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POTENTIAL HEALTH RISK ASSESSMENT OF HEAVY METALS [Cd, Cu AND Fe] CONCENTRATIONS IN SOME IMPORTED FROZEN FISH SPECIES CONSUMED IN NIGERIA

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ABSTRACT

The concentrations of three heavy metals [Cd, Cu and Fe] were determined in three species of imported frozen fish widely consumed in Nigeria. Appropriate weights of the dried pulverized fish samples were digested following FAO/SIDA wet digestion method with HNO_3 and H_2O_2 [1 : 1]. The accuracy and precision of the determinations in the study were evaluated using LOD, LOQ and certified standard reference material [DORM-1 for Dog fish]. The human health risk of consumption of the fishes was assessed by calculating the daily exposure rates (D), hazard quotients (HQ) and the total hazard indices (THI) of the metallic toxicants. The chemical analysis results showed that the concentration levels of the elements are of the ranges of 0.001-0.150 mg/Kg; 1.05-12.06 mg/Kg and 8.21 to 85.06 mg/Kg for Cd, Cu and Fe, respectively. Also results of the potential health risk assessment showed that the total hazard index of all the toxicants indicated no risk status from the consumption of the fish species studied.

Key words: Fish, ICP-OES, Heavy metals, Risk assessment, Wet digestion.

INTRODUCTION

The pollution of aquatic environment by heavy metals is a serious global problem. Environmental contamination by heavy metals are not like organic pollutants that may lose toxicity with biodegradation because metals cannot be degraded further and their toxicity can be long lasting[1]. Hence, metals and other fluvial contaminants, in suspension or solution do not simply flow downstream, they are complexed with other compounds, settled to the bottom and ingested by plants and animals or adsorbed to sediments. Consequently, aquatic organisms may acquire heavy metal burden directly from the water via gills or food chain

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mechanism[2]. Fish is a valuable and cheap food item and source of protein to man. Fish are often at the top of aquatic food chain and may concentrate large amount of some heavy metals, which often times endanger public health through consumption of contaminated sea food and irrigated food crops. Hence heavy metals bioaccumulate in fish tissues, which are finally transferred to other animals through food chain.

Fish contribute about 55% of the protein intake in Nigeria, which is one of the largest importers of fish with per capita consumption of 7.52 Kg per annum and a total consumption of 1.2 million metric tons with import making up about 2/3 of the total consumption[3]. The Nigerian Government made a tariff reduction in year 2001 on all fishery products from 25% to 5% and since then, Nigeria has become a major destination for imported seafood.

Recent study reported that the level of Cu in imported frozen fish was higher than the recommended maximum limit by World Health Organization (WHO) while Cd and Fe had values lower than the set limit[4]. However, hazard assessments of these toxicants in imported frozen fish have not been reported.

Health risk assessment is a very important tool to evaluate the consequences of human action and measures the adverse effect to public health. The objectives of this work were to determine the concentrations of some heavy metals in three popular imported frozen fish species consumed in Nigeria and predict their potential health consequences that can serve as scientific basis for decision and policy making.

EXPERIMENTAL

Materials and method

Sample collection and preparation

Specimens of three popular species of imported frozen fish namely, Atlantic Mackerel (*Scomber Scombrus*), Sardines (*sardinella sindesis*) and Stock Fish (Cod) (*Gadus Mangala*) were purchased from retailers in popular fish markets in Nigeria. These species were imported iced from Mauritania, South Africa, Norway and Thailand into Nigeria and highly cherished as sources of protein especially by low income population in the country. From each species, 10 fish were randomly collected and transported in ice-packed coolers to the laboratory. The samplings were replicated quarterly interval for one year as a result of information from the importers that new batch of importation arrived at such interval.

The fish muscles and gills were carefully removed with a plastic knife and oven dried at 105°C to a constant weight. The dried fish samples were homogenized using clean mortar and pestle. The homogenized samples (1.0 g each) were digested in a digestion flask in pent plicate (n = 5) according to FAO/SIDA manual[5]. A 3 mL portion of freshly prepared 1 : 1 HNO_3/H_2O_2 was added and the content heated for 1h on a hot plate in a fume cupboard at 160°C until the solution was clear. It was filtered with whatman no 1 filter paper into a 25 mL standard flask and diluted to mark with deionized water transferred to plastic bottles. Sample blanks (n = 10) were carried through the whole digestion processes. Also, the standard reference material (Dorm-1 for dog fish) provided by the National Research Council of Canada was carried through the whole digestion processes as the samples and blanks. The resulting solutions were analyzed using ICP-OES and the results expressed as mg/kg dry weight of fish sample.

Reagents, chemicals and equipment

All reagents used were of analytical grade. Deionized water was used throughout. Multi-elemental standard solution of 1000 mg/L of the metals (Cd, Cu and Fe) were purchased from Merck (Darmstalt, Germany) for calibration. HNO_3 (65%) and H_2O_2 (30%) used for wet digestion were also purchased from Merck. Agilent 710 –ES axial ICP-OES was used. The instrument includes are echelle polychromator and a megapixal CCD detector providing full wavelength coverage from 177 to 785 mm. ICP Expert II software version 1.0 was used for instrument operation parameters of the instrument operations. The operating parameters of the instrument are shown in Table 1.

Table 1: Operating parameters of ICP-OES machine

Parameter	Instrument operating condition
Power	2 Kw
Plasma gas flow:	Argon at 15 $dm^3\ min^{-1}$
Auxiliary gas flow:	1.5 $dm^3\ min^{-1}$
Nebulizer pressure:	250 kPa
Pump ratio :	15 rpm
Pump tubing:	White-white (inlet) Bleu-blue (outlet)
Sample uptake rate:	1.2 mL min^{-1}
Replicate read time:	30 s
Background correction:	fitted
Replicate readings:	3

ICP-OES determination

The calibration standards were prepared by diluting the stock multi elemental standard solution (1000 mg/L) in 0.5% v/v HNO_3 and the calibration curves for all the studied heavy metals were of the range 0.01 to a 1.0 mg/L. The emission intensities were obtained for the most sensitive lines with no spectral interference. The emission wavelengths used for the determinations were Cd (228.802 nm and 226.502 nm), Cu (223.009 nm) and Fe (238.204 nm).

The limit of detection (LOD) and limit of quantitation (LOQ) of the analytical method were estimated by analyzing ten replicate blank solutions and are expressed as the analyte concentration in the sample blank plus three times standard deviation and ten times the standard deviation respectively as shown by the following equations[6].

$$LOD = X_{b1} + 3S_{b1}$$

$$LOQ = X_{b1} + 10S_{b1}$$

Where, X_{b1} is the mean concentration of the and S_{b1} is the standard deviation of the blank

Precision was determined against standard reference material provided by the National Research Council of Canada (DORM-1 for dog fish) by carrying the material through same processes of digestion and analysis as the fish samples and sample blanks.

Potential human risk assessment

The human health risk assessment associated with the fish consumption was characterized using Hazard Quotient (HQ) and Total Hazard Index (THI) developed by United States Environmental Protection Agency following the model below[7].

$$D = (C \times IR \times AF \times EF \times CF) \div BW$$

Where D = Exposure dose (mg/kg/day); C = Contaminant concentration (mg/Kg)

IR = Intake rate of contaminated fish (mg/day); AF = Bioavailability factor (unitless)

EF = Exposure factor (unitless); CF = Conversion factor (10^{-6} Kg/mg)

BW = Body weight (Kg).

In this study, the IR for an adult was calculated from the annual per capita consumption of fish to be 36600 mg/day and for the child to be one third of the adult consumption which is 12200 mg/day[3]. Also the average body weights off adult and child are 70 Kg and 16 Kg, respectively[7]. Bioavailability factor (AF) represent as a percent the total amount of the toxicant ingested that actually enters the blood stream and is available to possibly harm a person and is assumed to be 1 (100%) for screening purposes. EF is taken as 1 as fish intake rate is a daily average.

Hazard Quotient (HQ) is the ratio between exposure dose (D) and oral reference dose (RfD)

HQ =D ÷ RfD and

Total Hazard Index (THI) = $\sum HQ = HQ_{Cd} + HQ_{Cu} + HQ + HQ_{Fe}$

THI calculation for individual fish species

RfD values were taken from US EPA table developed for ingestion as estimates of daily exposures to a substance that are likely o be without a discernable risk of deleterious effects to the general population during a lifetime of exposure[8]. If the hazard quotient (HQ) or total hazard index (THI) is below one, no health risk may occur as a result of ingestion of the fish and the greater the value of HI and THI above 1, the greater is the level of risk associated with the fish consumption[9,10]. Hence, THI = 0.0 to 1 means no hazard; 1.1 to 10 means moderate hazard while greater than 10 means high hazard or risk.

RESULTS AND DISCUSSION

Accuracy and precision of analytical method

The result of the detection and quantiation limits carried out is shown in Table 2.

Table 2: LODs and LOQs of analyzed metals

Metal	LOD μgg^{-1}	LOQ μgg^{-1}
Fe	0.35	1.08
Cu	0.31	0.96
Cd	0.23	0.69

Fish analysis

The analytical result of the 60 samples made from three species of fish widely consumed in Nigeria is shown in Table 3. The mean concentrations of Cd, Cu and Fe in the imported fish species examined are shown in Table 3.

Table 3: Concentrations (Average ± sd, n = 3, 95% confidence level) for fish samples obtained by ICP-OES (dry weight) expressed in mg/Kg

Scientific name	Common name	Cd	Cu	Fe
Scomber Scombrus	Mackerel	0.105 ± 0.01	12.06 ± 0.06	60.60 ± 0.02
Sardinella sindesis	Sardine	0. 150 ± 0.03	4.89 ± 0.01	45.06 ± 0.05
Gadus mangala	Stock fish (cod)	ND	3.11 ± 0.02	39.62 ± 0.05

Potential human health risk assessment

The exposure doses, hazard quotient and the total hazard indices of the toxic metals under study are shown in Table 4.

Table 4: Exposure doses (x 10^{-3}) mg/kg/day; Hazard quotient (HQ) and total hazard indices (THI) of the toxicants (Cd, Cu and Fe)

Fish species	Exposure dose	Cd	Cu	Fe	THI
Scomber Scombrus	Child	0.080	9..195	46.208	
	HQ child	0.080	0.230	0.066	0.376
	Adult	0.055	6.306	31.685	
	HQ adult	0.055	0.158	0.045	0.258
Sardenlla Syndesis	Child	0.114	3.729	34.358	
	HQ child	0.114	0.093	0.049	0.256
	Adult	0.078	2.557	23.560	
	HQ adult	0.078	0.064	0.034	0.716
Gadus mangala	Child	ND	2.371	30.210	
	HQ child		0.059	0.043	0.102
	Adult		1.626	20.716	
	HQ adult		0.04	0.030	0.071

The LOD of the assayed metals ranged from 0.23 μgg^{-1} for Cd to 0.35 μgg^{-1} for Fe. The LOQ varied from 0.69 μgg^{-1} for Cd to 1.08 μgg^{-1} for Fe. The results obtained from analysis of the reference material were within the range of certified value with 81-98% recovery and procedural replications showed RSD less than 10% (n = 5) for all the metals determined.

Analysis of variance (P = 0.05) indicates a significant difference in the mean concentration of Fe when compound with other metals found in the fish species. The pattern of metal content in the fish species is Fe > Cu > Cd.

Cd was detected in some of the fish species with a range from 0.001 ± 0.00 to 0.150 ± 0.03 mg/Kg, which is the lowest among other metals examined. The concentration of Cd found in the fish samples were well below the permissible limit of 2.0 mg/Kg recommended by Food and Agricultural Organization (FAO)[11] and USEPA[12]. Acute exposure to Cd can results in chronic obstructive lung disease, renal diseases and fragile bones among others[13]. The concentrations of Cu in the study ranged from 1.050 ± 0.03 mg/Kg to 12.06 ± 0.06 mg/Kg, which is far below the permissible limit of 30 mg/Kg[12]. Cu is an essential element in animals but has emetic action and is also toxic to man and other animals Fe was found in high concentration in all the fish samples with an average value of 48.37 ± 0.35 mg/Kg, which could be attributed to its bioavailability in the environment and its essential role in haemoglobin. The elements Cu and Fe are valuable to human beings as nutrients for healthy living but if present in levels above certain limits, they constitute a potential hazard to health when consumed[14].

Many studies have reported lower values of these metals in some fish species and samples[15-17]. Elevated levels and differences in heavy metal distribution in fish species might be attributed to their differences in feeding habits, habitats, ecological needs, metabolism, biology and physiology[18].

The daily exposure (D) of Cd in studied *Scomber Scombrus wa*s 0.0013 mg per day for a child of 16 yrs and 0.0056 mg per day for a 70 yr old Nigerian. The values are lower than the tolerated daily intake of 0.07 mg per day for an adult[19]. Also the daily exposures of Cu and Fe from *scomber scombrus* consumption were 0.140 mg per day and 0.739 mg per day equivalent to 22.99% and 6.60% of RFDs of Cu and Fe respectively for a child. For an adult of average weight of 70 Kg, the daily exposures of Cu and Fe from *scomber scombrus* consumption were 0.441 mg per day and 2.22 mg/day equivalent to 15.77% and 4.53% of their respective RFDs[19]. These exposures are lower than the tolerable daily intake of the metals in fish[19]. Furthermore, the hazard quotients of the metals (Cd, Cu and Fe) from *scomber scombrus* were lower than one (HQ < 1) which indicated no risk status for consumption of the frozen fish in Nigeria.

The intake of the studied metals from consumption of *sardinella syndesis* were 0.0182 mg/day of Cd, 0.060 mg/day of Cu and 0.550 mg/day of Fe for a 16 Kg child and 0.0055 mg/day of Cd, 0.179 mg/day of Cu and 1.64 mg/day of Fe for a 70 Kg adult. The daily exposures of the metals were lower than daily tolerable intake or daily ref recommended by USEPA-IRIS[19]. The hazard quotients of the metals were all less than one (HQ < 1) indicating no hazard or risk from consumption of imported frozen *sardinella sindesis*.

Similarly, the intake of the metals from consumption of *Gadus mangala* were 0.038 mg/day of Cu and 0.483 mg/day of Fe for a 16 Kg child while an adult of 70 Kg ingest 0.114 mg/day of Cu and 1.450 mg/day of Fe from *Gadun mangala* consumptions. The hazard quotients (HQ) of these metals consumption were of no risk level having values less than one. Cd was below detection level in *Gadus mangala.*

The total hazard indices of the metals ingestion in all the fish species were under the classification of no risk level (THI < 1) of recommended by USEPA-IRIS[10]. Hazard quotient based risk assessment method does not promote a quantitative estimate for the probability of an exposed population experiencing a reverse health effect but rather provides an indication of the risk level due to exposure to pollutants[19].

The result of this study was in contradiction with the work by Udo and co-worker, who found the levels of Cu and Fe very high in mackerel, *scomber scombrus* imported into Nigeria, which lead to their conclusion that the fish species was no longer suitable for human consumption, especially with respect to Cu and its carcinogenic nature in human system[4].

REFERENCES

1. R. B. Clark, Marine Pollution. Charendo Press, Oxford, UK (1992).
2. J. Arshad, J. Muhammad and A. Sajid, Int. J. Agric. & Biol., **9(1)**, 139-142 (2007).
3. S. A. Adewuyi, B. B. Philip and I. A. Ayinde et al., J. Hum. Ecol., **31(3)**, 178-184 (2010).
4. P. J. Udo and V. N Arazu, Pakistan J. Nutri., **10(12)**, 1158-1162 (2011).
5. FAO/SIDA. Manual of Methods in Aquatic Environmental Research, Part 8: Analysis of Metals and Organochlorines in Fish, FAO Fisheries Technical Paper, 156-212 (1983).

6. Federal Registers, Analytical Procedures and Methods Validation, Chemistry, Manufacturing and Controls (Notices) **65**, 776-777 (2000).

7. Agency for Toxic Substances and Disease Registry (ATSDR 2005), Public Health Assessment Guideline Manual Available at http://www.epa.Gov/iris/indes hmtl. (Accessed: March 10, 2012).

8. US EPA (IRIS, 2010), Integrated Risk Information System Available at http://www.epa. gov/iris/index. html (Accessed: March 10, 2012).

9. I. Grzetic and R. H. A Gharian, J. Serb. Chem. Soc. (2008), **73**, 35-40 (1983).

10. H. Y. Lai, Z. Y. Hseu, T. C. Chen, B. C. Chen, H. Y. Guo and Z. S. Chen. Int., J. Environ. Res. Public Health, **10**, 3595-3614 (2010).

11. FAO/WHO, Food and Agricultural Organization/World Health Organization, Evaluation of Certain Food Additives and Contaminations Hg, Pb and Cd, 16th Report, Rome, **84** (1983).

12. USEPA, Guidance for Assessing Chemical Contaminants, Data for use in Fish Advisories, Vol. 1, Fish Sampling and Analysis 3rd Edition EPA 823-R-95-007, Office of Water Washington DC. (2000).

13. ASTDR Report, Case Studies in Environmental Medicine, Heavy Metal Toxicity, US Department of the Health and Human Service, 2-22 (2001).

14. F. E. T Asuquo, I. Ewa-Oboho, E. F Asuquo and P. J. Udo, The Environ. (2004), **24**, 29-36 (2001).

15. S. M. Al-Weher, Jordan J. Bio. Sci., **9(1)**, 41-46 (2008).

16. J. C. Akpan, F. I. Abdulrahmin and O. A. Soddipo et al., J. Appl. Sci. Environ. Sanit., **4.2**, 103-114 (2009).

17. E. N. Ahmed, Egyp. J. Aquatic Biol. and Fish., **7**, 139-154 (2003).

18. G. Ewa-Szarek, A. Anthoni and G. Robert, Int. J. hydrol., **35(4)** 31-352 (2006).

19. N. S. Charry, C. T. Tamala and D. D. S. Rai, Ecotoxicol. Environ. Saf., **69**, 513-52 (2008).

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