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Bioaccumulation and human health risk assessment of heavy metals in fish cultured in metropolitan city lakes: A case study in Bengaluru, South India

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ABSTRACT

Food safety is a global concern, yet little research has focused on the heavy metal (HM) pollution and health risks associated with both cultured and captured fish. This study evaluates the bioaccumulation and human health risks of HM (Cd, Cu, Zn, Cr, and Pb) in various tissues of aquacultured fish, Oreochromis niloticus, from major lakes in Bengaluru. Over two years (August 2021 to July 2023), spanning different seasons, fish samples were collected and analyzed for heavy metal presence using Atomic Absorption Spectrophotometry (AAS). Bioaccumulation levels were assessed, along with Estimated Daily Intake (EDI), Target Hazard Quotients (THQ), Hazard Index (HI), and Target Risk (TR). Results showed varying bioaccumulation patterns across different tissues and lakes. The highest concentrations of heavy metals were found in gonad and liver tissues HI values for all muscle tissue were>1, indicating potential health risks from metal consumption. TR levels were found to be unacceptable in the case of Cd and Cr which may lead to the risk of cancer and damages the normal functionality of the kidneys and liver. This study highlights the health threats associated with consuming freshwater fish from Ulsoor and Agara lakes in Bengaluru, underscoring the importance of regular lake monitoring to mitigate heavy metal impacts on human health and the ecosystem.

1- Introduction

Freshwater ecosystems, particularly lakes, play a pivotal role in providing essential habitats for a diverse array of fish species. However, the proximity of these ecosystems to human settlements and industrial activities exposes them to the pervasive threat of heavy metal contamination [1]. This concern is particularly acute in urban lakes situated within metropolitan areas, where fish populations face heightened risks due to prolonged exposure and the gradual bioaccumulation of toxic metals, such as mercury, lead, cadmium, arsenic, copper, and other hazardous heavy metals throughout the aquatic food chain [2] and poses severe risks to the well-being of fish populations in these environments [3].

Heavy metals can accumulate in fish through various pathways, primarily water, food, and sediment. Small aquatic organisms such as plankton and other invertebrates may ingest metals from water and sediment. Fish can accumulate metals over time when they consume these organisms [4]. Fish are effective bioindicators of heavy metal pollution in aquatic environments due to their constant contact with water. If the aquatic environment contains elevated levels of heavy metals, these metals can be absorbed directly through the fish's gills and skin [2, 5–7] and the accumulation of heavy metals can lead to various changes, including cytoplasmic vacuolation, disorganization, necrosis, sinusoidal dilation, and edema [8], multiple detrimental effects like genotoxicity, mutagenicity, immunosuppression, cytotoxicity, histopathological changes and reproductive abnormalities in fish [9]. This poses a potential health risk to humans when consuming contaminated fish through the food chain [10, 11].

Numerous studies have consistently reported the suboptimal water quality and metal accumulation in the water and sediment of Bengaluru lakes, attributing these concerns to diverse factors such as rainwater runoff, industrialization, sewage discharge, and other anthropogenic activities [12–15]. Despite this extensive exploration, the extent of bioaccumulation of heavy metals in fish populations within Ulsoor and Agara lakes remains inadequately documented. Furthermore, the fish sourced from these lakes are a significant component of the local diet, emphasizing the need for an in-depth investigation into the bioaccumulation of selected heavy metals, namely Cd, Cu, Zn, Cr, and Pb, within various tissues of aqua-cultured tilapia, *Oreochromis niloticus*.

Our previous study revealed that water and sediment samples collected from Ulsoor and Agara lakes are contaminated with heavy metals and are above the permissible limit [16]. This study seeks to fill the existing knowledge gap and aims to evaluate the level of bioaccumulation of heavy metals in fish sourced from Ulsoor and Agara lakes across different seasons and to assess their potential human health risk, if any, associated with the consumption of fish. Given the limited information available on heavy metal bioaccumulation in these lakes, this research aims to contribute valuable insights into the ecological and public health implications of consuming freshwater fish from Bengaluru lakes.

2-Materials and Methods

2-1- Study area

Two major lakes of Bengaluru, Ulsoor lake and Agara Lake (12.983°N 77.619°E & 12.921°N 77.641°E), were chosen as the focal points due to their location, aquaculture of *O. niloticus*. They were also susceptible to pollution resulting from heightened concentrations of hazardous elements, primarily due to the influx of rainwater runoff into the lakes. The specific sampling sites are depicted in Figure 1.

2-2- Fish sampling

Oreochromis niloticus samples (body weight 120 to 180 g), were collected randomly from Ulsoor and Agara Lakes of Bengaluru with the help of local fishermen using cast nets and other indigenous traps twice a month during the two consecutive calendar years (from August 2021 to July 2023) in different seasons such as pre-monsoon (PrM) (February to May), monsoon (M) (June to September), post-monsoon (PoM) (October to January). After collection, fish are transported to the laboratory promptly placing them in an ice box. Upon arrival, fish were dissected carefully with a metal-free ceramic knife and the vital tissues such as gill, muscle, kidney, liver and gonad were separated. The dissected tissues were washed with double-distilled water and put in Petri dishes reaching a constant weight and were sealed using polythene bags and stored at 20ºC until further analysis.

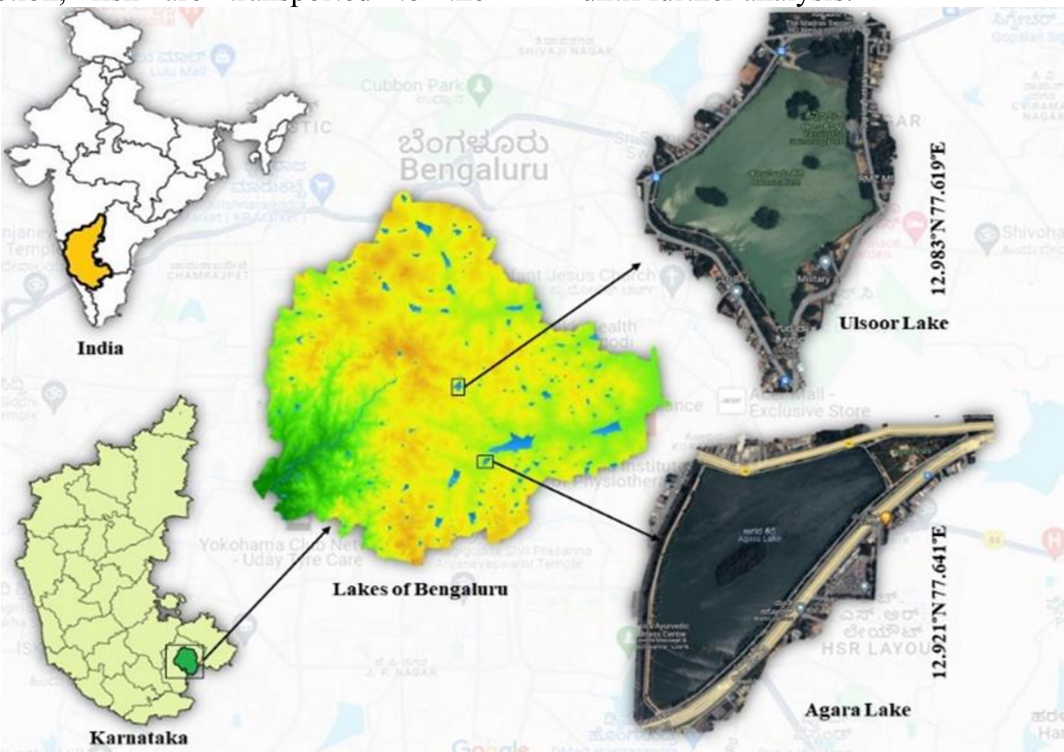

Figure 1: Location map showing Ulsoor and Agara lakes of Bengaluru

2-3- Heavy metal analysis

For heavy metal analysis, 1 g of each tissue, in 5 duplicates, was dried at 60 ºC for 24 h in a hot air oven (Beston Industries, Cochin, India) and ground into powder. Then, 1 g of the powdered tissue was taken, and a mixture of nitric acid and hydrochloric acid (3:1) was added and kept for 12 h [17]. Thereafter, the digestion process was performed on a hotplate at a temperature of 40 ºC for 2 h inside the fume hood. Once digestion was complete, the samples were diluted using Millipore water and filtered using Whatman's filter paper (No: 45) to remove any solid particles or impurities. The volume of the filtered solution was adjusted to 50 mL using a volumetric flask (AOAC 1990) [18]. The obtained solution was used to analyse the heavy metals using an Atomic Absorption Spectrometer (AAS: Shimadzu AA6880, Japan). The absorption wavelengths of selected heavy metals are as follows: Cd: 228.8 nm, Cu: 324.8 nm, Zn: 213.9 nm, Cr: 357.9 nm, and Pb: 220.35 nm. The results were expressed as mg/kg on the dry weight for fish tissue.

2-4- Potential health risk assessment

2-4-1 Estimated daily intake

The Estimated Daily Intake (EDI) of heavy metals from eating the fish edible tissue is based on the mean concentrations of heavy metals in the fish samples and the average daily consumption. EDI (mg/kg/day) has been calculated using the equation:

$$\mathrm{EDI} = \frac{\mathrm{DFC} \times \mathrm{C}}{\mathrm{WAB}}$$

Where DFC represents daily food consumption; C represents metal concentration in the fish sample, and WAB is average body weight. Since specific regional daily consumption rates for fish among men and women were unavailable, the study adopted assumed values of 29.41g/person/day for men and women. Additionally, the average body weights of 65kg for a man and 55kg for a woman were considered.

2-4-2 Target hazard quotient

The evaluation of health risks linked to the consumption of fish species involved the utilization of the target hazard quotient (THQ). The calculations were conducted following the established approach for comprehensive risk analysis by the US Environmental Protection Agency as outlined in their guidelines (USEPA 2010).

$$THQ = \frac{EF \times ED \times FIR \times C}{RfD \times WAB \times TA} \times 10^{-3}$$

In the assessment, the variables were as follows: EF represented Exposure Frequency (365/days/year), ED stood for Exposure Duration (equivalent to the average lifetime, based on [19], FIR represented Food Ingestion Rate (29.41kg/person/day), C denoted the metal concentration in fish tissues (mg/kg), RfD referred to the oral reference dose (mg/kg/day), WAB represented average body weight (65kg for men and 55kg for women), and TA indicated exposure time for non-carcinogens (365 days/year×ED). According to the USEPA (1997) [20], the oral reference doses for Cd, Cu, Zn, Cr and Pb were 0.001, 0.04, 0.03, 1.3 and 0.004 mg/kg/day, respectively.

2-4-3 Hazard Index

The hazard index (HI) from THQ is expressed as the total of the hazard quotients (USEPA [21], HI value <1 considered to be safe and values >1 considered to pose a health risk.

$$HI = THQ_{Cd} + THQ_{Cu} + THQ_{Zn} + THQ_{Cr} + THQ_{Pb}$$

2-4-4 Target carcinogenic risk

Following the risk-based concentration table provided by the USEPA for the specific region, the values representing target carcinogenic risk (TR) (lifetime cancer risk) were computed for both As and Pb using the subsequent equation:

$$TR = \frac{EF \times ED \times FIR \times C \times CSFo}{WAB \times TA} \times 10^{-3}$$

Where TR denotes the target lifetime cancer risk, TA signifies the average time span for carcinogens (365 days/year×ED), and CSFo represents the oral carcinogenic slope factor derived from the USEPA's Integrated Risk Information System (2010) [22]. The CSFo values attributed to Pb, Cr and Cd were 0.0085, 0.5 and 1.5 mg/kg/day respectively. Calculated TR values below 10^{-6} are deemed negligible, those falling between 10^{-6} and 10^{-4} are generally considered acceptable, and TR values exceeding 10^{-6} are deemed unacceptable according to the USEPA's guidelines (1989) [23].

2-5 Statistical analysis

All the collected data were subject to statistical analysis like mean, minimum, maximum, and standard deviation and one-way and two-way Analysis of variance (ANOVA) using the Data Analysis package of MS Excel 2010 (Microsoft Inc.). ANOVA was carried out to compare the means of different combinations and parameters; where a significant P value was observed, differences between individual means were tested using DMRT (Duncan's Multiple Range Test) at 0.05 level of significance.

3-Results and Discussion

3-1 Bioaccumulation of heavy metals in fish tissues

The present study reports the seasonal variation of heavy metals (Cd, Cu, Zn, Cr and Pb) accumulation in various tissues (gill, muscle, kidney, liver and gonad) of *O. niloticus* collected from the two major lakes of Bengaluru namely Ulsoor and Agara lake and are presented in Tables 1 and 2. Heavy metals can cause detrimental impacts on the physiology growth and reproduction of fishes, along with compromising their biodiversity and ecosystem services. These effects encompass teratogenic, mutagenic and carcinogenic consequences, as well as issues such as kidney issues, damaging liver, cardiovascular diseases, and potential mortality [24]. Industrialisation and urbanisation are two major sources of heavy metal contamination in most of the lakes that affect aquatic organisms' storage, water balance and biotic diversity [6, 7, 25]. The results revealed that the Cd was found in high concentration in the gonad (1.68±2.54 and 3.05±2.54 mg/kg in Ulsoor and Agara fish respectively), Cu in kidney and liver (83.89±74.76 mg/kg and 212.40±33.80 mg/kg in Ulsoor and Agara fish respectively), Zn in the gill (43.46±3.16 and 46.66±2.25 in Ulsoor and Agara lake respectively), Cr in kidney and muscle (20.81±35.62 and 8.12±3.64 mg/kg in Ulsoor and Agara lake respectively). Pb in the kidney of Ulsoor lake (19.39 ± 27.99 mg/kg) and gill tissue (17.58±14.84 mg/kg) of Agara lake fish samples. It is observed that most of the heavy metals registered high concentrations during the PrM season, except Cr (M season) and Pb (M and PoM seasons). In certain months, the accumulation of heavy metals (mean value) varies significantly (standard deviation). This variance may be due to varying environmental parameters such as rainfall, temperature and ritual activities.

The order of bioaccumulation of the observed heavy metals was as follows: gonad > liver > muscle > kidney > gill; liver > kidney > gills > muscle > gonad; gill > liver > kidney > muscle > gonad; kidney > gill > muscle > gonad > liver; kidney > gill > liver > muscle > gonad respectively for Cd, Cu, Zn, Cr and Pb in *O. niloticus* collected from Ulsoor lake. In Agara lake fish, gonad > liver > kidney > gill > muscle; liver > kidney > gills > muscle > gonad; gill > liver > muscle > kidney > gonad; gill > kidney > liver > muscle > gonad; liver > gill > muscle > gonad > kidney respectively. Among the various tissues, a high accumulation of metals was registered in the gill, kidney and liver, the accumulation depends on the function and age, physiology, metabolism, food habitat and properties of metals [26]. The gills are the initial contact point between waterborne metals and fish, crucial for respiration and osmoregulation [27]. Given their sensitivity, metals can be absorbed through the gill surface. The fish kidney, responsible for metal excretion, may not efficiently eliminate certain metals, resulting in accumulation and potential damage [26]. While the liver detoxifies and transforms metals like cadmium and lead, inefficient elimination may lead to metal retention and oxidative stress [28]. However, a series of metal bioaccumulations in the gill and muscle of both the lake samples were found to be the same in the order of Zn > Cu > Pb > Cr > Cd. Similarly, kidney and liver samples also showed the same trends in the order of Cu > Zn > Pb > Cr > Cd. Whereas, in the case of gonad accumulation registered in the order of Zn > Cu > Cr > Pb > Cd in Ulsoor lake samples and Zn > Cu > Pb > Cr > Cd in Agara lake gonad samples. The accumulation pattern of heavy metals in fish tissues during different seasons are as follows: Cu > Zn > Cr > Pb > Cd in PrM, Zn > Cu > Pb > Cr > Cd in M season and Cu > Zn > Pb > Cr > Cd in PoM season in Ulsoor lake. Whereas, fish samples from Agara lake showed a consistent pattern of Cu > Zn > Cr > Pb > Cd in PrM, Cu > Zn > Pb > Cr > Cd during both M and PoM seasons.

In general, Cu and Zn were high in all the organs *O. niloticus* collected from both lakes throughout the study period. The accumulation of Cu in fish samples varied from 0 to 197.48 mg/kg (in Ulsoor lake) and 0 to 231.22 mg/kg (in Agara lake) and was found to be similar in that reported in fish obtained from Kaali River and Ox-box Lake reported [5, 29]. However, it was found several times lower than the previous report on Bellandur lake [30], Hebbal lake [4] and Kuntbhyog lake [31], Mumbai harbour [32]. Whereas, the accumulation of Zn ranged from 1.30 to 48.95 and 0.38 to 49.62 mg/kg in Ulsoor and Agara lake fish respectively and the level was significantly higher than the values reported for Bellandur lake, Hebbal lake and Kuntbhyog lake [4, 30, 31, 33]. However, high levels of

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Zn were noted in the heavy metal profile of Dhanbad lake, Ox-box Lake, Kaali River, Mahanadi River, and Mumbai harbour [5, 29, 31, 32, 34, 35] was similar. The concentration of Pb varied in the range of 0 to 61.17 mg/kg in Ulsoor lake and 0 to 58.59 mg/kg in Agara lake. The results of the present study showed very close similarities with the report of Madiwala Lake, Dhanbad lake, Ox-box Lake, Kaali River [5, 29, 35, 36]. Similarly, the concentration of Pb in this study was found to be higher than in previous reports from Bellandur, Hebbal, Kuntbhyog lakes and in Mumbai harbour [4, 30–32]. On the other hand, the Pb concentration was found to be lesser than the reports from the Mahanadi River [34]. The Cr concentration registered between 0 to 78.04 mg/kg in Ulsoor lake and 0 to 24.35 mg/kg in Agara lake. The concentration of Cr in this study registered close similarity to the previous study held in Mumbai harbour [32], Madiwala lake [36] and Dhanbad lake [35].

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Table 1: Distribution of heavy metals in fish samples collected from Ulsoor lake during different seasons (2021-2023).

Metal	Tissue	Seasons			Mean	Range	P value
		PrM	M	PoM			
Cd	Gill	1.07±1.09[a]	0.12±0.15[b]	0.25±0.17 [b]	0.48	0.01 to 2.67	<0.001
	Muscle	1.29±1.61[a]	0.35±0.66[b]	0.39±0.53[b]	0.68	0.01 to 3.72	<0.001
	Kidney	0.86±0.72[a]	0.26±0.34[c]	0.54±0.48[b]	0.55	0.02 to 1.93	<0.001
	Liver	1.22±1.41[a]	0.30±0.52[b]	0.59±0.48[b]	0.70	0.02 to 3.39	<0.001
	Gonad	1.68±2.54[a]	0.24±0.43[b]	0.20±0.18[b]	0.71	0.005 to 6.00	<0.001
Cu	Gill	3.88±1.32[b]	25.14±39.10[a]	26.24±33.18[a]	18.42	0 to 84.25	0.006
	Muscle	8.70±9.73[a]	19.93±34.37[a]	19.89±21.56[a]	16.17	0 to 71.94	0.130
	Kidney	83.89±74.76[a]	28.60±31.78[b]	39.49±22.42[b]	50.66	3.49 to 197.48	<0.001
	Liver	59.82±36.63[a]	39.79±25.07[b]	58.89±46.73[a]	52.83	5.77 to 114.95	0.066
	Gonad	4.36±6.49[b]	23.60±44.01[a]	1.32±2.14[b]	9.76	0 to 90.02	0.002
Zn	Gill	43.43±3.16[a]	37.01±8.38[b]	30.50±5.52[c]	37.08	24.44 to 47.95	<0.001
	Muscle	30.57±7.88[a]	19.49.8.38[b]	16.15±3.69[b]	22.07	8.10 to 39.51	<0.001
	Kidney	41.39±7.66[a]	20.08±8.58[c]	24.58±5.39[b]	28.67	13.09 to 48.63	<0.001
	Liver	38.81±11.02[a]	34.82±2.55[a]	20.88±7.66[b]	31.50	14.66 to 48.95	<0.001
	Gonad	18.48±8.07[b]	30.86±13.10[a]	15.42±13.38[b]	21.58	1.30 to 41.14	<0.001
Cr	Gill	5.62±1.58[b]	16.24±25.37[a]	7.23±11.35[b]	9.70	0 to 54.21	0.025
	Muscle	7.80±2.58[b]	17.20±26.57	1.75±1.24[b]	8.92	0.04 to 57.68	<0.001
	Kidney	5.90±3.46[b]	20.81±35.62[a]	5.71±9.24[b]	10.81	0 to 78.04	0.009
	Liver	5.83±3.01[b]	13.21±19.76[a]	4.47±7.34[b]	7.84	0 to 46.34	0.015
	Gonad	5.06±3.44[b]	3.24±5.39[b]	0.63±0.81[b]	8.80	0 to 61.06	<0.001
Pb	Gill	4.24±2.12[b]	17.20±28.76[a]	13.06±17.49[ab]	11.50	0.75 to 61.17	0.035
	Muscle	3.76±4.29[b]	14.03±21.83[a]	12.51±15.81[a]	10.10	0.06 to 47.24	0.028
	Kidney	6.84±6.99[b]	19.39±27.99[a]	9.76±10.82[b]	12.00	0.09 to 60.50	0.019
	Liver	3.77±3.62[b]	14.21±26.11[a]	14.00±17.69[a]	10.66	0 to 54.20	0.045
	Gonad	2.09±3.57[b]	12.29±23.03[a]	9.19±10.40[ab]	7.84	0 to 47.33	0.026

Different alphabets on the same column represent statistical significance (P<0.05).

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Table 2: Distribution of heavy metals in fish samples collected from Agara lake during different seasons of (2021-2023)

Metal	Tissue	Seasons			Mean	Range	P value
		PrM	M	PoM			
Cd	Gill	0.68±0.57[ab]	1.32±2.53[a]	0.14±0.16[b]	0.71	0.01 to 5.23	0.013
	Muscle	0.60±0.67[a]	0.14±0.22[b]	0.11±0.12[b]	0.28	0.02 to 1.62	<0.001
	Kidney	2.26±2.93[a]	0.41±0.54[b]	0.25±0.12[b]	0.97	0.03 to 6.95	<0.001
	Liver	1.92±0.76[a]	0.58±0.83[b]	0.51±0.53[b]	1.00	0.06 to 3.10	<0.001
	Gonad	3.05±5.57[a]	0.22±0.37[b]	0.22±0.23[b]	1.17	0.02 to 11.46	<0.001
Cu	Gill	20.42±15.35[a]	28.59±48.15[a]	23.91±26.50[a]	24.31	0 to 100.76	0.629
	Muscle	5.22±5.58[b]	26.95±42.18[a]	21.35±26.50[a]	17.84	0 to 89.05	0.012
	Kidney	112.57±86.99[a]	105.68±65.94[a]	76.90±59.83[a]	98.41	20.68 to 198.68	0.131
	Liver	212.40±33.80[a]	150.68±67.82[b]	149.62±82.52[b]	170.90	26.75 to 231.22	<0.001
	Gonad	4.25±3.10[b]	33.87±61.16[a]	2.76±3.12[b]	13.63	0 to 125.84	0.001
Zn	Gill	46.66±2.25[a]	36.8.36±8.38[b]	32.50±4.80[c]	38.45	23.93 to 49.61	<0.001
	Muscle	31.37±7.56[a]	24.02±11.69[b]	20.15±3.94[b]	25.18	9.75 to 37.84	<0.001
	Kidney	30.41±7.56[a]	22.65±13.52[b]	16.51±2.32[c]	23.19	8.47 to 40.13	<0.001
	Liver	43.31±4.17[a]	27.71±12.76[b]	23.42±6.28[b]	31.48	12.59 to 49.62	<0.001
	Gonad	13.33±9.06[b]	33.18±11.74[a]	16.64±8.25[b]	21.05	0.38 to 40.24	<0.001
Cr	Gill	5.97±2.80[a]	3.24±5.39[a]	6.97±11.36[a]	5.39	0.06 to 24.35	0.135
	Muscle	8.12±3.64[a]	3.52±5.82[b]	1.59±1.08[b]	4.41	0 to 12.75	<0.001
	Kidney	6.37±2.62[a]	2.61±5.09[b]	5.58±7.01[a]	4.85	0 to 15.86	0.022
	Liver	7.49±3.59[a]	3.56±5.66[b]	2.21±3.35[b]	4.42	0 to 12.27	<0.001
	Gonad	4.82±3.81[a]	2.72±5.30[b]	0.75±0.75[c]	2.76	0 to 10.80	<0.001
Pb	Gill	4.35±4.39[b]	16.57±27.32[a]	17.58±14.84[a]	12.84	0.06 to 58.59	0.009
	Muscle	3.52±5.51[b]	14.92±26.03[a]	15.27±14.32[a]	11.24	0 to 54.87	0.015
	Kidney	2.20±3.05[b]	12.88±23.39[a]	16.84±16.14[a]	10.64	0 to 48.48	0.003
	Liver	7.79±8.35[a]	15.68±24.97[a]	17.10±14.59[a]	13.52	0.20 to 53.44	0.088
	Gonad	3.84±7.42[b]	12.83±23.58[a]	16.10±16.98[a]	10.92	0 to 48.86	0.020

Different alphabets on the same column represent statistical significance (P<0.05).

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On the other hand, the observed concentration of the study was found to be several times lower than the studies that happened in Ox-box Lake [5], Kaali River [29].

However, Cd registered a low concentration compared to all other metals, the range was between 0.05 to 6.00 mg/kg in Ulsoor lake and 0.01 to 11.46 mg/kg in Agara lake. The observed Cd concertation in the current study showed almost similar to that reported in Madiwala lake and Dhanbad lake [35, 36]. Similarly, an elevated level of Cd was reported in the Kaali River by [29], Ox-box Lake by [5] and in Mahanadi River [34], a relatively lower level of Cd was found in Kuntbhyog lake [31].

Bioaccumulation of these heavy metals, either essential or non-essential, like Cd, Pb Cr and Cu in fishes and other aquatic organisms poses threats causing toxicity through detrimental impacts on their reproductive capabilities, development and overall well-being by disrupting essential physiological functions like enzyme activity, ion regulation, diminished feeding, respiratory distress, and even fatality [35, 37–40]. Within aquatic ecosystems, these can interfere with nutrient cycling, modify community compositions, and impact the overall health of the ecosystem [39, 41]. Overall, the accumulation of Cd and Zn in both lake fish samples was found to be high in PrM, decrease in the water levels and high evaporation rate may lead to sedimentation of metals leading to increased concentration [42]. Cu levels are found higher during PoM and M seasons in Ulsoor and Agara lakes, respectively. This is due to increased runoff and erosion of contaminated soil and sediment, which results in bioaccumulation in fish [43] (Udayakumar et al. 2014). High levels of Cr and Pb were found in M season in Ulsoor lake fishes, and in Agara lake Cr levels were high in PrM and Pb in PoM season. Resuspension and contaminated sediment along with the toxic elements entered through rainwater lead to the accumulation of heavy metals in fish tissues [35] (Pal and Maiti 2018).

3-2 Correlation analysis:

Pearson's correlation of heavy metals in different tissues revealed a significant positive relationship among the tissues (Table 3). Cd showed a positive correlation between all the tissues of Ulsoor lake samples except the gonad and kidney; whereas, in Agara lake, kidney and muscle, liver and muscle, liver and kidney and gonad with muscle and kidney exhibited a significant strong positive relation ($p<0.01$). On the other hand, muscle and gill of both lake samples and gonad with gill and muscle showed a strong correlation for Cu. Similarly, Zn also did not show the relation between the tissues except muscle and gill of both lakes and liver with gill and muscle of Agara lake tissues. However, the relation of Cr in all the tissues of Ulsoor lake samples showed a significant positive relationship ($p<0.01$). In Agara lake only muscle and gill, liver and muscle, gonad and muscle and gonad and liver exhibited a strong relationship. Surprisingly all the tissue samples showed a significant strong correlation ($p<0.01$) between the tissues for Pb in the tissue samples of both the lakes.

The overall results revealed that the bioaccumulation of heavy metals such Cd, Cu, Cr and Pb in various tissues of *O. nilotics* was higher than the permissible limit set by various agencies. However, the accumulation level of Zn is below the permissible limit, except FEPA (2003) [47] (Table 4). Moreover, heavy metals present in fish pose potential risks to human health throughout the food chain, given that fish serve as a crucial source of protein and nutrients for many individuals. Risks associated with human exposure include neurotoxicity, chronic toxicity, endocrine disruption and immunotoxicity [49]. Since muscle is the only edible part in the selected fish species this study analysed the human risk assessment for muscle tissue alone.

Table 3: Correlation of heavy metals between the tissues compared in all the seasons (2021-2023).

		Ulsoor lake					Agara Lake				
		Gill	**Muscle**	**Kidney**	**Liver**	**Gonad**	**Gill**	**Muscle**	**Kidney**	**Liver**	**Gonad**
Cd	Gill	1					1				
	Muscle	0.930**	1				0.408**	1			
	Kidney	0.840**	0.934**	1			0.319**	0.985**	1		
	Liver	0.934**	0.974**	0.929**	1		0.524**	0.876**	0.808**	1	
	Gonad	0.953**	0.917**	0.766**	0.928**	1	0.234*	0.962**	0.983**	0.729**	1
Cu	Gill	1					1				
	Muscle	0.860**	1				0.940**	1			
	Kidney	0.090	0.035	1			0.086	0.119	1		
	Liver	0.281*	0.154	0.294*	1		-0.098	-0.196	0.505**	1	
	Gonad	0.671**	0.721**	0.271*	0.166	1	0.809**	0.814**	0.114	-0.343**	1
Zn	Gill	1					1				
	Muscle	0.859**	1				0.822**	1			
	Kidney	0.450**	0.731**	1			0.622**	0.645**	1		
	Liver	0.693**	0.753**	0.597**	1		0.828**	0.807**	0.736**	1	
	Gonad	0.036	-0.017	-0.257*	-0.025	1	-0.145	0.117	0.237*	0.0836	1
Cr	Gill	1					1				
	Muscle	0.909**	1				0.394**	1			
	Kidney	0.980**	0.963**	1			0.871**	0.567**	1		
	Liver	0.981**	0.936**	0.979**	1		0.661**	0.923**	0.748**	1	
	Gonad	0.850**	0.953**	0.917**	0.878**	1	0.348**	0.920**	0.530**	0.847**	1
Pb	Gill	1					1				
	Muscle	0.980**	1				0.997**	1			
	Kidney	0.910**	0.899**	1			0.990**	0.986**	1		
	Liver	0.987**	0.981**	0.903**	1		0.972**	0.975**	0.960**	1	
	Gonad	0.978**	0.947**	0.945**	0.975**	1	0.983**	0.986**	0.986**	0.979**	1

**Correlation is significant at the 0.01 level (2-tailed). *Correlation is significant at the 0.05 level (2-tailed).

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Table 4: Permissible limit preferred by various organisations

Agencies	Cd	Cu	Zn	Cr	Pb
FAO 1983 [44]	2	-	50	1	1 to 6
WHO 2006 [45]	-	3	50	0.15	2
EC 2001 [46]	-	1	-	1	-
FEPA 2003 [47]	-	1.3	15	0.15	2
MPEDA 1987 [48]	-	10	50	-	5

3-3 Estimated daily intake (EDI)

The Estimated daily intakes for the analysed metals in Ulsoor and Agara lake fish for both target groups (male and female) are depicted in Table 5. The findings indicate that Cd levels during PrM in Ulsoor lake fish, as well as Cu and Cr from both lakes across all seasons, and Pb in M and PoM from both lakes, exceeded the permissible limits set by various organizations (Table 3). The comparison of mean Estimated Daily Intake (EDI) values with Tolerable Daily Intake (TDI) values revealed that the calculated values suggest potential health risks associated with the consumption of the specific fish in the study area. On the country, Pal and Maiti. [35] reported comparatively high values of EDI in Dhanbad lake India. On the other hand, Pradip and Malik. [29] reported lower values of EDI from the Ganga River in India.

3-4 Target hazard quotient (THQ) and Hazardous index (HI)

THQ values for selected heavy metals in fish samples from Ulsoor and Agara lakes are presented in Table 6. The findings of the current study indicate that THQ values for Cr in all fish species were below unity for both male and female target groups. In contrast, Cd, Cu, Zn, and Pb exhibited the highest THQ values across all fish species from both lakes. According to the THQ, the fish species are deemed unsafe, posing potential risks associated with Cd, Cu, Zn, and Pb (THQ > 1). The Hazard Index (HI) values for fish species exceeded unity, as detailed in Table 6. According to the HI values, the consumption of tilapia fish from Ulsoor and Agara lakes may pose potential health risks. The prolonged exposure to these metals through the consumption of the selected fish species could lead to chronic health effects. Similarly, Pal and Maiti [35] reported THQ>1 for Pb was reported in the Dhanbad lake, India. Kumari et al (2018) reported a high value of (THQ>1) for Cr and Pb in edible fish of Jamshedpur Urban Agglomeration India.

3-5 Target carcinogenic risk (TR)

Target Risk (TR) assessment was conducted specifically for Cd, Cr and Pb, as these elements are categorized as carcinogenic. For carcinogens, TR values should be equal to or less than 10^{-6}, although they may extend up to 10^{-4}. In this study, the calculated TR values fell within the acceptable range for Pb but exceeded this range for both Cd and Cr. Since the Cd is an endocrine disruptor, it can affect progesterone, testosterone, estrogenic, and ovarian steroidogenic pathways, thus boosting the risk of breast cancer and ovarian cancer [50, 51] Consumption of high doses of Cr in the bloodstream causes blood cell damage, leading to lung cancer, and also damages the normal functionality of the kidneys and liver [52, 53]. Persistent entry of Pb in humans is carcinogenic and causes kidney cancer, brain cancer and stomach cancer by damaging the DNA and cell nature [54]. This suggests that the consumption of these freshwater fish species, with elevated Cd and Cr levels, could pose a considerable hazard risk to the human body. The chosen fish species (*O. niloticus*) in the examined urban lakes hold significant economic and dietary importance in the region. Therefore, evaluating the health risks associated with consuming these fish is necessary. It is essential to monitor and compare EDI and TDI to assess and manage potential health risks associated with various substances. The calculated EDI and TDI values showed potential health risks to both the targeted groups. THQ and HI for Cu, Cd, Zn and Pb exhibited THQ >1, indicating the potential health risks that can be caused by these metals, similarly, HI also exceeded the allowable limit and stands to be posing health hazardous to the community which includes these fishes in their diet.

Table 5: The estimated daily intake (EDI) and the tolerable daily intake (TDI) of heavy metals in the fish collected from Ulsoor and Agara lakes of Bengaluru.

		Ulsoor			Agara			
Heavy metals	**Population**	**PrM**	**M**	**PoM**	**PrM**	**M**	**PoM**	**TDI mg/day**
Cd	Men	0.583	0.158	0.176	0.271	0.063	0.049	0.5
	Women	0.689	0.187	0.208	0.320	0.074	0.058	FAO (1983)
Cu	Men	3.936	9.017	8.999	2.361	12.193	9.660	0.9
	Women	4.652	10.657	10.635	2.791	14.410	11.416	NIH
Zn	Men	13.831	8.818	7.307	14.193	10.868	9.117	60.0
	Women	16.346	10.421	8.635	16.774	12.844	10.774	WHO (1996)
Cr	Men	3.529	7.782	0.791	3.673	1.592	0.719	0.20
	Women	4.170	9.197	0.935	4.341	1.882	0.850	RDA (1989)
Pb	Men	1.701	6.348	5.660	1.592	6.750	6.909	3.0
	Women	2.010	7.502	6.689	1.882	7.978	8.165	JECFA (2000)

Table 6: Risk values (THQ, HI, and TR) of each metal contaminant in muscle tissue of fish collected from Ulsoor and Agara lake of Bengaluru, Karnataka, India.

Risk values	**HM**	**Population**	**Ulsoor**			**Agara**		
			PrM	**M**	**PoM**	**PrM**	**M**	**PoM**
THQ	**Cd**	**Men**	0.583	0.158	0.176	0.271	0.063	0.049
		Women	0.689	0.187	0.208	0.32	0.074	0.058
	Cu	**Men**	0.098	0.225	0.224	0.059	0.304	0.241
		Women	0.116	0.266	0.265	0.069	0.36	0.285
	Zn	**Men**	0.461	0.293	0.243	0.473	0.362	0.303
		Women	0.544	0.347	0.287	0.559	0.428	0.359
	Cr	**Men**	0.002	0.005	0.0006	0.002	0.001	0.0005
		Women	0.003	0.007	0.0007	0.003	0.001	0.0006
	Pb	**Men**	0.425	1.587	1.415	0.398	1.687	1.727
		Women	0.502	1.875	1.672	0.47	1.994	2.041
HI	**HI**	**Men**	1.571	2.27	2.06	1.204	2.419	2.323
		Women	1.856	2.683	2.435	1.423	2.859	2.745
TR	**Cd**	**Men**	0.008	0.002	0.002	0.004	0.0009	0.0007
		Women	0.01	0.002	0.003	0.004	0.001	0.0008
	Cr	**Men**	0.001	0.003	0.0003	0.001	0.0007	0.0003
		Women	0.002	0.004	0.0004	0.002	0.0009	0.0004
	Pb	**Men**	1.45E-05	5.40E-05	4.81E-05	1.35E-05	5.74E-05	5.87E-05
		Women	1.71E-05	6.38E-05	5.69E-05	1.60E-05	6.78E-05	6.94E-05

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The TR levels in Cd and Cr exhibited the unacceptable range in the case of men and women. Therefore, the potential health risks associated with metal exposure through fish consumption in an economically significant urban lake cannot be ignored by local populations.

It's important to be aware that consuming fish with high levels of heavy metals can lead to serious health issues. These can affect the nervous system, kidneys, and cardiovascular system. Pregnant women, children, and people with weakened immune systems are particularly at risk [55]. Consistently ingesting heavy metals can cause developmental delays, cognitive impairment, an increased risk of cancer and pose detrimental effects on the kidneys, bones, blood cells, nervous system, and respiratory system [55, 56]. Wilson's disease, a genetic disorder impacting copper metabolism [57], DNA damage and gastrointestinal problems [38]. and these effects may vary depending on the form of the metals and its concentration present in the fish [58]. It's critical to control and monitor the levels of heavy metals in fish and increase public awareness of the risks of heavy metal ingestion to prevent these health problems and guarantee the safety of fish consumers [35, 37].

4-Conclusion

In conclusion, the current research has revealed the heavy metal accumulation in various tissues of *O. niloticus* collected from Ulsoor and Agara lakes of Bengaluru. Bioaccumulation of heavy metals in fish tissues is generally higher during the pre-monsoon season than in the post-monsoon and monsoon periods, demanding increased attention due to elevated pollution and decreased water levels. The accumulation of Cd, Cu, Zn, Cr, and Pb exceeded allowable limits for fish samples as set by various organizations. The potential human health risk assessment such as EDI, TDI, THQ and TR values exceeded the threshold, suggesting a potential risk for overall consumer health including cancer. Thus, this study suggests that consuming tilapia fish from Ulsoor and Agara lakes in Bengaluru may not be safe for human health. Continuous monitoring of water and biota is recommended to control further pollution in the selected lakes.

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6-Competing Interests

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7-Authorship Contribution Statement

All authors contributed to the study's conception and design. Material preparation, data collection and analysis were performed by Ajjigudde Shreenivasa Shashank and Krishnakumar Velayudhannair. The first draft of the manuscript was written by Ajjigudde Shreenivasa Shashank and all authors commented on previous versions of the manuscript. All authors read and approved the final manuscript.

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