Bima Journal of Science and Technology, Vol. 8(2) June, 2024 ISSN: 2536-6041

DOI: 10.56892/bima.v8i2.690

Assessment of Heavy Metal Concentration in Commercially Important Fishes from Cross River System Itu, Nigeria

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ABSTRACT

The Cross River System provides critical economic and ecological services to people around its environment. However, human activities along the river make it a potential dumpsite for industrial, domestic, urban, and agricultural wastes. This research aimed to investigate the concentration of heavy metals in commercially important fish species from the Cross River System, Itu, Nigeria. Samples were collected monthly from June to September 2021. The concentration of heavy metals were analyzed using Atomic Absorption Spectroscopy (AAS) and the following order of increment was recorded Mn (29.467±0.04 mg/kg) > Cu (24.897±0.02 mg/kg) > Zn (18.190±0.00 mg/kg) > Fe (8.923±0.01 mg/kg) > Ni (0.065±0.00 mg/kg) > Cd (0.033±0.00 mg/kg) > Pb (0.030±0.00 mg/kg) > Cr (0.018±0.00 mg/kg). Similarly, the following order in decreased was also recorded Cr (0.011±0.00 mg/kg) < Pb (0.015±0.00 mg/kg) < Ni (0.015±0.01 mg/kg) < Cd (0.021±0.00 mg/kg) < Fe (7.62±0.52 mg/kg) < Zn (14.813±0.91 mg/kg) < Cu (20.133±0.03 mg/kg) < Mn (23.042±0.01 mg/kg) respectively. Monthly variation in heavy metals was also recorded. *Chrysichthys Nigrodigitatus* from Mbiabo Edere had the highest concentration of Zn, and Mn (18.00±1.00 mg/kg, 29.40±0.00 mg/kg), *Schilbe mystus* from Ikot Otu had the highest concentration of Cd (0.06±0.47 mg/kg). *Mormyrus rume* from Itu Head Bridge and Ikot Iwatt had the highest concentration of Ni (19.19±0.33 mg/kg) and Pb (0.431±0.92 mg/kg), similarly *Synodontis Schall* from Ikot Otu and Ikot Iwatt had the highest concentration of Cu (21.62±0.03 mg/kg) and Cd (0.09±0.51 mg/kg) respectively. The results of the concentration of heavy metal in this study were below the permissible limits recommended by the National Environment Standards and Regulations Enforcement Agency, the European Commission, and the Joint Expert Committee of the Food and Agricultural Organization/World Health Organization FAO/WHO for food. This implies that fish from the cross-river system contained low concentrations of heavy metals. However, there is a need for research to assess the health impacts of these heavy metals in fish and the potential for transfer to humans. Also, there is a need for routine heavy metal monitoring programmes in fish along the Cross River System to help in the execution of heavy metals regulatory standards by the government and relevant environmental health management agencies.

Keywords: Heavy metal, Toxicity, Fish, Cross River System.

INTRODUCTION

The persistent discharge of toxic chemical compounds by man into the aquatic environments incites the need to assess contaminant levels in aquatic ecosystems and organisms. However, the major groups of environmental contaminants in the aquatic ecosystem include persistent organic pollutants, radioactive waste, extractive residue, pathogens, litter, debris, and heavy metals with severe toxicological implications. Increased ecological and public health consequences of heavy metals toxicity, nonbiodegradability, prolonged biological half-life, and bio-accumulation in animal and human organs/tissues have received great attention in recent times (Asraf *et al.,* 2012; Ye *et al.,* 2012; Jacob *et al.,* 2024b). Aquatic organisms including fish are highly threatened by heavy metal pollutants due to their toxicological implications (Islam *et al.,* 2020; Biswas *et al.,* 2021).

Heavy metals are naturally occurring elements on the earth's crust with high atomic weight and about 5 times the density greater than water (Mahmuda *et al.,* 2020). Unfortunately, some natural phenomenon and anthropogenic activities significantly modify their geological cycles and biochemical balance, thereby enhancing their entry into the food web and human body (Mahmuda *et al.,* 2020).

Some heavy metals perform essential biological functions such as metabolism and constitute a major part of several key enzymes in living organisms (FAO, 1996). Although heavy metals are classified as essential (Zn, Cu, Fe, Mn, etc.) and non-essential or toxic heavy metals (As, Cd, Pb, Hg, etc), excessive heavy metals in the body can result in mutagenesis, carcinogenesis, tetratogenesis, deformation, liver damage, kidney dysfunction, cardiovascular abnormalities, impaired metabolism, and death (Tchounwou *et al.,* 2012; Suchana *et al.,* 2021; Al-Busaidi, *et al.,* 2011; Hina, *et al.,* 2011; El-Moselhy *et al.,* 2014).

Fish is a balanced food item consumed throughout the world due to its role in providing protein, energy, and essential minerals to the body (Pieniak, 2010; Jacob *et al.,* 2023). However, the accumulation of heavy metals in fish can be transferred to humans by feeding on contaminated food and water (trophic transfer (Zhuang *et al.,* 2009). Ingestion of contaminated aquatic food, suspended particles in water, and ion exchange between fish gills and water are the major routes of heavy metals accumulating in fish bodies (Mansour and Sidky, 2002). Therefore, Fish serves as an indicator organism to estimate heavy metal contaminants in freshwater ecosystems and associated human health risks (Pan *et al.,* 2012; Jacob *et al.,* 2023).

In Nigeria, fish from streams and rivers constitute a substantial portion of accessible protein (Jacob *et al.,* 2023). The Cross River System is an important river network in South-South Nigeria flowing in about three states from its source. The river receives large amounts of waste discharge ranging from industrial, and domestic effluents, to urban and agricultural wastewater which exposes organisms in this river to severe contaminants burden. Several studies have reported unprecedented levels of toxic heavy metals in aquatic environments and aquatic organisms in recent times (Tchounwou *et al.,* 2012; El-Moselhy *et al.,* 2014; Mahmuda 2020; Ali, *et al.,* 2020). Similarly, Odoemelam *et al.,* (2020), reported the concentration of heavy metals in water and sediments samples from the lower cross system. Unfortunately, there is little or no information on the concentration of heavy metals in commercially important fish species in the cross river system. Similarly, organisms in the cross river system are continuously exposed to heavy metal pollutants due to anthropogenic activities close to the river.

Most toxicity research on heavy metals in the cross river system has been focused on water and sediment pollution profiles. Information on the toxicity matrix in commercially important fish in this river is sparse. Assessment of heavy metals in fish organs and muscles is critical to provide information on health risks to humans who feed on these fish species. To protect the general public from

DOI: 10.56892/bima.v8i2.690

exposure to heavy metals through the consumption of contaminated fish food, there is a need to investigate the contaminant levels of heavy metals in commercially important fish in the cross-river system. Therefore, this study was designed to determine heavy metals (Zn, Cd, Cr, Pb, Ni, Mn; Cu, and Fe) concentration in commercially important fish species from the Cross River System to provide critical public health awareness information on the effect of heavy metal pollution.

MATERIALS AND METHODS

Description of the Study Area

Cross River system is located in the South-South region of Nigeria on longitude 7° 30' and 9°30' North and latitude 4°30' to 7°15' East (Fig. 1). The climatic condition of the area is marked by North-South movement of zones of discontinuity between maritime (Atlantic) and dry continental (Sahara) air masses. The movement of these air masses creates distinct dry and wet seasons in the area. However, the change from dry to wet season happens in a distinct gradual manner, while the change from wet to dry occurs rapidly. The maximum temperature and annual rainfall in this area is between 26-28°C and 362.5mm respectively (Opeh and Udo, 2017).

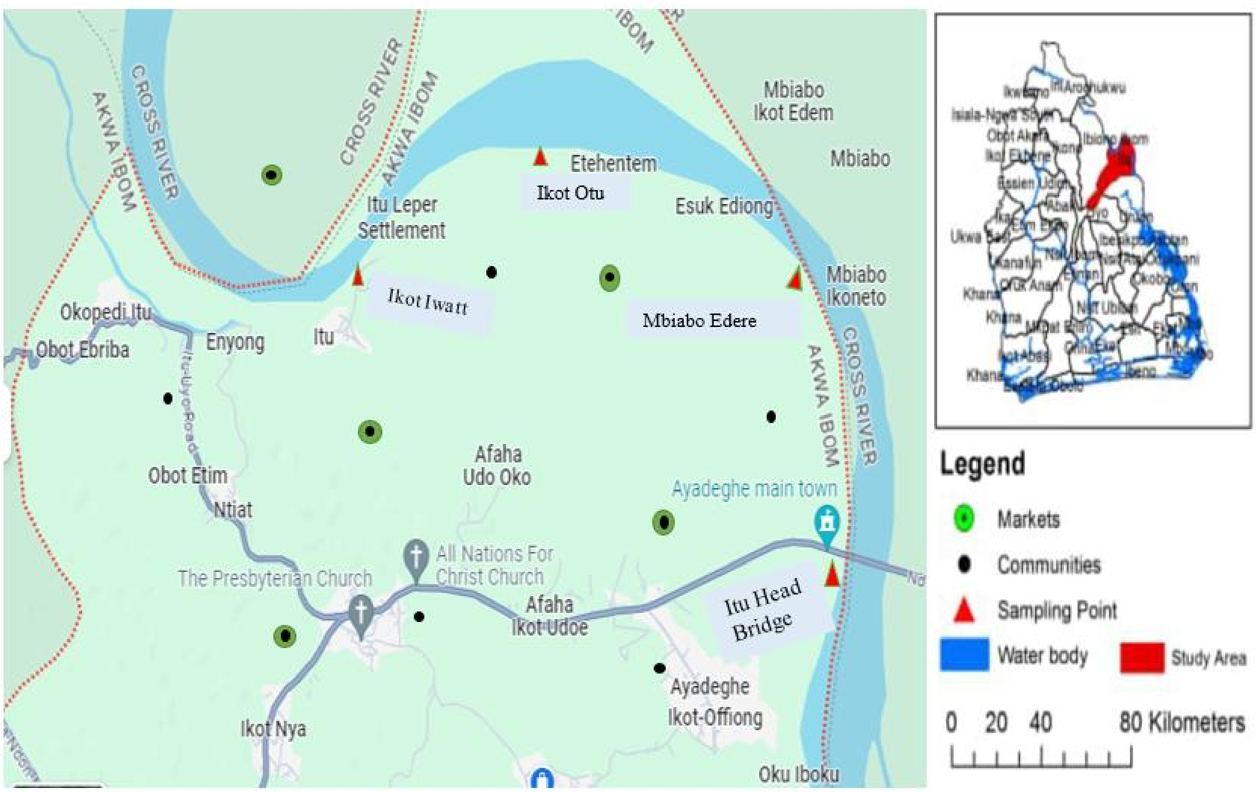

Figure 1: Map of Cross River System showing sampling stations

The Cross River System gets its source from the Cameroon Republic and flows through Ikom and Obubra in Cross River State, Afikpo in Ebonyi State, Okopedi in Itu, Oron, Ibaka in Mbo in Akwa Ibom State where it discharges into the Atlantic Ocean, covering about 352 kilometers. The densely populated rural dwellers in these areas predominantly depend on the river for their drinking water supply and fisheries resources. The River also provides

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places of abode and recreation, means of livelihood and transport, and a discharge route for residential, agricultural, and industrial sewage. It also provides critical ecological functions such as water purification and temperature regulation to the surrounding ecosystem. In Itu River Head Bridge, however, agricultural activities, transportation, and trading constitute major viable economic activities that sustain the dense human population in this area, these activities make the river a potential dumpsite for waste generated in this area.

Sample collection

Four sampling stations namely Ikot Iwatt, Ikot Otu, Mbiabo Edere, and Itu Head Bridge were chosen for this study. Also, four commercially important fish species namely *Chrysichthys nigrodigitatus*, *Synodontis schall*, *Mormyrus rume,* and *Schilbe mystus* (Fig. 2) were randomly collected from landings of artisanal fishermen in each of the sampling stations. A total of 250 samples were collected from June – September 2021.

Figure 2: Samples of Fish Species collected for this study (a: *Synodontis schall*, b: *Mormyrus rume*, c: *Chrysichthys nigrodigitatus*, d: *Shilbe mystus*)

No sex difference was considered. During collection, samples were packed in nylon bags, labeled, put in an ice chest, and transported to the Department of Fisheries and Aquatic Environmental Management Laboratory, University of Uyo for sorting and identification (Adesulu and Sydenham, 2007). Laboratory protocols were properly observed to avoid the introduction of metals from laboratory equipment into the fish muscle during sample preparation. In the laboratory, all the fish were thawed and dissected carefully. Muscular tissues on the dorsal surface of each fish were collected and preserved in a specimen bottle until preparation.

Sample Preparation and Thermal Digestion

About 100 g of fish muscle excised from individuals of each species were dried at 105^0 for 24 h in a hot-air oven. The sample was placed into the oven in clean white paper.

Then, the dried samples were ground into a fine powder. 1 g of each sample was digested at 130ºC in an electro-thermal heater for 30 minutes thereafter, treated with HCl and HNO_3, (3:1 v/v).To make the volume up to 100 ml, 20 ml of distilled water was added to the digested sample and transferred into a clean volumetric flask after cooling. Filtration was done using the Whatman Filter paper. Lastly, the filtrated solutions were kept in sealed and labeled plastic bottles until when it was used.

Blank Solution Preparation

Briefly, a blank solution containing all the similar digestion inputs except the sample was prepared by following standard procedure. The blank solution was used to identify whether impurities or contaminants from other chemicals were based on the result (Mortuza, and Misned, 2015). The blank value found through the analysis by atomic absorption spectrophotometer (AAS) was subtracted from each of the sample values to find the original value.

Sample analysis, method validation

Heavy metal concentrations were determined using a flame atomic absorption spectrophotometer, AAS (UNICAM939/59). Acetylene gas and air were used as fuel and oxidizer, respectively. The concentrations of heavy metals were determined with the support of calibration curves. Calibrations were done using standard solutions following the manufacturer's instructions.

After the establishment of the calibration curves, the limit of detection (LOD) was measured for the heavy metals of interest. The results (LOD) were measured using the equation adopted by (Sahnquillo *et al.,* 2003). Briefly, 3.3 * (S.Dblank/m), where SD stands for the standard deviation of blanks and m is the sensitivity of the calibration curve.

All chemicals and reagents were of analytical reagent-grade quality. Before use, all glass and plastic ware were soaked in 14% HNO3 for 24 hours. The washing was done with distilled water.

Data Analysis

Data were subjected to One-way analysis of variance (ANOVA) using SPSS (IBM version, 25). Results were considered significant at ($P \leq 0.05$) level.

RESULTS

Variations in Heavy Metals Concentration

The mean concentration of heavy metals recorded in the four commercially important fish species for each sampling month is presented in Table 1. The highest concentrations of each of the respective heavy metals were Pb (0.030±0.00 mg/kg), Cu (24.897±0.02 mg/kg), Cd (0.033±0.00 mg/kg), Cr (0.018±0.00 mg/kg), Zn (18.190±0.01 mg/kg), Ni (0.065±0.00 mg/kg), Mn (29.467±0.04 mg/kg) and Fe (8.923±0.01 mg/kg). While the lowest concentrations were Pb (0.015±0.00 mg/kg), Cu (20.133±0.03 mg/kg), Cd (0.021±0.00 mg/kg), Cr (0.011±0.00 mg/kg), Zn (14.813±0.91 mg/kg), Ni (0.015±0.01 mg/kg), Mn (23.042±0.01 mg/kg), and Fe (7.62±0.52 mg/kg) respectively.

There was a significant difference ($P \leq 0.05$) in heavy metal concentrations among the different sampling months. However, Mn had the highest heavy metal concentration (29.467±0.04 mg/Kg) while lead (Pb) had the lowest heavy metal concentration (0.015±0.00 mg/kg). For individual heavy metals, there was a significant difference ($P \leq 0.05$) across the period of the study period. However, there were no significant differences ($P \leq 0.05$) in the mean concentration of chromium during the study period.

Table 1: Monthly variations in heavy metals concentration (mg/kg) in fish from the Cross River system

Months	Species	Pb	Cu	Cd	Cr	Zn	Ni	Mn	Fe
June	*Synodontis schall*	0.025±0.00^{b}	24.807±0.06^{b}	0.026±0.00^{b}	0.016±0.00^{e}	18.080±0.00^{a}	0.065±0.00^{d}	29.420±0.36^{a}	8.870±0.24^{e}
	Mormyrus rume	0.022±0.00^{c}	24.787±0.02^{b}	0.026±0.00^{b}	0.011±0.00^{e}	16.217±0.2^{bc}	0.030±0.01^{de}	29.467±0.04^{a}	8.737±0.12^{f}
	Schilbe mystus	0.022±0.00^{c}	23.513±0.71^{d}	0.022±0.00^{b}	0.015±0.00^{e}	15.523±0.12^{e}	0.053±0.01^{de}	28.27±0.10^{e}	8.683±0.02^{fg}
	Chrysichthys nigrodigitatus	0.022±0.00^{c}	20.160±0.01^{j}	0.023±0.00^{b}	0.015±0.00^{e}	16.220±0.82^{bc}	0.042±0.01^{de}	28.900±0.03^{b}	7.620±0.52^{h}
July	*Synodontis schall*	0.022±0.00^{c}	23.623±0.02^{c}	0.026±0.00^{a}	0.015±0.00^{e}	18.017±0.02^{a}	0.055±0.00^{de}	28.673±0.02^{c}	8.923±0.01^{e}
	Mormyrus rume	0.024±0.00^{b}	23.417±0.51^{e}	0.027±0.00^{b}	0.018±0.00^{e}	16.283±0.86^{b}	0.055±0.00^{e}	28.423±0.02^{d}	8.727±0.22^{f}
	Schilbe mystus	0.027±0.00 ^{a}	22.423±0.01^{g}	0.024±0.00^{b}	0.013±0.00^{e}	15.413±0.01^{f}	0.015±0.00^{de}	27.153±0.03^{g}	8.657±0.53^{g}
	Chrysichthys nigrodigitatus	0.019±0.00^{e}	20.133±0.03^{j}	0.021±0.00^{b}	0.016±0.00^{e}	15.930±0.04^{d}	0.047±0.00^{e}	27.533±0.01^{f}	7.650±0.03^{h}
August	*Synodontis schall*	0.019±0.00^{d}	22.672±0.81^{f}	0.024±0.00^{b}	0.013±0.00^{e}	18.003±0.10^{a}	0.050±0.00^{de}	28.627±0.00^{c}	8.907±0.61^{e}
	Mormyrus rume	0.017±0.00^{fg}	22.351±0.00^{h}	0.020±0.00^{b}	0.013±0.00^{e}	16.143±0.21^{c}	0.015±0.00^{e}	28.407±0.41^{d}	8.740±0.52^{f}
	Schilbe mystus	0.015±0.00^{g}	22.427±0.92^{g}	0.023±0.00^{b}	0.014±0.00^{e}	14.937±0.82^{g}	0.037±0.00^{de}	27.133±0.01^{g}	8.653±0.61^{g}
	Chrysichthys nigrodigitatus	0.019±0.00^{d}	22.073±0.23^{i}	0.020±0.00^{b}	0.012±0.00^{e}	14.813±0.91^{h}	0.019±0.01^{e}	27.510±0.01^{f}	7.647±0.07^{h}
September	*Synodontis schall*	0.029±0.00 ^{a}	24.897±0.02^{a}	0.026±0.00^{b}	0.015±0.00^{c}	18.190±0.01^{a}	0.019±0.00^{i}	26.653±0.16^{h}	8.910±0.01^{a}
	Mormyrus rume	0.030±0.00 ^{a}	24.823±0.72^{b}	0.025±0.00^{b}	0.014±0.00^{c}	16.223±0.02^{c}	0.057±0.14^{i}	27.322±0.79^{h}	8.757±0.54^{b}
	Schilbe mystus	0.025±0.00^{b}	23.510±0.01^{d}	0.022±0.00^{b}	0.012±0.00^{c}	15.623±0.63^{b}	0.015±0.11^{i}	27.415±0.61^{h}	8.687±0.02^{c}
	Chrysichthys nigrodigitatus	0.023±0.00^{c}	20.173±0.02^{j}	0.033±0.00^{b}	0.013±0.00^{c}	16.227±0.01^{b}	0.015±0.00^{i}	23.042±0.01^{h}	7.650±0.03^{d}
P-value		0.0012*	0.0001*	0.4782	0.0001*	0.0001*	0.0005*	0.0004*	0.0003*

- Indicate significant differences in heavy metal concentration

DOI: 10.56892/bima.v8i2.690

Table 2: National and International Permissible Limit (mg/kg) of metal concentration in fish

Heavy metals	MFA	FAO	WHO/FEPA	WHO	USFDA	USEPA
Zn	100	30.00	30.00	-	-	0.0766
Ni	-	-	0.10	-	-	
Pb	2.0	0.50	2.00	-	0.5	0.0058
Fe	-	-	0.50	1-3	-	0.1
Cd	1.0	0.50	0.50	-	0.01 – 0.21	0.008
Cr	-	0.15 – 1.0	-	0.05 – 0.15	-	0.05
Cu		0.05	< 1.0			1.0
Mn		0.1 - 0.5	-	2.5	-	-

Concentration Heavy Metal in the Different Fish Species

The concentrations of heavy metals in *Chrysichthys nigrodigitatus* for the different sampling stations are presented in Table 3. The highest concentration of respective heavy metals was as follows: Pb (0.02±0.79 mg/kg), Cu (24.08±0.64 mg/kg), Cd (0.23±0.41 mg/kg), Cr (0.05±0.34 mg/kg), Zn (18.00±1.00 mg/kg), Ni (1.19±0.51 mg/kg) Mn (29.40±0.01 mg/kg) and Fe (6.90±0.66 mg/kg). While the lowest concentrations of heavy metals were Pb (0.01±0.29 mg/Kg), Cu (21.89±0.06 mg/Kg), Cd (0.12±0.17 mg/Kg), Cr (0.01±0.32 mg/Kg), Zn (10.03±0.03 mg/Kg), Ni (0.05±0.51 mg/Kg), Mn (26.50±0.04 mg/Kg), and Fe (4.87±1.02 mg/Kg).

The concentration of heavy metal in *Schilbe mystus* for the different sampling stations is presented in Table 4. The highest concentrations of heavy metals were: Pb (0.72±0.79 mg/kg), Cu (22.67±0.01 mg/kg), Cd (0.06±0.47 mg/kg), Cr (0.08±0.53 mg/kg), Zn (19.23±0.03 mg/kg), Ni (0.08±0.15 mg/kg) Mn (23.44±0.21 mg/kg) and Fe (5.87±0.12 mg/kg). While the lowest were: Pb (0.01±0.53 mg/Kg), Cu (17.62±0.03 mg/Kg), Cd (0.02±0.31 mg/Kg), Cr (0.01±0.32 mg/Kg), Zn (10.00±0.91 mg/Kg), Ni (0.03±0.25 mg/Kg), Mn (17.67±0.01 mg/kg), and Fe (3.60±0.14 mg/kg).

The concentration of heavy metal in *Synodontis schall* from the different sampling stations is presented in Table 5. The highest concentrations of heavy metals were: Pb (0.83±0.53 mg/kg), Cu (21.62±0.03 mg/kg), Cd (0.09±0.51 mg/kg), Cr (0.07±0.83 mg/kg), Zn (20.10±0.01 mg/kg), Ni (0.08±0.26 mg/kg) Mn (18.05±0.06 mg/kg) and Fe (7.24±0.05 mg/kg). While the lowest were: Pb (0.19±0.05 mg/kg), Cu (13.39±0.06 mg/Kg), Cd (0.05±0.71 mg/Kg), Cr (0.02±0.63 mg/Kg), Zn (16.30±0.03 mg/Kg), Ni (0.02±0.13 mg/Kg) Mn (13.03±0.05 mg/kg), and Fe (3.94±0.03 mg/Kg).

The concentration of heavy metal in *Mormyrus rume* for the different sampling stations is presented in Table 6. The highest concentrations of heavy metals were: Pb (0.41±0.92 mg/kg), Cu (24.15±0.06 mg/kg), Cd (0.09±0.02 mg/kg), Cr (0.05±0.38 mg/kg), Zn (19.19±0.33 mg/kg), Ni (0.08±0.55 mg/kg) Mn (26.64±0.01 mg/kg) and Fe (9.10±0.06 mg/kg). While the lowest were: Pb (0.19±0.24 mg/Kg), Cu (19.22±0.03 mg/Kg), Cd (0.02±0.51 mg/Kg), Cr (0.01±0.56 mg/kg), Zn (13.08±0.01 mg/Kg), Ni (0.01±0.91 mg/kg), Mn (21.62±0.01 mg/kg), and Fe (6.01±0.02 mg/kg) respectively for each sampling station.

Table 3: concentration of Heavy metals in *Chrysichthys nigrodigitatus* for the different sampling stations

Heavy metals	Mbiabo Edere	Ikot Otu	Ikot Iwatt	Itu Head Bridge
Pb	0.02±0.31	0.02±0.12	0.01±0.92	0.02±0.79
Cu	24.08±0.64	23.06±0.23	22.67±0.10	21.89±0.06
Cd	0.12±0.17	0.07±0.92	0.23±0.41	0.02±0.58
Cr	0.01±0.26	0.05±0.34	0.01±0.32	0.02±0.34
Zn	18.00±1.00	14.01±0.81	10.03±0.03	17.13±0.32
Ni	0.06±0.51	0.05±0.51	0.05±0.06	1.19±0.51
Mn	29.40±0.01	28.67±0.41	28.62±0.01	26.50±0.04
Fe	4.87±1.02	5.92±0.03	6.90±0.66	6.60±0.54

Table 4: Concentration of heavy metal in *Schilbe mystus* for the different sampling stations

Heavy metals	Mbiabo Edere	Ikot Otu	Ikot Iwatt	Itu Head Bridge
Pb	0.01±0.53	0.14±0.06	0.61±0.62	0.72±0.79
Cu	18.80±0.16	17.62±0.03	22.67±0.01	20.64±0.05
Cd	0.03±0.57	0.06±0.47	0.02±0.31	0.02±0.68
Cr	0.06±0.16	0.08±0.53	0.01±0.32	0.06±0.58
Zn	10.00±0.91	13.01±0.16	18.00±0.33	19.23±0.03
Ni	0.06±0.71	0.03±0.25	0.08±0.15	0.05±0.91
Mn	23.44±0.21	17.67±0.01	21.62±0.1	19.65±0.04
Fe	5.87±0.12	4.81±0.05	4.90±0.66	3.60±0.14

Table 5: Concentrations of heavy metals in *Synodontis schall* for the different sampling station

Heavy metals	Mbiabo Edere	Ikot Out	Ikot Iwatt	Itu Head Bridge
Pb	0.83±0.53	0.42±0.06	0.51±0.92	0.19±0.05
Cu	20.10±0.06	21.62±0.03	16.67±0.01	13.39±0.06
Cd	0.05±0.71	0.08±0.92	0.09±0.51	0.05±0.87
Cr	0.03±0.51	0.02±0.63	0.04±0.72	0.07±0.83
Zn	20.10±0.01	19.00±0.02	16.30±0.03	19.32±0.03
Ni	0.02±0.13	0.02±0.75	0.08±0.26	0.05±0.21
Mn	16.01±0.05	13.03±0.05	18.05±0.06	17.00±0.04
Fe	7.24±0.05	3.94±0.03	7.06±0.06	4.60±0.24

Table 6: Concentrations of heavy metal in *Mormyrus rume* for the different sampling stations

Heavy metals	Mbiabo Edere	Ikot Otu	Ikot Iwatt	Itu Head Bridge
Pb	0.19±0.24	0.24±0.05	0.41±0.92	0.03±0.49
Cu	24.15±0.06	19.22±0.03	21.67±0.01	21.89±0.06
Cd	0.04±0.75	0.09±0.02	0.02±0.51	0.04±0.48
Cr	0.02±0.27	0.04±0.53	0.01±0.56	0.05±0.38
Zn	13.08±0.01	16.01±0.06	14.00±0.33	19.19±0.33
Ni	0.05±0.51	0.08±0.55	0.05±0.96	0.01±0.91
Mn	26.64±0.01	24.00±0.01	22.39±0.08	21.62±0.01
Fe	6.97±0.02	6.01±0.02	9.10±0.06	7.60±0.04

DOI: 10.56892/bima.v8i2.690

DISCUSSION

Globally, Fish muscle constitutes a major edible component of fish food and provides humans with nutrients such as protein, low-level cholesterol fatty acids, vitamins, and essential minerals (Agbugui *et al.,* 2011). Pollution from anthropogenic activities contaminates the aquatic environment with heavy metals, pesticides, polycyclic aromatic hydrocarbons, nonylphenol, and other contaminants of emerging concern thereby rendering fish and other aquatic resources unsuitable for consumption (Obot and Jacob, 2024; Jacob *et al.,* 2024a). Fish and other aquatic organisms can bioaccumulate and store contaminants in their body for a long time which creates a pathway for biotransformation and transferred to man through trophic interactions. Hence, the consumption of contaminated food material including aquatic for remains the major source of human exposure to the risks of heavy metal pollution (Liu *et al.,* 2010).

Heavy metals are hazardous environmental contaminants with severe toxicological consequences in humans (Indrajit *et al.,* 2011, Lopez-Alonso 2012). This study was carried out to investigate the concentration of heavy metals in four commercially important fish species from the Cross River System, Itu Local Government Area, Akwa Ibom State, Nigeria, and to provide first-hand information on the potential of consuming contaminated fish food from this river. Herein, there was a significant difference ($P \leq 0.05$) in the measured heavy metals concentration among the different fishes. However, there was no significant difference ($P \geq 0.05$) in the concentration of Cd during the study period.

The concentrations of Pb in the fish samples were below the National Environment Standards and Regulations Enforcement Agency (NESREA, 2007) recommended level (10 mg/Kg) for Pb in edible products. Similarly, our result did not agree with the reports of (Ahmad *et al.,* 2009; Opaluwa *et al.,* 2012; Hossain, *et al.,* 2022). The biological function of Pb has not been fully established. Pb is a comparatively unreactive element with a weak metallic characteristic and an amphoteric nature. Pb is a toxic, non-biodegradable metal, and can affect the central nervous system. Numerous processes such as mining and smelting of metalliferous ores, burning of leaded gasoline, municipal sewage, and industrial wastes enriched in Pb, and paints introduce Pb into the aquatic environment.

About 30 mg/kg of Zn is allowed under (FAO, 1983) guidelines Table 2. However, our result was agreed with the reports of (Rahman *et al.,* 2012; Hossain *et al.,* 2022). They reported low Zn Concentrations in the Meghna River and Bangshi River respectively. High concentrations of Zn in the body may result in Zn poisoning, diarrhea, and fever (Chi *et al.,* 2007). This may suggest low toxicity of zinc in fishes from the Cross River system. Another exposure route also requires further investigation in this respect. However, these contaminants may originate from sewage discharge due to agricultural activities, urban/domestic discharge, electroplating, smelting and refining, mining, and biosolids (Ramzy *et al.,* 2021). Excessive Zn intake resulted in severe toxicological effects such as electrolyte imbalance, nausea, anemia, and lethargy (Prasad, 1984).

The WHO and FAO (1983) recommended level for Cd was set at 1 mg/kg. Moreover, the European Community legislation (2001) and the Codex Committee on Food Additives set the maximum recommended limits for Cd contaminants as 0.05 mg/kg and 0.5 mg/kg, respectively. In this study, the concentrations of Cd analyzed in the current study were below FAO/WHO and European Union

DOI: 10.56892/bima.v8i2.690

recommendation levels. The presence of heavy metals in fish Cross River System in south-south Nigeria showed differences among the economically important species. (Saha *et al.,* 2020) reported Cd contamination levels in various commercial feeds and collected fishes in aquaculture systems in Bangladesh which ranged from (0.23-1.28 mg/kg) and (0.87-1.35 mg/kg), respectively. This does not agree with the report of this study. Human exposure to Cd contamination for a long time can lead to renal failure, bone softening, and prostate cancer (Thomson *et al.,* 2008; Gray *et al.,* 2005). Cr concentrations in all the species sampled were below the (WHO, 1989) recommended standard for edible items.

The significant role of Cr includes the metabolism of Lipids and glucose in the diet (Ahmed, *et al.,* 2015; Ali *et al.,* 2018). The absorption of lipids, proteins, and glucose can be slowed down and denatured by Cr deficiency (Calabrese, 1985). Consequently, excessive Cr consumption can result in kidneys, liver, and lungs deterioration and impaired respiration (Ali, *et al.,* 2020). The concentration of Cr reported in this study was below the concentration recommended by European Community legislation (2001) and the Codex Committee on Food.

Ni concentrations for the four fish species were lower than the mean Ni concentration in fishes from the tropical estuary of the Karnafhuli River in Bangladesh was reported at 0.233 mg/kg, which was higher than the value reported in the study. Fish from Bangladesh's subtropical coastal area had higher mean Ni concentrations of 0.4 ± 0.3 mg/g (Hossain, *et al.,* 2022). Ni is one of the ubiquitous elements. At extremely low concentrations, Ni can elicit severe toxicological responses such as lung inflammation, fibrosis, emphysema, cancer, and tumors (Forti *et al.,* 2011). Similarly, Aleksandra and Urszula (2008) reported that Ni toxicity may result in skin irritability, and toxicity of the lungs, neurological system, and mucosal membranes. The major sources of Ni include Volcanic eruptions, landfills, forest fires, bubble bursting and gas exchange in the ocean, soil weathering and geological materials, industrial effluents, kitchen appliances, surgical instruments, steel alloys, automobile batteries are the major source of Ni in the aquatic environment (Ramzy *et al.,* 2021)

The concentrations of Cu recorded in this study for the different fish species were higher than the concentration reported by (Mortuza and Misned, 2015) in cultured fishes of Rajshahi. (Odoemelam *et al.,* 2020 reported (36.00 mg/kg) copper concentration in surface water and sediment of the lower Cross River System in Akwa Ibom State, Nigeria. This did not agree with the findings in this study. Copper is an essential heavy metal and a critical component of a living organism. Cu plays a role in the production of hemoglobin, myelin, melanin, and the normal functioning of the thyroid gland. Deficiency of Cu can result in hernias, aneurysms, and blood vessel breakage manifesting as bruising or nosebleeds (Osredkar and Sustar 2011). Consequently, extreme exposure to Cu can lead to cellular damage and neurotoxicity (Parmar, and Thakur 2013). The main source of Cu is the electroplating industry, mining, biosolids, smelting, and refining (Ramzy *et al.,* 2021).

Manganese enhances growth and good health in children (Zarith and Mohd, 2015). Severe skeletal and reproductive abnormalities can be caused by Mn deficiencies in humans (Zarith and Mohd, 2015). Also, congenital malformations in offspring and low efficiency of the reproductive system can be caused by Mn deficiencies (Saha and Zaman, 2013). However, a high concentration of Mn is toxic and may lead to neurologic and psychological disorders (Perl and Olanow, 2007; Saha and Zaman, 2013; Jacob *et al.,* 2024b). The main sources of Mn are Municipal wastewater discharges, sewage sludge, mining, and mineral processing, emissions from alloy, steel, and iron production, and combustion of fossil fuels (Ramzy *et al.,* 2021).

About 0.03 g (30 mg) of iron is required by the body per day typically for erythropoiesis (Winter*et al.,* 2014; Coates, 2014). A substantial amount of iron required by the body is obtained from the macrophagic-mediated digestion of short-lived reticulocytes and the subsequent recycling of iron from the breakdown of the heme prosthetic group found in hemoglobin. Iron is an essential micronutrient with severe toxicity to biological systems. It generates free radicals by interconverting between ferrous (Fe^{2+}) and ferric (Fe^{3+}) forms. Iron plays a critical role during the formation of heme. A molecule with the functionally prosthetic group for oxygen-binding proteins like hemoglobin and myoglobin (Zhang, 2014). It also helps in the formation of iron-sulfur clusters that mediate redox and electron transfer reactions for a large number of proteins including cytochromes, ferredoxins, and dehydrogenases. Most biochemical reaction in living organisms is enhanced by the Fe cofactor. However, the toxicity of Fe in the body may resulted in cancer (Puliyel *et al.,* 2015; Bystrom and Rivella 2015; Torti, and Torti, 2013), ischemia (Zhao*et al.,* 1997; Minhas *et al.,* 2014), lung diseases (Ghio, 2009), aging (Mallikarjun, 2014), neurodegenerative diseases (Levi *et al.,* 2014; Stankiewicz *et al.,* 2014; Mena *et al.,* 2015; Belaidi, and Bush 2015), iron mediating programmed cell death (necroptosis-type PCD called ferroptosis) (Xie, *et al.,* 2016).

Accumulation and toxicity of heavy metals in fish occur by natural and anthropogenic processes. Heavy metal contaminants exhibit different toxicological effects at different concentrations in fish due to the affinity between heavy metals and fish organs. The formation of metal-binding proteins such as metallothioneins in metabolic organs makes the accumulation of heavy metals vary significantly among different fish species (Torreblanca, *et al.,* 2014; Waheed *et al.,* 2014; Kalay and Canli, 2000).

CONCLUSION

This study provides valuable insight into the levels of heavy metals in ecologically and economically important fish species in the Cross River System, Itu Local Government Area, Akwa Ibom State, Nigeria. The concentration of the heavy metals analyzed from all the fish species was lower than the recommended limits of major regulatory bodies such as NESREA, EC, and FAO/WHO. In our study, we noticed that bioaccumulation and bio concentration of heavy metals vary enormously with the different fish species. This may be due to their food and feeding habits and trophic levels. Since fish constitute a major source of food and cheap protein for people in Akwa Ibom state and Nigeria, bioaccumulation of heavy metals in fish bodies and organs can be transferred to humans by feeding on contaminated fish food. This may result in serious illnesses such as cardiovascular diseases, neurodegenerative disorders, and cancer. To prevent continuous pollution of the river, industrial effluents, domestic sewage, and other pollutants should be properly treated before discharge into the environment. Furthermore, proper environmental monitoring programmes should be put in place to ensure fish quality and aquatic life in the Cross River System.

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