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Abstract

Heavy metal content and its associated hazard risk to human health were assessed in dried fish species commonly consumed in Benin city metropolis. In this study, three commercially available dried fish species Elops lacerta (ten pounder), Clarias gariepinus (catfish) and Cynoglossus senegalensis (sole fish) were purchased from selected markets in Edo State, Southern Nigeria. Result indicates that the heavy metal content was higher in Elops lacerta fish than the other fish species. On average, levels of Cd (0.23±0.03 mg/kg), Cu (10.50±2.87 mg/kg), Pb (0.14±0.03 mg/kg) and Zn (24.97±2.41 mg/kg) in all three fish species were below the maximum acceptable limits except for Fe (44.44 ±3.18 mg/kg). The metal pollution index (MPI) in fish species was below 100. Estimated daily intake (EDI) for the metals was lower than the tolerable daily intake (TDI). Target hazard quotient (THQ) of all metals was below 1, but the total THQ values for associated metals were above 1 for children, suggesting a noncarcinogenic health risk to children. Target carcinogenic risk (TR) was within the acceptable limits of 10⁻⁴ to 10⁻⁶ indicating low carcinogenic risk to the consumer. We recommend that the dried fish species in the studied areas should be closely monitored to protect consumer health, especially in children.

Keywords: Dried fish, health risk, heavy metals, hazard quotient, cancer risk

INTRODUCTION

Fish consumption is a common protein source in many developing nations, including Nigeria. Approximately 41% of Nigeria's total animal protein intake comes from fish products and worldwide 6% of all protein is consumed (Food and Agriculture Organization, 2012). Fish is regarded as a relatively less expensive source of protein, vitamins, lipids, and minerals than animal protein and is linked with a wide array of health benefits to man (Neff et al., 2014). Public health authorities have recommended fish eating in equivalent amounts to 1-2 servings per week to prevent diet-related chronic illnesses and lower the risk of heart and stroke diseases (Zhao et al., 2019). Production volumes of fish had fallen below expectations over the last decades in Nigeria, with 3.6 million metric tonnes of domestic fish demand reported annually, while current production stands at 1.1 million tonnes (Nnodim, 2014), leaving a gap between supply and demand of about 2.5 million metric tonnes for 200 million plus Nigerians. United Nation Food and Agriculture Organization in 2018 reported that fish consumption has outpaced fish production due to world population growth. In Nigeria, household fish consumption is 13.3 kg annually, which is low compared to the global average of 20.3 kg per capita (FAO, 2018), raising enormous challenges for food and nutrition security. The Nigeria fisheries industry has contributed significantly to the country's economy and poverty

alleviation efforts accounting for 3.25% of the country's gross domestic product (Olaoye and Ojebiyi, 2018). To enhance the country's fishing industry, prudent postharvest treatment procedures are essential to increase shelf life and its uses, as fresh fish is a highly perishable food that deteriorates rapidly unless preserved (Mahmud et al., 2018). Dry fish is the second most commonly consumed fish form in most Nigerian households and a preferred cheap method of preservation (Onyeneke et al., 2020). This method removes the water content from the fish at varying degrees, inhibiting the growth of microorganisms present in the fish over smouldering wood, sawdust, or other sources of energy using traditional kilns (Ako and Salihu, 2004). Dried fish can be contaminated with heavy metals through its surrounding water body, preservation processing and handling (Alquezar et al., 2006). Heavy metals are ubiquitous and can bio-accumulate in the tissues (muscles, gills, liver, scales, skin) of fish (Canli and Atli, 2003), over a long period of time. When these metals accumulate beyond the permissible levels, they can become harmful to humans, even at minute levels (Jaishankar et al., 2014). In light of growing concerns regarding the quality and safety of food in several regions of the world, limited data is available on assessing the potential human health risk of dried fish species commonly consumed by the Nigerian population. Thus, this study determined the potential heavy metal contamination levels as well as the human health risk associated with adults and children consuming these dried fish species since dried fish has a unique position in Nigerian cuisine and their harvesting and processing remain crude.

MATERIALS AND METHODS

Sample Collection and Preparation

Three commonly consumed fish species; *Elops lacerta* (ten pounder), *Clarias gariepinus* (cat fish) and *Cynoglossus senegalensis* (sole fish) were purchased from fish mongers in Benin City metropolis. These fish samples were collected in triplicate, from New Benin (6°21′ 4.6728 5°37′ 48.3888E″) Uselu (6°22′ 26.2128N 5036′ 49.1688E″) markets and transported to the laboratory with sterile polythene bags and labelled properly. Using stainless steel scissors, the muscle of the fish samples was collected and cut into tiny portions in the laboratory. Agate mortar and pestle were used to grind the samples into a fine powder, sieved with a clean, airtight plastic sieve (2 mm), and stored in a vacuum desiccator in preparation for heavy metals analysis.

Digestion and Analysis of Fish

Fish samples were digested with the procedure described by Poldoski (1980). One (1) g of the dried powdered fish was transferred into 250 mL bottled flask to this 10 mL of conc HNO₃ and 10 mL H_2O_2 was added left for an hour. The flask was heated gently at 160 °C in a sand bath on the hot plate until a volume reduction to 5 mL. The digested sample was cooled and transferred to 25 mL volumetric flask filled with de-ionized water and retained in plastic bottles for further heavy metals analysis using Atomic Absorption Spectrophotometer (Perkin Elmer 1996).

Metal Pollution Index

The metal pollution index (MPI) was determined following the equation reported by Usero *et al.* (2016).

MPI= $(CM_1 X CM_2 X CM_3 X ... CM_n)^{1/n}$ ------(1)

 CM_1 , the first metal level, CM_2 , the second metal level, CM_3 , the third metal level and CM_n , the nth metal level in the sample of the species of interest (mg/kg dry weight).

Human Health Risk Assessment Estimated Daily Intake

The daily estimated intake (EDI) of heavy metals for adults and children were determined using the following (Griboff *et al.*, 2017)

EDI= (C X IRD)/BW ------ (2)

C, heavy metal level in the fish muscle of the species (mg/kg dry weight), IRD, daily mean fish intake rate (55.8 g/day for adults, 52.8 g/day for children) and BW, average body weight (60 kg for adults and 30 kg for children).

Target Hazard Quotient

The target hazard quotient (THQ), is the ratio of the exposure of metals and their reference dose (RFD). A ratio value of 1 indicates a non-significant hazard to human health, while a ratio value greater than 1 indicates hazards to consumers (USEPA, 1989). This is calculated using equation (3) indicated below

$THQ_{S} = (EF X ED X IRD X C) / (RFD X BW X AT) ------ (3)$

THQ, target hazard, EF, frequency of exposure (365 day/year), ED, exposure duration (70 years for adults, 6 years for children), IR, ingestion rate (55.8 g/day for adults, 52.8g/ day for children), C, heavy metal level in fish (mg/kg/dw), RFD, oral reference dose (μ g/kg/day,100 for Fe, 1 for Cd, 40 for Cu, 4 for Pb, 300 for Zn), BW, average body weight (60kg for adult, 30kg for children), AT, average exposure time for non-carcinogenic (ED X 365 days).

Total Target Hazard Quotient

The sum of all the Total Target Hazard Quotient (THQ) amounts was determined to calculate the total non-carcinogenic risk. The total THQ was considered as hazard index (HI) to assess the risk of multiple metal contamination in fish. This is calculated using equation (4) indicated below.

$HI = \sum THQ = THQ (Fe) + THQ (Cd) + THQ (Cu) + THQ (Pb) + THQ (Zn) ------ (4)$

Target Carcinogenic Risk

Target carcinogenic risk (TR) is the likelihood to develop cancer over the course of a person's lifetime when exposed to a carcinogen substance (Zhong *et al.*, 2018). The acceptable limit for lifetime exposure to carcinogens is 10⁻⁴ to 10⁻⁶. This is calculated using equation (5) indicated below.

TR= (EF x ED x IRd x C x CSF) / (BW x AT) x 10^{-3} ------ (5) TR is the target carcinogenic risk and CSF is the oral carcinogenic slope factor (pb-0.009, Cd-0.6) (USEPA, 2011).

Statistical Analysis

Data were analyzed using SPSS 16 and Microsoft Excel 16 packages. The means and standard errors of the metal levels in the various dried fish species were calculated. Significance of difference in metal levels between various fish species was examined using one-way variance of analysis (ANOVA). For each metal, (p< 0.05) mean levels between various fish species were investigated using multivariate post hoc Duncan tests.

RESULTS AND DISCUSSION

Concentration of heavy metals

Table 1 presents the mean concentrations and standard errors of heavy metals found in the dried fish species (*Elops lacerta, Clarias gariepinus, and Cynoglossus senegalensis*). In general, the highest mean level was found in Fe, among the three different fish species. Heavy metals concentrations were in this order Fe>Zn>Cu>Pb>Cd. Compared to other studies on dried fish, this study's fish heavy metals levels (Table 1) were higher than those collected from Benin and Warri metropolis (Edosomwan *et al.*, 2016). Mean Fe concentrations were as high as (60.95 mg/kg) in *Elops lacerta*. The lowest Fe accumulation (34.98 mg/kg) was in *Cynoglossus senegalensis* fish species. According to Anisha *et al.* (2023), dried fish from Kerala coast, South West India had the highest concentration of Fe, which is comparable to this study's finding. Dried fish species of *Elops lacerta* had a higher Fe content than the other two species (p< 0.05) based on their mean differences.

Species	Fe	Cd	Cu	Pb	Zn	MPI
	(Mg/kg)	(Mg/kg)	(Mg/kg)	(Mg/kg)	(Mg/kg)	
	X±S. E	X±S. E	X±S. E	X±S. E	X±S. E	
	(Min-Max)	(Min-Max)	(Min-Max)	(Min-Max)	(Min-Max)	
Elops lacerta	60.95 ± 5.18^{b}	0.13 ± 0.03	14.52 ± 2.01^{b}	0.18 ± 0.03	34.10 ± 3.99^{b}	3.71
(Ten pounder)	(49.69-71-04)	(0.10 - 0.15)	(9.73-19.51)	(0.15 - 0.20)	(24.63-41.50)	
Clarias gariepinus	37.39 ± 2.09^{a}	0.10 ± 0.00	9.91 ± 0.67^{a}	0.10 ± 0.00	24.78 ± 2.10^{a}	2.47
(Catfish)	(32.54-42.73)	(0.10 - 0.10)	(8.19-11.43)	(0.10 - 0.10)	(19.84-0.11)	
Cynoglossus	, , , , , , , , , , , , , , , , , , ,	. ,	, ,	· · · ·	, , , , , , , , , , , , , , , , , , ,	19.3
senegalensis	34.98 ± 2.29^{a}		7.08 ± 0.57^{a}		19.03±1.13ª	
(Sole Fish)	(29.19-39.16)	ND	(5.98 - 8.62)	ND	(16.83 - 22.00)	
Mean	· · · · · · · · · · · · · · · · · · ·		()		()	
	44.44 ±3.18	0.23±0.03	10.50±2.87	0.14±0.03	24.97±2.41	12.61
F (P) Values Guidelines	16.96(0.001)	0.333(0.667)	8.(0.008)	0.333	0.010	
WHO/FEPA (2003)	0.5	3.0	3.0	2.0	60	
NESREA (2009)	NA	0.050	NA	0.20	NA	
FAO (2012)	30	0.05	30	0.5	30	

Table 1: Heavy Metals Concentrations (mg/kg) in Dried Fish Species from Markets in Benin City

Vertically different letters (a and b) indicate statistical differences between fish species at p < 0.05 (Duncan test). Mean (\mathbf{X}), Standard error (S.E) Not available (NA), Not detected (ND).

Nonetheless, Fe mean values in the different fishes were above the WHO (2003), NESREA (2009) and FAO (2012) limits. Fe is an essential element that is involved in arrays of metabolic processes in an organism (Abbaspour *et al.*, 2014). However, it can pose potential hazards that can endanger both human and animal health at either low or high dose levels. Mean Cd levels in all the fish species were below the NESREA (2009) and FAO (2012) limits. *Elops lacerta* (0.13 mg/kg) fish species had the highest mean Cd value while *Clarias gariepinus* had the lowest mean level (0.10 mg/kg). In *Cynoglossus senegalensis*, Cd was not detected in the fish species.

Egbeja et al. (2019) reported a higher mean level of Cd for dried Clarias gariepinus (0.34 mg/kg) from Anyiyba market in Kogi State. Cu mean levels in the fish muscle analyzed varied from 7.08 to 14.52 mg/kg, with the highest concentration (14.52mg/kg), in *Elops lacerta*. However, the lowest mean level (7.08 mg/kg) in *Cynoglossus senegalensis* was far above the WHO (2003) guideline of 3.0 mg/kg, but below the FAO (2012) maximum limit of 30 mg/kg. There was no significant (p>0.05) difference between the mean Cd level among the dried fish species. Pb concentrations in the analyzed fish samples varied from 0.10 to 0.18 mg/kg, with *Elops lacerta* having the highest mean value (0.18 mg/kg). However, the lowest mean value (0.10 mg/kg)in Clarias gariepinus was far above the WHO (2003), NESREA (2009) and FAO (2012) guidelines of Pb. Statistically, no significant difference was found in the mean values of Pb within the different fishes (p > 0.05). Similar to Fe, Cd, Cu, and Pb the highest mean concentration of Zn (34.10 mg/kg) was found in *Elops lacerta*, while the lowest mean level (19.03 mg/kg), was recorded in Cynoglossus senegalensis. Akter et al. (2019) reported that dried fishes from the coastal region of Bangladesh had a lower mean Zn level range from 0.15-1.81 ppm. Zn concentrations in Elops lacerta was below the maximum permissible levels for FAO (2012), but not above WHO (2003). Difference in the mean levels of Zn in the three fish species showed that *Elops lacerta* had higher Zn content than other species (p < 0.05). The estimated MPI ranged from 2.47- 19.2 mg/kg (Table 1). The highest values were found in Cynoglossus senegalensis, while the least value was detected in Clarias gariepinus fish. All the dried fish species showed slight pollution in consumption and this poses non-human health risk implications. The MPI values of dried fish species in this present study were lower than the study of Rakib et al. (2021) on dried fish consumed in Bangladesh

Human Health Risk Assessment

The estimated EDI values for heavy metals in dried fish species in adults and children are presented in Table 2. The EDI values and tolerable daily intake (TDI) were compared. The findings from this study revealed that the EDI values of the five heavy metals were within the TDI values indicating a low potential for consumers' health to be at risk. The EDI values obtained from this study are higher than the findings of Moslen and Miebaka (2017). The target hazard quotient of each heavy metal (Table 2) for adults and children in the dried fish species was below the maximum limit of 1. Total hazard quotient in the dried fish species of all the combined heavy metals suggested the presence of non-carcinogenic human health risk, especially in children who consume the fish species studied.

quotients													
	Elops la	icerta	Clarias	gariepinus	Cynogle senegai	ossus lensis		Elops la	icerta	Clarias	gariepinus	Cynogle senegale	ossus ensis
	EDI (µg/kg/l	bw/day)	EDI (µg/kg/	/bw/day	EDI (µg/kg/	bw/day)	TDI (μg/kg/bw/day)	EDI (µg/kg/	bw/day)	EDI (µg/kg/l	bw/day)	EDI (µg/kg/l	bw/day)
	Adult	Children	Adult	Children	Adult	Children		Adult	Children	Adult	Children	Adult	Children
Fe	56.68	106.65	15.70	65.43	14.69	61.22	800	0.08	0.15	0.05	0.09	0.05	0.09
Cd	0.12	0.22	0.04	0.18	-	-	0.8	0.12	0.23	0.09	0.18	BDL	0.18
Cu	13.51	25.42	4.16	17.35	2.97	12.39	500	0.34	0.64	0.23	0.43	0.16	0.43
Pb	0.16	0.31	0.04	0.18	7.99	-	1.50	0.11	0.21	0.06	0.12	BDL	0.12
Zn	31.71	59.33	10.40	43.11	-	33.11	300	0.11	0.19	0.08	0.15	0.06	0.15
TTHQ								0.76	1.14	0.76	1.42	0.27	0.96

 Table 2: Average daily intake of heavy metals by adults and children and respective hazard quotients

TDI - tolerable daily intake, BDL- Below detection limit, EDI- estimated daily intake, THQ- target hazard quotient and TTHQ- total target hazard quotient

Table 3: Es	stimated carcinogenic risk in Elops lacerta	arcinogenic risk in adult and children for dried fish species Elops lacerta Clarias gariepinus					
		CR					
	Adult	Children	Adult	Children			
Cd	7.25 x 10 ⁻⁵	5.9 x 10 ⁻⁶	5.58 x10-5	4.5 x 10 ⁻⁶			
Pb	3.52 x 10 ⁻⁵	2.8 x 10 ⁻⁶	1.95x 10 ⁻⁵	1.6 x 10 ⁻⁶			

CR-Carcinogenic risk

The target carcinogenic risk (TR) was estimated (Table 3) for Cd and Pb in dried fish species and were within acceptable limits, suggesting that the dried fish species from Benin City markets were safe for human consumption.

CONCLUSION

Five heavy metals in the muscle of dried fish species purchased from markets in Benin City metropolis were investigated for their bioaccumulation rate and human health risk to potential consumers. It was observed that Fe ranked highest among the heavy metals in the tissue of all the fish species and exceeded the acceptable limits. The consumption of dried fish species poses non- human health risks but, collectively the detection of these heavy metals indicated a non-carcinogenic risk to children than adult. Thus, constant monitoring is recommended to avoid an unexpected health hazard that can occur over a long period of consuming these fish species.

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