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Heavy Metal Tissue Concentrations and Human Health Risks of Commonly Consumed Fish Species *Coptodon zilli* and *Chrysichthys nigrodigitatus* Sold from Markets in Warri, Metropolitan City, Delta State, Nigeria.

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ABSTRACT: Contamination of heavy metals in aquatic organisms is a significant cause of concern worldwide due to the fact that these contaminants may bioaccumulate in tissues and pose health risks to humans who consume these fish species. Therefore, the objective of this paper was to investigate the tissue concentrations of Cu, Pb, Zn, Fe and Cd in two commonly consumed fish species, *Coptodon zilli and Chrysichthys nigrodigitatus*, from markets in Warri Metropolitan City, Delta State, Nigeria, using Atomic Absorption Spectrophometer (Model AA 500L). The metals were ranked in order of Fe> Zn>Cu>Pb>Cd in the tissues of both fish species. The mean values of Fe, Zn and Cd in both fish species were above the permissible limits of FAO/WHO. The estimated daily intake (EDI) was below the provisional tolerable intake (PTI). The target hazard quotient and hazard index of all metals were below 1, indicating that the consumption of these fish species is not hazardous to the population of Warri community. However, continuous monitoring of the levels of heavy metals in various fish species commonly eaten is recommended, as the possible accumulation of these metals can eventually pose a human risk to the populace.

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Environmental pollution is a global concern, with heavy metals being a significant group of contaminants. Human activities introduce heavy metals into the aquatic environment, a significant portion is taken up by the sediment and can be recycled through chemical, physical, and biological processes. This leads to the dissolution of the residual pollutants in the water column, which are then absorbed by the aquatic organisms and affect biological processes, particularly in productive aquatic habitats (Gheorge *et al.*, 2017). Globally, fish intake has risen in recent years as a result of growing recognition of its nutritional advantages (Barawo *et al.*, 2018). Fish is a notable protein source, abundant in minerals, vitamins, and omega-3 fatty acids that are crucial for fostering optimal human growth and development (Leech, 2019). Nevertheless, humans can consume fish that may contain harmful substances, such as heavy metals, in high concentrations. This then predisposes them to the health risks associated with consuming these fish (Isangedighi and David, 2019). Varol *et al.* (2019) reported health risks that may occur in the liver, kidney, heart, reproductive organs, circulatory system, and developmental abnormalities, particularly in children but also in adults that consume this contaminated aquatic food. Consequently, different

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species of fish are commonly utilised as biomarkers for investigating the accumulation of toxic metallic compounds in aquatic habitats (Kwaansa-Ansah *et al.*, 2019).

In most metropolitan cities in Nigeria, such as Warri City, fresh fish products are often purchased from marketplaces and their provenance is often unknown. Most fresh species are obtained from highly disrupted aquatic ecosystems affected by human activities. This increases the susceptibility of those who consume these fish species to a higher risk of health problems. Thus, the objective of this paper was to investigate the tissue concentrations of Cu, Pb, Zn, Fe and Cd in two commonly consumed fish species, *Coptodon zilli* and *Chrysichthys nigrodigitatus* from markets in Warri Metropolitan City, Delta State, Nigeria.

MATERIALS AND METHODS

*Sample Collection and Preparation***:** Samples (N=30) of *Coptodon zilli* and *Chrysichthys nigrodigitatus* were purchased from fish mongers at two main markets in Warri City metropolis. Fish were collected in duplicates, from Ogbe-Ijoh (5°29'0"N, 5°44'0"E) and Mclver (5°30'27.5"N 5°50'00.9"E) respectively from January – March 2022. Samples were purchased once a month and, the samples collected were transported to the laboratory in clean black polyethylene bags with ice to prevent the samples from decaying. Fish were identified using standard freshwater fishes of Nigeria guides (Idodo-Umeh, 2003). All fish samples were dissected and the livers, gills and muscles were carefully removed for metals analysis.

Digestion and Heavy Metal Analysis: The microwave digestion of 2 g of fish flesh was carried out in Teflon containers using 6 mL of concentrated HNO_3, 2 mL of 30 % H_2O_2 and 2 mL of HF. The HF was disposed of by introducing H_3BO_3, in accordance with the US EPA Method 3052 in 1996. The solution was transferred into a volumetric flask and then diluted with milliQ water until the total volume reached 100 mL. The solution was diluted by adding 10 mL of HNO_3 to 1mL of the solution (Suresh, 2011). The glassware and plastic containers were washed with a 10 % HNO_3, solution, followed by thorough rinsing with milliQ water. The concentrations of heavy metals (Fe, Cd, Cu Zn and Pb) were quantified using Atomic Absorption Spectrometry (AAS) (Model AA 500L).

Human Health Risk Assessment: Estimated Daily Intake (EDI): The Estimated Daily Intake (EDI) was determined using the following (Korkmaz *et al*., 2012).

$$EDI = Cmetal\ x\ \frac{W}{M} \quad (1)$$

C; heavy metals in the fish organs (mg/kg); W, daily average intake of fish (0.025 kg/day for children and 0.068 kg/day for adults); M, bodyweight (30 kg for children and 70 kg for adults).

Target Hazard Quotient (Non-carcinogenic risks): Target hazard quotient was determined via the following (Varol *et al.*, 2017)

$$\mathrm{THQ} = \frac{(EFxEDx\ IRdx\ C)}{(\mathrm{RfD}\ x\ \mathrm{BW}\ x\ \mathrm{AT})} \quad (2)$$

Target Hazard Quotient (THQ) is a measure of the risk that is not related to cancer. EF; exposure frequency per year (365 days), ED; exposure duration (70 years for adults and six years for children), Ird; ingestion rate (52.5 g/day for children and 55.8 g/day for adults), C; heavy metal concentration in aquatic food, RfD; oral reference dose expressed (Cu-40, Zn-300, Fe-400) and BW; average body weight (70 kg for an adult and 30 kg for a child).

Health risk assessment of the toxicants was determined by interpreting the results of the Hazard Quotient (HQ) and Hazard Index (HI). Values below one for either HQ or HI indicate no danger, whereas higher values indicate an increased risk level for the manifestation of long-term health hazards caused by toxicants.

Hazard Index (HI): Hazard index (HI) has been established to evaluate the possible risk to human health resulting from exposure to certain heavy metals. The hazard index is determined by adding together the Target Hazard Quotients for each heavy metal, as computed using the below equation (Korkmaz *et al*., 2012).

$$\mathrm{THQ} = \sum \mathrm{THQ} = \mathrm{THQFe} + \mathrm{THQCu} + \mathrm{THQZn} \quad (3)$$

It presupposes that the extent of the negative impact will be directly related to the combined amount of exposure to various metals. Additionally, it assumes comparable operational processes that have a direct and proportional impact on the specific organ being studied.

Statistical Analysis: All data was analysed using SPSS 16 and Microsoft Excel 16. Data was presented as mean ± standard error. Significant difference ($p<0.05$) between sampling stations was determined using a t-test. Values were considered significant at $p<0.05$.

RESULTS AND DISCUSSION

Heavy Metal Concentration in Fish Tissues: The presence of significant heavy metals concentrations in an aquatic ecosystem is a major concern due to their

toxicity, persistent nature, and potential to bio-magnify within the food chain (Maurya and Malik, 2018). The summary of the average mean concentration of heavy metals in *Coptodon zilli* and *Chrysichthys nigrodigitatus* from the fish market in Warri metropolis in fish organs is presented in Table 1. The mean concentration of heavy metals was in the order liver>gills>muscle for Fe> Zn>Cu>Pb>Cd in the tissues of both fish species. The findings indicate that the metal concentrations in all fish organs exceeded those found in the muscles. This has been documented by several other studies (Rajkowska and Protasowicki, 2013; Rajeshknur and Li, 2018). Liang *et al.* (1999) reported elevated metal levels in the internal organs of fish and their significant function in storing trace metals. Iron (Fe) mean ranged from 31.31 mg/kg to 601.73 mg/kg, the highest and lowest values were found in *Coptodon zilli* across the various fish tissues and markets.

Table 1: Mean ± SD Concentrations of Heavy Metals (mg/kg) in Fish Tissues in *Coptodon zilli* and *Chrysichthys nigrodigitatus* from Market in Warri City

	Fish Species	Heavy Metals				
		Gills				
		Fe	Pb	Cd	Cu	Zn
Ogbe-Ijoh	*C. zilli*	218.18 ±4.69	5.05 ± 4.95	ND	11.31 ±1.11	92.32±10.89
	C. nigrodigitatus	226.97±37.65	0.15±0.05	ND	10.35±3.85	82.26±16.36
Mclver	*C. zilli*	214.75± 65.47	0.20 ± 0.00	ND	9.11 ±0.61	103.32±16.00
	C. nigrodigitatus	162.75±33.98	0.15±0.00	ND	9.78±1.03	79.02±0.48
		Liver				
Ogbe-Ijoh	*C. zilli*	601.73±3.60	0.38±0.13	0.13±0.03	39.36±9.25	195.3±19.70
	C. nigrodigitatus	583.05±102.47	0.34±0.06	0.15±0.00	44.52±18.90	197.85±8.55
Mclver	*C. zilli*	489.5±11.87	0.18±0.08	0.10±0.00	40.51±1.59	176.13±25.87
	C. nigrodigitatus	217.42±168.02	0.15±0.05	ND	28.25±4.05	149.52±21.09
		Muscles				
Ogbe-Ijoh	*C. zilli*	46.15±8.05	ND	ND	3.11±0.61	35.03±1.31
	C. nigrodigitatus	43.8±24.70	ND	ND	2.85±0.87	28.19±1.47
Mclver	*C. zilli*	31.31±12.91	ND	ND	3.28±0.33	35.58±4.15
	C. nigrodigitatus	49.9±21.56	ND	ND	3.39±1.34	40.96±15.24

Not detected (ND

The Fe content was high beyond the permissible limits of 189 mg/kg set by FAO/WHO (1985), in almost all the fish tissues. Fe is a vital mineral necessary for human growth and development. Insufficient iron levels in the body can lead to the development of anaemia, which can lead to significant health consequences. Nevertheless, excessive amounts of iron can result in acute symptoms like constipation, stomach discomfort, vomiting, and diarrhoea. Prolonged exposure to excessive dosages beyond the maximum allowable limit can result in organ failure, coma, seizures, and mortality (National Institutes of Health, 2022). Copper (Cu) is a crucial micronutrient required for the proper development and metabolic function of living organisms (Padrilah *et al.*, 2018). Cu was found at high concentrations in the livers of all two species of fish. *Coptodon zilli* and *Chrysichthys nigrodigitatus* had the minimum (3.11 mg/kg) and maximum (44.52 mg/kg) Cu concentrations, respectively. A high Cu concentration can be detrimental to the kidney and liver (ATSDR, 2021).

Lead (Pb) and cadmium (Cd) were not detected in the muscles of the fish species which are important parts for human consumption, however they were detected in the livers and gills of both species from the two sampling locations. Both metals were above the permissible limits set by FAO 2012 at 0.5 mg/kg for Pb and 0.05 mg/kg for Cd. The observed detection of heavy metals levels in the gills and liver indicate the detoxifying capability of these organs to toxic metals through the presence of metal binding proteins called metallothioneins; it is probable this may be the reason why metals were not detected in the muscle (Subotic *et al.*, 2013).

Both Pb and Cd have harmful health risks associated with neurological, respiratory and cardiovascular disorders (Balili-Mood *et al.*, 2021). The fish exhibiting the highest average Zn contents varied between 28.19 and 197.85 mg/kg. Both fish species examined for Zn concentrations were above the acceptable limits as suggested by the EC (2014). However, humans have been associated with potential health risks from chronic exposure to Zn compounds.

Mean Percentage Composition in the Tissues of Coptodon zilli and Chrysichthys nigrodigitatus: The mean percentage content found in the organs of both species is represented in Figure 1. Ranked in the order of Fe>Zn>Cu>Pb>Cd, it clearly indicates that both fish species bioaccumulated Fe and Zn the most from their polluted environments thereby depositing these trace metals in their tissues.

Human Health Risk Assessment: The EDI values varied between 0.003 and 0.060 for both the metals and the adult and child population. The levels of EDI were shown to be less than the Tolerable Daily Intake (TDI). This indicates that there is no adverse health risk associated with consuming the selected fish species from this study area. The THQ serves as a framework for assessing the average daily consumption of pollutants in the diet and comparing it to the reference dose (Allauddin *et al.,* 2020). A THQ value greater than 1 implies a significant potential danger, whereas a THQ value less than 1 suggests a minimal and insignificant risk. Humans are predisposed to the combined effects of contaminants. As a result, a cumulative risk index is computed, which takes into account all the heavy metals present in each unique fish species. The Hazard Index (HI) refers to the cumulative total of all the THQs (Target Hazard Quotient) for each specific fish species (USEPA, 2005). In this study, the THQ and HI values were both below 1, indicating that there is no significant health risk associated with consuming fish species from this particular area.

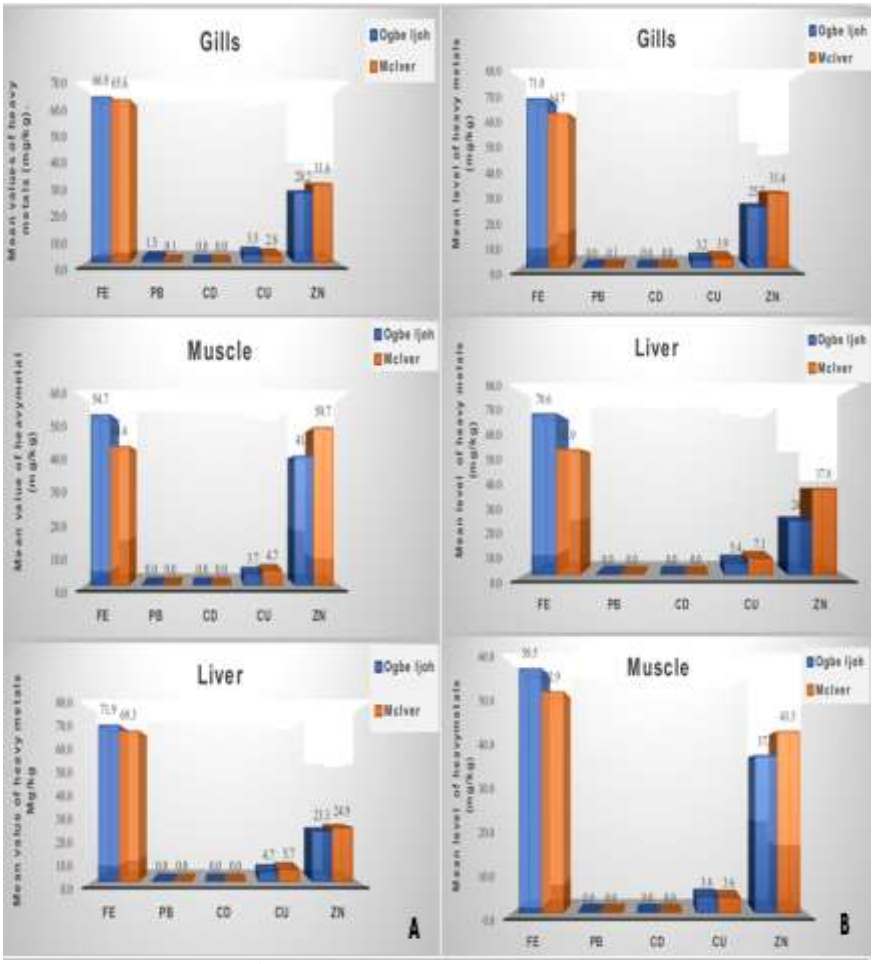

Fig 1: The Percentage Composition of Heavy Metals (A) *Coptodon Zilli* (B) Chrysichthys *nigrodigitatus* Fish Species

Table 2: Estimated Daily Intake (EDI mg/kg), Tolerable Daily Intake (TDI mg/kg), Target Health Quotient (THQ) and Hazard Index (HI).

Heavy metal	***C. zilli*** **EDI**		**TDI**	***C. nigrodigitatus*** **EDI**		***C. zilli*** **THQ**		***C. nigrodigitatus*** **THQ**	
	Adult	Children		Adult	Children	Adult	Children	Adult	Children
Fe	0.060	0.051	0.8	0.066	0.057	0.07	0.24	0.72	0.31
Cu	0.003	0.003	0.5	0.068	0.004	0.06	0.14	0.06	0.065
Zn	0.051	0.044	0.3	0.047	0.041	0.09	0.2	0.20	0.09
HI						**0.22**	**0.58**	**0.98**	**0.46**

Conclusion: The present study investigated the levels of five heavy metals (Fe, Pb, Cd, Cu, and Zn) in fish consumed by the Warri population and assessed their human health risk. The results indicated that the levels of Fe, Cu, and Zn in fish exceeded the guidelines set by international organisations. However, the estimated

weekly intake of these metals through fish consumption remained below the acceptable limit. The target and hazard risk assessments revealed that exposure to these metals does not pose a threat to human health in the Warri population. Among the metals studied, Fe was found to be the primary risk factor for human health, followed by Cu and Zn. To prevent the potential accumulation of these metals. It is recommended for continuous monitoring of the levels of heavy metals in various fish species commonly consumed widely by the populace in Warri City.

Declaration of Conflict of Interest: The authors declare no conflict of interest

Data Availability Statement: Data are available upon request from the first author or corresponding author.

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