. rita* at Prayagraj as well as in *L. rohita* and *R. rita* at Mirzapur was beyond their permissible limits of 0.3 mg/kg recommended in fish (FAO, 1991). Thus, the present study showed that fishes caught from the GRW in UP were contaminated with Pb, Cd, Cr, and As and might be suffering badly due to the metal pollution of the GRW. Besides HM accumulation analysis, the BAF values of metals in the tissues of fish caught from the GRW showed that elements bioaccumulate significantly in greater amounts than their quantities found in the water at different places in UP. Additionally, even though BAF values of Pb, Cr, Cd, Al, Fe and Cu were relatively higher for most of the tissues in all fish species (*L. rohita*, *R. rita, P. pangasius*, and *C. idella*), significant bioaccumulation of Be, B and Co in fish tissues compared to their lower amounts in the GRW at studied at various sampling locations in UP indicates great health concern for fishes living in contaminated conditions of water.

Fish is not only one of the primary sources of proteins and other nutritional elements. They also constitute a greatly palatable component of our daily meals. Hence, efforts have been made to understand the reason for the decline in the quantity and taste of the fish caught from the Ganga River, which got dangerously polluted in the examined cities of UP. This was done by analyzing and comparing the changes noticed in the nutritional composition of muscles of the fish caught from the GRW at locations similar to the river water sampling in UP. The main biomolecules that decide the taste of fish are proteins, lipids, and glycogen. Accumulation of metals & other toxicants in fish tissues could often disturb the equilibrium between these important biomolecules and alter their taste and nutritional value, which are also primary concerns for human health. Irrespective of sex, healthy specimens of three different fish species, namely *R. rita*, *L. rohita*, and *P. pangasius*, were collected from the Ganges River from three sites of each of the four different cities covering a stretch of 370 km from Kanpur, Prayagraj, Varanasi and Mirzapur in UP, India during 2019 to 2022 in the six months interval gaps and were collated together for analysis. The average length of fish ranged from 15 ± 5 cm and weighed 150 ± 15 g, respectively. The fish were sacrificed, and their muscles were subjected further to analyze total protein, glycogen, and lipid concentrations. The total concentrations (mg/g wet wt. of tissue) of protein, glycogen, and lipid in fish muscles were estimated following the methods of Lowry et al. (1951), Carroll et al. (1956), and Folch and Stanely (1957), respectively. The value of total protein content in muscles of *R. rita, L. rohita*, and *P. pangasius* ranged between 58.00 ± 10.92 to 183.75 ± 16.93, 83.85 ± 7.72 to 220.54 ±7.56, and 74.80 ± 7.39 to 178.68 ± 13.71, respectively among the different sampling locations of cities in UP. It was noticed that protein levels in the muscles of all three fish of the Ganga River were greater at Varanasi, followed by Mirzapur and Prayagraj, compared to Kanpur (Upstream taken as a control for comparison). It could be due to the extensive metallic pollution of the Ganges and its fish inhabitants at Varanasi and Mirzapur than upstream at Kanpur. Similarly, the value of glycogen in muscles of *L. rohita, R. rita,* and *P. pangasius* ranged between 12.30 ± 1.06 to 45.14 ± 5.36; 12.87 ± 1.36 to 44.89 ± 5.33 and 12.64 ± 2.82 to 42.69 ± 7.30, respectively among the different sampling locations of cities in UP. The value of muscle glycogen in *L. rohita* was highest, followed by *R. rita* and *P. pangasius*. However, at the place of most contaminated sites of Varanasi and Mirzapur, muscle glycogen content declined significantly ($p < 0.05$) in fish compared to those caught from relatively less polluted Ganges at upstream Kanpur and Prayagraj. Meanwhile, the values for the total lipid concentration in muscles of *L. rohita*, *R. rita,* and *P. pangasius* ranged between 3.25 ± 0.02 to 9.72 ± 0.73; 2.16 ± 0.04 to 6.79 ± 0.98; and 2.7 ± 0.01 to 7.15 ± 0.62, respectively among the different sampling locations of cities in UP. It was observed that lipid contents in Varanasi and Mirzapur fish muscles were significantly ($p < 0.05$) less than the lipid concentration of muscles in fish caught from upstream Ganga at Kanpur and Prayagraj. Thus, the current work illustrates significant alterations in the different biochemical profiles of fish muscles that could lead to their altered physiology and nutritional value. Hence, the discharge of this toxic effluent in the Ganga River should be prohibited. Further, contamination of the GRW and its impacts on fish should also be regularly monitored to give any emergency advisory to protect the fish species from metal pollution and, thereby, human health.