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Dietary Intake, Carcinogenic and Non-Carcinogenic Risk Potentials of Lead, Cadmium, Mercury and Arsenic Exposure via Consumption of Dried Crayfish in Calabar, Nigeria

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Abstract

Intense pressure from both onshore and offshore oil exploration and exploitation activities, together with the accompanying urbanization and industrialization has resulted in massive contamination of land and water resources in Niger Delta, Nigeria. Whereas crayfish is very sensitive to contaminant in the aquatic environment and constitute an important part of human diet, its quality and safety from environmental pollutant is of serious health concern. Evaluation of dietary intake, potential carcinogenic and non-carcinogenic risk of lead, cadmium, mercury and arsenic exposure via consumption of dried crayfish purchased from major markets in Calabar, Nigeria was carried out between June and August 2021. Thirty-six composite samples of dried crayfish purchased from 180 vendors were used for the study. Heavy metals concentrations were determined using Atomic Absorption Spectrophotometer (Model AA-6800, Japan) after wet digestion. Metals concentrations (Mg/kg) were of the ranges 0.02 - 0.24, 0.14 - 0.86, 0.32 - 0.72, 0.04 - 0.19 for Pb, Cd, Hg and As respectively. The mean content of cadmium and mercury exceeded FAO/WHO and Commission of European Communities maximum levels for crustaceans. Average Estimated Daily Intake for each of the metals was found to be above the recommended daily intake level except for arsenic. The average estimated daily intake values for Cd and Hg were also above the tolerable upper intake level. Average Target Hazard Quotient of all the metals and Hazard

Index of all the markets were below 1.00. The Incremental Lifetime Cancer Risk of the metals was greater than the standard tolerable regulatory risk (10^{-4}) for carcinogens. Consumption of crayfish purchased from major markets in Calabar could pose a range of carcinogenic and non-carcinogenic human health risks.

Keywords

Oil Industry, Heavy Metals, Crayfish, Human Health Risk, Carcinogenic, Non-Carcinogenic

1. Introduction

The Niger Delta region of Nigeria is blessed with one of the best quality crude oils in the world and is the home of Nigeria's oil industry (Omotola, 2006; Odulari, 2008). With an average daily production of 2.07 million barrels per day (mbpd) in the first quarter of 2020 (NBS, 2017) which accounts for over 90% of Nigeria's foreign exchange earnings (NBS, 2017), this oil is the main stay of Nigeria's economy. Intense pressure from both onshore and off shore oil exploration and exploitation activities is continually degrading the quality of the Niger Delta environment. These, together with the rapid urbanization and industrialization have been linked to large scale air, water and land pollution, poor crop yield, fish migrations and contamination of land and water resources (Ayuba, 2012). Oil spills routinely occur in the Niger Delta with an estimated 3.1 million barrels of crude oil reported to have been spilled between 1976 and 2014 in the region (Chinedu and Chukwuemeka, 2018; Ubiogoro & Adeyemo, 2017). The aquatic systems are the ultimate repositories of the contaminants given that over 90% of crude oil in Nigeria is drilled offshore. Over 50% of the Delta is aquatic with extensive network of rivers, tributaries, creeks and estuaries. The Niger Delta aquatic system supports a rich mangrove swamp ecosystem with extensive mud flats and swamps which also act as important short- or long-term sink for contaminants (Adekola & Mitchell, 2011; US-NOAA, 2017). Tidal regimes in the Niger Delta play a dominant role in the redistribution of this pollutant across the length and breadth of the network of adjourning rivers, creeks and estuaries.

The accumulation of toxic heavy metals to hazardous levels in the aquatic ecosystem has become a problem of global concern as it not only disrupts the aquatic ecosystem but also poses serious threat to public health (Ubiogoro & Adeyemo, 2017). Heavy metals are not biodegradable and easily accumulate in living organisms including fish. Consequently, human beings are potentially exposed through the food chain. Most of the metals have significant potentials to become increasingly more concentrated at successively higher trophic levels and thus posing more risk.

Studies have shown that invertebrates such as crayfish bioaccumulate more pollutants including toxic heavy metals such as nickel and mercury in their muscles and exoskeleton, and cadmium, zinc, lead and mercury in their hepatopancreas (Kouba et al., 2010; Rajeshkumar & Li, 2018). Within species, bioaccumulation factor (BAF) differs for different pollutants. BAF also varies among species for the same pollutant. Site specific environmental conditions can also affect the bioaccumulation factor significantly. The quantification of the concentration of toxic chemicals in tissues of organisms is the basis of biomonitoring. Crayfish in particular have been reported to be very sensitive to contamination in the aquatic ecosystem. With the high degree sensitivity, the organisms are highly responsive to changes in the ecosystem and have been used as bioindicators in the aquatic environment (Malinovska et al., 2020; Alcorlo et al., 2006). They play a significant role in food chain as they feed on smaller creatures/invertebrates and dead plants and as well serve as food for fish and mammals. Crayfish is therefore a keystone species as they play a significant role in determining the overall community structure within the aquatic ecosystem (Malinovska et al., 2020).

Fish and meat/milk from cattle, goats and sheep are the most common sources of animal protein in Nigeria. Adekunmi et al. (2017) reported that the most consumed animal protein sources in Osun State, Southern Nigeria were; fish 41.0% and milk 42.0% for breakfast while for dinner, beef 62.2% and fish 45.1%. With the rising insecurity occasioned by Boko Haram in the north east, banditry in the north west, famers headers classes in north central, Militancy in south-south, secession agitation in south east and south western Nigeria and general inflation, the price of animal protein has been on the increase. In the southern part of the country for instance, fish and fishery products now constitute more than 60% of the total protein intake for adults especially in the rural areas (Okelola et al., 2019). Rural population in Nigeria was reported to be 48.04% of the total population in 2020 according to world bank c ollection of development indicators (Trading Economics, 2021), and the price for fish has almost become impossible for people with low purchasing power to afford. To meet their daily protein requirement, people with average income and below, have settled for cheap and readily available source of protein such as crayfish and periwinkle. This has caused a high demand of crayfish among the populace with supply not meeting demand.

In west Africa, what is known as crayfish is technically shrimps/prawns and comprise of a mixture of matured shrimps, post larvae stages of shrimps and other tiny crustaceans often harvested in estuaries and coastal waters (Okelola et al., 2019; Kainga and Kingdom, 2012). Crayfish is a good source of high quality and easily digestible protein with low fat. It also contains all the nine essential amino acids and essential fatty acids such as omega 3 and omega 6 in addition to minerals (sulphur, phosphorus, calcium and iron) and vitamins (fat soluble vitamin A, D, E and K, and water soluble vitamins C and B complex) (Simonyan, 2016). The α-amino acid (lysine) present in crayfish is used in the biosynthesis of protein. The high polyunsaturated fatty acids present in crayfish are important in lowering blood cholesterol levels. Other medicinal values of crayfish include its use in the reduction of heart related problems and goiter (Etim et al., 2020). The American Heart Association recommends individuals eat fish at least twice a week in order to meet the recommended daily intake of omega-3 fatty acid.

Crayfish is reported as the second largest fishery in the marine/estuarine fisheries in the lower Cross River Basin (Etim et al., 2020). It is estimated that about 12,000 metric tons of crayfish is produced annually in Nigeria and reported to generate about 20 million US dollars annually to the Nigerian economy (Etim et al., 2020; Ele & Nkang, 2014). Crayfish production and marketing is a profitable business. Kainga and Kingdom (2012) reported 1.64 Naira return on every Naira invested in Yenogoa metroplolis, Bayelsa State, Nigeria. A return on investment of 16816.42 Naira and 35734.50 Naira for Akwa Ibom and Abia state respectively have also been reported by Simonyan (2016) for an average of 8 and 10 bags of crayfish equivalent to 2102.05 Naira and 3573.45 Naira per bag. Average crayfish whole sale and retail margin of 18.95% and 18.98% for Nsidung, 29.82% and 28.87% for Watt, and 35.31% and 32.48% for Ikaika Oqua markets with profit of N3000, N2600 and N3500 for Nsidung, Watt and Ikaika Oqua market respectively have been reported for Calabar (Ele & Nkang, 2014). Crayfish fishery has provided business and economic activity for many Nigerian riverine Delta region fisherfolks who source their livelihood from production and marketing of smoke-dry crayfish (Okeke & Nwankwo, 2020). Akwa Ibom, Cross River, Rivers and Bayelsa States are the major producers and highest consumers of crayfish in Nigeria. Due to their relative size, crayfish is not eaten as table shrimps, rather they are smoked or sundried, ground and used as seasoning. It is an indispensable food item in the diet of the people of southern Nigeria.

Due to their feeding habits and lifestyle of burrowing and molting, crayfish are known to be accumulators of both organic and inorganic contaminants. Crayfish of all species feeds on plants and animals (living, decaying and detritus). Two major ways through which heavy metals enter into the aquatic food chain are through the digestive track (ingestion of food and water) and non-dietary routes (across the permeable membranes such as muscles and gills) (Lake et al., 2018). The presence of these toxic metals in crayfish can invalidate its nutritional, medicinal and economic benefits (Bawuro et al., 2018). Monitoring the levels of these chemical substances in crayfish and other food fishes is essential for the evaluation of the quality of the aquatic environment, given that the levels of the chemicals in fish usually reflect the levels found in the sediment and water of the environment from which they are sourced and most importantly to safeguard human health (Okeke et al., 2016). Periodic monitoring could serve as early warning signal because toxic effects in the exposed persons occur only when the metabolic, excretory, sequestration and detoxification mechanisms are no longer able to counter uptake (Lake et al., 2018). Bioaccumulation of these highly persistent and non-biodegradable toxic substances takes place when the rate of intake exceeds the rate of elimination. Serious threats like renal failure, liver damage, cardiovascular diseases and even death have been identified as some of the deleterious effects of heavy metals to human health (Lake et al., 2018; Bawuro et al., 2018).

In view of the wide consumption of crayfish harvested from areas with a long history of environmental pollution resulting from oil spills and improper waste management, it becomes imperative to evaluate dietary intake and hazard potentials of selected toxic heavy metals exposure via consumption of crayfish sourced from the Niger Delta.

2. Materials and Methods

2.1. Description of Study Area

Calabar, the capital of Cross River State, Nigeria lies between longitude 8˚15'E and 8˚26'E and between latitude 04˚55'N and 4˚58'N. Administratively, the metropolitan city is made up of two Local Government Areas (Calabar Municipality and Calabar South) (Udiba et al., 2020). It has an area of 406 square kilometers (Udofia et al., 2016) and a population of 579,000 as at 2020 (PC, 2020). The city is drained by two major rivers—the Great Kwa River with its creeks and tributaries on the east and the Calabar River also with its creeks and tributaries on the west. The two rivers originate from Oban Hills and flow in uni-direction through equatorial rain forest changing from fresh water ecology to mangrove swamp ecology before discharging into Cross River estuary which subsequently empties into the Gulf of Guinea (Atlantic Ocean), as shown in **Figure 1**. Calabar enjoys a tropical climate (Udiba et al., 2020). The Cross River Estuary and its systems are rich in fish and shell fish including Shrimps. Artisanal shrimp fishery in the Estuary is based on *Nematopalaemon hastatus.* Mean catch composition by weight of 70% *Nematopalaemon hastatus*, 14% *Parapaeniopsis atlantica*, 3% *Eshippolysmata hastatoides* and 13% bycatch has been reported (Ofor & Kunzel, 2013). Three species (*Nematopalaemon hastatus*, *Parapaeniopsis atlantica Eshippolysmata hastatoides*) therefore dominate the catch of crayfish in the region. Efiat, Bakasi and Oron are major landing areas for fish and fishery product within the Estuary (Ofor & Kunzel, 2013). Calabar is a metropolitan city with; Watt market, Marian market, 8miles market, and Mbukpa market as the major markets (Etim et al., 2020). Nsidung Beach market is the landing point for Crayfish in Calabar.

2.2. Sample Collection

Crayfish samples used for this study was purchased from the four major markets (8 miles market, Marian market, Watt market and Mbukpa market) in Calabar Metropolis. Three transects (Market lanes) in fish and fishery products section of the different markets were selected for the study. One cup of crayfish was purchased from each of five randomly selected vendors on each of the crayfish

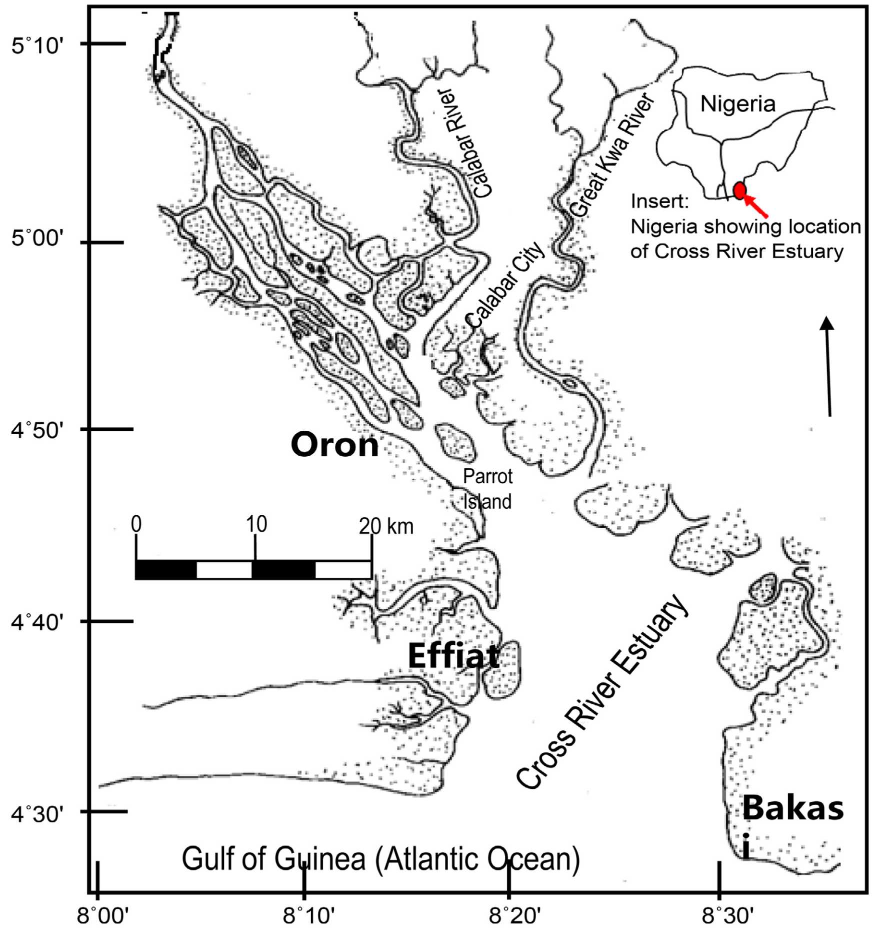

Figure 1. Map of cross river estuary showing major crayfish landing points.

lanes selected (every 6th vendor on a line of between 30 and 35 vendors). The crayfish samples from each lane were pooled together to form a composite sample, designated sampling point 1, 2 and 3 for line 1, 2 and 3 respectively, packaged in precleaned polyethylene bags and transported to Zoology and Environmental Biology laboratory, University of Calabar for sample preparation. A total of twelve composite samples was obtained per month. Sampling was conducted once a month for three months (from June to August, 2021), bringing the total number of samples to thirty-six (purchased from 180 vendors).

2.3. Sample Preparation

The dry crayfish from each market was oven dried at 40˚C for about two hours (until it creeps) before pounding into powder. The crayfish powder was thoroughly mixed and 5 g weighed into a conical flask and, digested with nitric acid and perchloric acid in ratios 3:1 on a hot plate. The digest was filtered into 50 mL volumetric flask and made up to the mark with distilled deionized water.

2.4. Sample Analysis

Lead, cadmium, mercury and arsenic concentrations in the digest were determined using Atomic Absorption Spectrophotometer (model AA-6800, Shemadzu, Japan) at National Research Institute for Chemical Technology (NARICT), Zaria, Nigeria. The calibration curve was prepared by running different concentrations of the standard solutions.

2.5. Analytical Quality Assurance

To establish the correctness and genuineness of results obtained, suitable precautions and quality assurance procedures were adopted. Samples were handled with care to avoid cross contamination. All the glassware used were cleaned in a proper manner. Double distilled water and analytical grade reagents [perchloric acid (British Drug House, England) and nitric acid (Rieldel-deHaen, Germany)] were used all through sample preservation, preparation and metal analysis. In order to evaluate the trustworthiness of the analytical method adopted for metal determination, a blank and combined standards were run with each batch of samples to detect background contamination and also monitor consistency between batches. Result of the analysis was validated by digesting and analyzing standard reference materials (animal blood coded IAEA-A-13) following the same procedure. Subsequently, the certified reference values and the analyzed values of the metals were compared to establish with certainty, the reliability of the method.

2.6. Statistical Analysis

Shapiro Wilks test was used to test for normality and Z score was used to test for outliers. Data were subjected to measures of central tendencies (mean and standard deviation) and statistical test of significance. Variation of metal contents of crayfish between the four markets on one hand and between the three sampling months on the other hand was evaluated using analysis of variance test. Probabilities less than or equal to 0.05 were considered statistically significant. Duncan multiple tests was employed for multiple comparison when equality of variance was assumed (homogeneity of variance greater than 0.05) while Donnette T was adopted for equality of variance not assumed (homogeneity of variance less than 0.05). SPSS software 23.00 for windows was used for the statistical analysis.

2.7. Evaluation of Dietary Intake and Hazard Potentials

The United States Environmental Protection Agency Recommended health risk assessment model (US-EPA, 1989) was adopted for the estimation of dietary intake, non-carcinogenic and carcinogenic hazard potential of lead, cadmium, mercury and arsenic exposure via consumption of dried crayfish. Health risk assessment essentially involves evaluation of hazard or toxicity of an agent and exposure to the agent (Risk = Hazard x Exposure) (Boguski, 2021). While hazard describes the potential of a chemical agent to cause harm, risk describes the likelihood or probability of the chemical agent to cause harm under defined conditions. Evaluation of health risk in this study was based on the assumption that ingested metals from crayfish is equal to absorbed concentration by the individual and that cooking process has no effect on the heavy metal content of the crayfish.

2.7.1. Non-Carcinogenic Health Risk Assessment

The non-carcinogenic health risk of the metals was assessed first by estimating level of exposure. The Dietary Intake (level of exposure) was evaluated using Estimated Daily Intake (EDI). Then the systematic toxicity or non-carcinogenic risk for single element was evaluated as Target Hazard Quotient (THQ) and the potential non carcinogenic risk due to more than one element as Hazard Index (HI).

1) *Estimated Daily Intake* (*EDI*)

The Estimated Daily Intake (EDI) of lead, cadmium, mercury and arsenic was determined according to Addo et al. (2013) using Equation (1)

$$\mathrm{EDI} = \frac{\mathrm{EF} \times \mathrm{ED} \times \mathrm{FIR} \times \mathrm{Cm}}{\mathrm{BAW} \times \mathrm{AT}} \quad (1)$$

where EF is the exposure frequency (365 days/year), ED is exposure duration (was adopted from UN-WPP (2022) as 55.44 years equivalent to the life time expectancy for Nigeria), FIR is the fish ingestion rate (The FIR for Nigerians of 0.02 kg/person/day was adopted from Oguguah et al. (2017) and used for edible tissues of Crayfish, Cm is the concentration of metal in Crayfish (Mg/kg), WAB is the average body weight for adult (60.7 kg) and AT is the average exposure time-age (EF × ED).

The fish ingestion rate (0.02 kg/person/day) apply to fresh fish, the concentration of metals measured in this study refereeing to dry weights were recalculated to fresh weight based on the available information on the mean moisture content of Crayfish from the area to ensure consistency between the unit used for fish ingestion rate and measured concentration data. This was done following the U.S. Environmental Protection Agency (U.S. EPA), Office of Research and Development (ORD), National Centre for Environmental Assessment's guidance and risk assessments for intake of fish and shell fish (US-EPA, 2011). The conversion of metal concentrations measured in dry weight to wet weight was done using moisture content percentage of 9.54 (Iwar & Amu, 2021) according to Equation (2) (US-EPA, 2011).

$$\mathrm{Cww} = \mathrm{Cdw}\left[\frac{100 - \mathrm{W}}{100}\right] \quad (2)$$

where: Cww = wet weight concentration, Cdw = dry weight concentration and W = Average moisture content of crayfish in Nigeria (adopted from Iwar & Amu, 2021 as 9.54%).

2) *Target Hazard Quotient* (*THQ*)

Estimation of potential hazard to human health (Target Hazard Quotient-THQ) was computed using Equation (3).

$$THQ = \frac{EF \times ED \times FIR \times Cm}{RfD \times WAB \times AT} \quad (3)$$

where RfD is the oral reference dose for metal (Mg/kg body weight per day). RfD is an estimate of daily oral exposure for the human population which does not cause harmful or damaging effect during lifetime (Guerra et al., 2012). The methodology for estimation of target hazard quotients (THQ) was adopted from *USEPA Region screening levels* (*RSLs*)—*Generic table*, 2020 (US-EPA, 2020). The value of RfD for Pb (0.0035 Mg/kg per day) was taken from WHO (2008) and ATSDR (2019). The RfD values for Cd (0.001 Mg/kg per day), Hg (0.0003 Mg/kg per day) and As (0.0003 Mg/kg per day) were taken from integrated risk information system (US-EPA, 2010).

3) *Hazard Index* (*HI*)

The hazard index was computed as the sum of the Target Hazard Quotients of the heavy metals under study as described in Equation (4) (Guerra et al., 2012).

$$HI = \Sigma THQ = THQ_{Pb} + THQ_{Cd} + THQ_{Hg} + THQ_{As} \quad (4)$$

where ΣTHQ is the summation of target hazard quotients of all metals under study, THQ_{Pb} is the target hazard quotients for lead, THQ_{Cd} is the target hazard quotients for cadmium, THQ_{Hg} is the target hazard quotients for mercury and THQ_{As} is the target hazard quotients for arsenic.

2.7.2. Carcinogenic Health Risk Assessment

Carcinogenic risk was evaluated as the incremental likelihood of a person developing cancer disease due exposure to carcinogenic or potential carcinogenic metal using Incremental Life Time Cancer Risk (ILCR). Cumulative Cancer Risk (CCR) was used to assess carcinogenic risk due to exposure to more than one carcinogenic metal.

1) *Incremental Lifetime Cancer Risk* (*ILCR*)

Incremental cancer risk due to exposure to a given cancer causing metal through the consumption of crayfish was computed following Abba et al. (2020) using Equation (5)

$$ILCR_m = EDI_m \times CSF_m\text{-oral} \quad (5)$$

where EDI_m is the estimated daily intake for the metal and CSF_m is the cancer slope factor-oral for the metal.

2) *Cumulative Cancer Risk* (*CCR*)

The cumulative cancer risk (CCR) due to exposure to many cancer causing metals from human intake of crayfish is believed to be the total of a person metal incremental lifetime cancer risk as suggested by Liu et al. (2013) and was computed using Equation (6).

$$CCR = \sum ILCR = ILCR_{Pb} + ILCR_{Cd} + ILCR_{Hg} + ILCR_{As} \quad (6)$$

3. Results

3.1. Analytical Quality Assurance

Results of the analysis of standard reference materials (coded animal blood IAEA-

A-13) employed to evaluated the accuracy and precision of the analytical procedure adopted in the study shows that the analyzed values of the metals were very close to the certified reference values (Table 1) suggesting the reliability of the method.

3.2. Total Heavy Metal Content of Crayfish Obtained from Major Markets in Calabar

Results obtained from the determination of lead, cadmium, mercury and arsenic contents of crayfish obtained from major markets in Calabar are presented in Table 2. Comparison of metal concentrations in crayfish across major markets in Calabar for the months under study are presented in Figures 2-5.

Table 1. Results of analysis of reference material (animal blood IAEA-A-13) compared to the certified reference value (Mg/kg).

Element (mg/kg)	Pb	Ni	Hg	As	Zn
A Value	0.20	1.20	4.00	1.20	14.2
R value	0.18	1.00	3.92	1.17	13.7

A Value = Analyzed value; R Value = Reference value.

Table 2. Metal concentrations (Mg/kg, dw) of crayfish purchased from major markets in Calabar metropolis, Nigeria.

Months (2021)	Sampling point	8 Miles Market				Marian Market				Watts Market				Mbukpa Market			
		Pb	Cd	Hg	As	Pb	Cd	Hg	As	Pb	Cd	Hg	As	Pb	Cd	Hg	As
June	1	0.02	0.43	0.59	0.09	0.06	0.34	0.65	0.08	0.04	0.37	0.49	0.07	0.03	0.18	0.32	0.09
	2	0.03	0.21	0.36	0.08	0.02	0.21	0.41	0.06	0.11	0.32	0.48	0.12	0.02	0.54	0.37	0.12
	3	0.14	0.42	0.38	0.06	0.03	0.25	0.36	0.12	0.03	0.14	0.42	0.15	0.06	0.34	0.52	0.18
	Mean ± SD	0.06 ± 0.07^a	0.35 ± 0.12^a	0.44 ± 0.13^a	0.08 ± 0.02^a	0.04 ± 0.02^a	0.27 ± 0.07^a	0.47 ± 0.016^a	0.10 ± 0.03^a	0.06 ± 0.04^a	0.28 ± 0.12^a	0.46 ± 0.04^a	0.11 ± 0.04^a	0.04 ± 0.02^a	0.35 ± 0.18^a	0.40 ± 0.10^a	0.13 ± 0.05^a
	Range	0.02 - 0.14	0.21 - 0.43	0.36 - 0.59	0.06 - 0.09	0.02 - 0.06	0.21 - 0.34	0.36 - 0.65	0.06 - 0.12	0.03 - 0.11	0.14 - 0.37	0.42 - 0.49	0.07 - 0.15	0.02 - 0.06	0.18 - 0.54	0.32 - 0.52	0.09 - 0.18
July	1	0.05	0.32	0.52	0.04	0.03	0.43	0.48	0.09	0.04	0.51	0.46	0.08	0.04	0.43	0.51	0.04
	2	0.05	0.43	0.53	0.11	0.05	0.19	0.47	0.09	0.03	0.27	0.43	0.18	0.06	0.38	0.32	0.18
	3	0.03	0.12	0.45	0.15	0.04	0.37	0.46	0.13	0.05	0.14	0.41	0.14	0.06	0.28	0.42	0.18
	Mean ± SD	0.04 ± 0.01^a	0.29 ± 0.16^a	0.55 ± 0.11^a	0.1 ± 0.56^a	0.04 ± 0.01^a	0.33 ± 0.12^a	0.47 ± 0.01^a	0.10 ± 0.02^a	0.04 ± 0.01^a	0.31 ± 0.19^a	0.43 ± 0.03^a	0.13 ± 0.05^a	0.05 ± 0.04^a	0.36 ± 0.08^a	0.42 ± 0.10^a	0.1 ± 0.02^a
	Range	0.03 - 0.05	0.12 - 0.43	0.45 - 0.67	0.04 - 0.15	0.03 - 0.05	0.19 - 0.43	0.46 - 0.48	0.09 - 0.13	0.03 - 0.05	0.14 - 0.51	0.41 - 0.46	0.08 - 0.18	0.04 - 0.12	0.28 - 0.43	0.32 - 0.52	0.04 - 0.18
August	1	0.15	0.56	0.57	0.12	0.03	0.54	0.65	0.14	0.22	0.45	0.72	0.14	0.13	0.49	0.43	0.16
	2	0.08	0.86	0.52	0.09	0.24	0.71	0.65	0.13	0.11	0.72	0.48	0.13	0.20	0.47	0.56	0.04
	3	0.11	0.54	0.67	0.16	0.07	0.76	0.58	0.16	0.08	0.78	0.46	0.15	0.09	0.76	0.67	0.19
	Mean ± SD	0.11 ± 0.04^a	0.65 ± 0.18^a	0.58 ± 0.08^a	0.12 ± 0.04^a	0.11 ± 0.11^a	0.67 ± 0.12^b	0.63 ± 0.04^a	0.14 ± 0.02^a	0.14 ± 0.07^a	0.65 ± 0.18^a	0.55 ± 0.03^a	0.14 ± 0.01^a	0.15 ± 0.07^b	0.57 ± 0.16^a	0.55 ± 043^a	0.13 ± 0.03^a
	Range	0.08 - 0.15	0.54 - 0.86	0.52 - 0.67	0.09 - 0.16	0.03 - 0.24	0.54 - 0.76	0.58 - 0.65	0.13 - 0.16	0.08 - 0.22	0.45 - 0.78	0.42 - 0.48	0.13 - 0.15	0.09 - 0.23	0.47 - 0.76	0.43 - 067	0.04 - 0.19

Mean with different superscript along the same column indicates significant (ANOVA, $p < 0.05$) difference in metal concentration between the months.

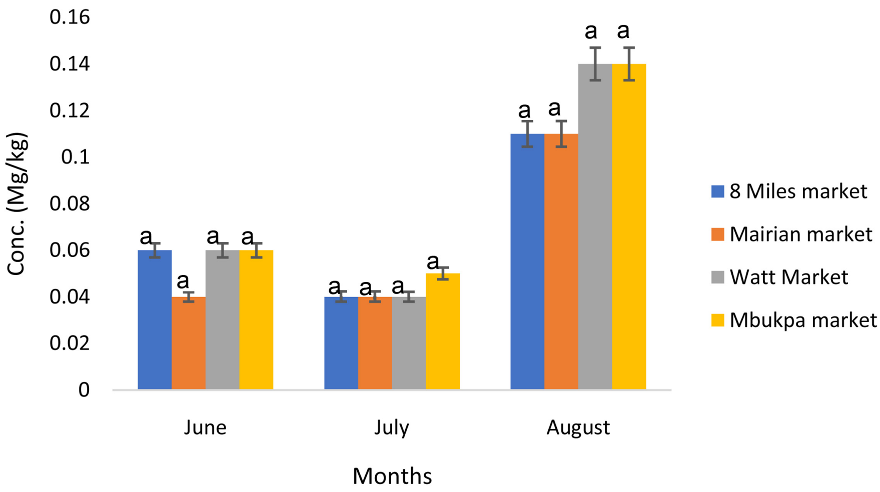

Figure 2. Comparison of mean lead concentrations of crayfish across major markets in Calabar Metropolis, Nigeria. Bars with same alphabet above the error bar within the month indicates no significant difference in lead concentration.

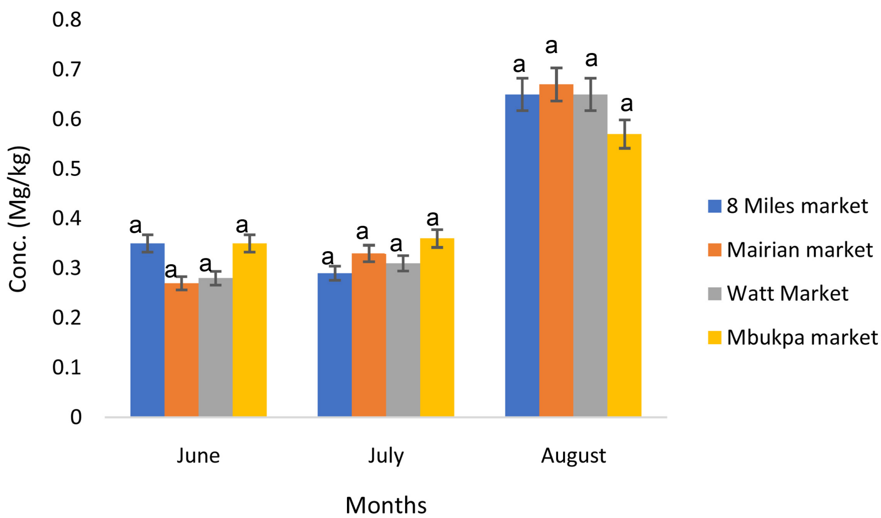

Figure 3. Comparison of mean cadmium concentrations of crayfish across Major Markets in Calabar metropolis, Nigeria. bars with same alphabet above the error bar within the month indicates no significant difference in cadmium concentration.

Concentrations of lead (Mg/kg dw) ranged from 0.02 - 0.24 (Table 2). The lowest concentration (0.02 Mg/kg) was recorded across all the markets in June, Watt market being the only exception. The highest concentration (0.24 Mg/kg) was recorded at Watt market in August. The difference in lead content of cray fish between the months was not significant (ANOVA, $p > 0.05$ in all the markets except at Mbukpa market where the lead concentration in August was significantly higher than June and July. The difference in lead concentration between June and July at the market was however not significant at 95% confidence level. The difference in lead concentration between the four markets throughout the study was not significant (ANOVA, $p > 0.05$) (Figure 2).

Table 2 indicates that cadmium content (Mg/kg dw) of crayfish in the study

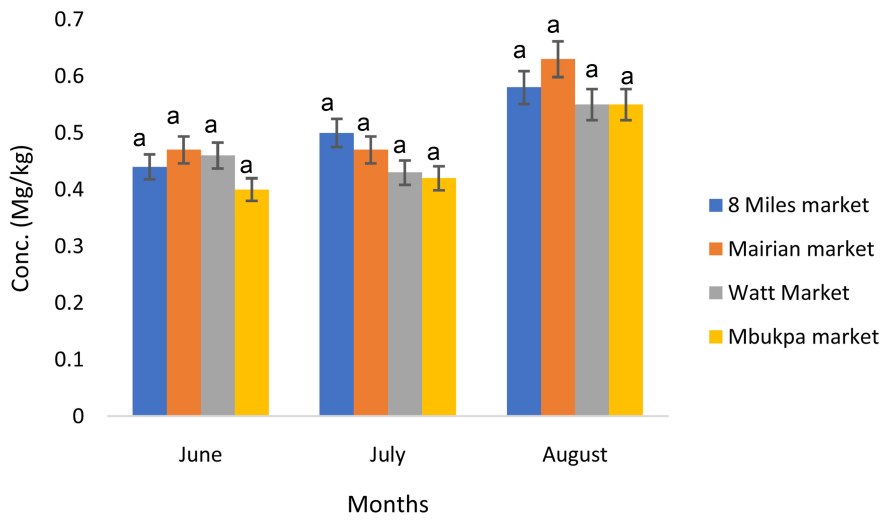

Figure 4. Comparison of mean mercury concentrations of crayfish across major markets in Calabar Metropolis, Nigeria. Bars with same alphabet above the error bar within the month indicates no significant difference in Mercury concentration.

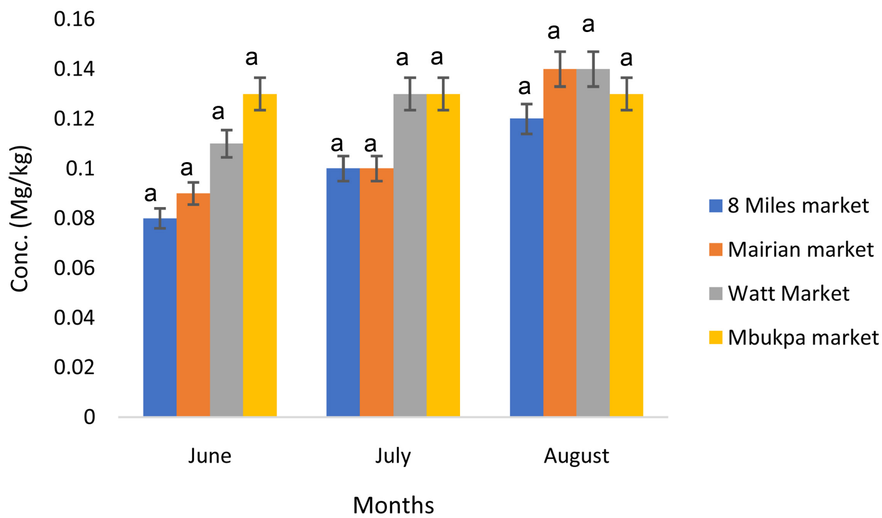

Figure 5. Comparison of mean arsenic concentrations of crayfish across major markets in Calabar Metropolis, Nigeria. Bars with same alphabet above the error bar within the month indicates no significant difference in arsenic concentration.

ranged from 0.14 - 0.86. The lowest concentration was measured at Marian Market in June and July and the highest at 8 Miles in August. The difference in cadmium concentration between the sampling months was significant (ANOVA, $p < 0.05$) only for samples purchased from Marian market where the concentration in August was found to be significantly higher than June and July. The difference in cadmium concentration between the markets studied was not significant (ANVA, $p > 0.05$) throughout the study (Figure 3).

Mercury content of crayfish in the study ranged from a minimum of 0.32 Mg/kg recorded for Mbukpa market in June and July to a maximum of 0.72 Mg/kg recorded for Watt market in August (Table 2). The difference mercury concentration between the months under consideration was not significant (ANOVA, $p > 0.05$). The difference in mercury content of crayfish between the four markets was also not significant at 95% confidence level (**Figure 4**).

Table 2 indicates that concentrations (Mg/kg) of arsenic ranged between 0.04 and 0.19. The lowest concentration was recorded at 8 Miles market in July and Mbukpa market in August. The highest value was recorded at Mbukpa market in August (**Table 2**). The difference in arsenic content of crayfish between the months was not significant (ANOVA, $p > 0.05$) all through the study. The difference in arsenic concentrations between the markets was also not significant (ANOVA, $p > 0.05$) (**Figure 5**).

3.3. Evaluation of Dietary Intake and Hazard Potentials

3.3.1. Non-Carcinogenic Health Risk Assessment

1) *Estimated Daily Intake* (*EDI*)

The average values of estimated daily intake (Mg/kg bw /day) for lead, cadmium, mercury and arsenic in the study were; 0.021, 0.028, 0.156 and 0.030 for 8 Miles market, 0.019, 0.126, 0.156 and 0.034 for Marian market, 0.024, 0.123, 0.143 and 0.039 for Watt market and 0.024, 0.128, 0.136 and 0.036 for Mbukpa market.

2) *Target Hazard Quotient* (*THQ*)

The average values of target hazard quotient for lead, cadmium, mercury and arsenic in the study were; 0.006, 0.128, 0.520 and 0.099 for 8 Miles market, 0.005, 0.126, 0.521 and 0.113 for Marian market, 0.007, 0.123, 0.477 and 0.126 for Watt market and 0.008, 0.127, 0.454 and 0.120 for Mbukpa market.

3) *Hazard Index* (*HI*)

The mean hazard index for the markets under study were 0.753 for 8 miles market, 0.765 for Marian market, 0.733 for Watt market and 0.709 for Mbukpa market.

3.3.2. Carcinogenic Health Risk Assessment

1) *Incremental Life Time Cancer Risk* (*ILCR*)

The average incremental life time cancer risk for lead, cadmium and arsenic were 1.8×10^{-6}, 4.9×10^{-2}, 4.5×10^{-2} for 8 Miles market, 1.6×10^{-6}, 4.8×10^{-2} and 5.1×10^{-2} for Marian market, 2.4×10^{-6}, 4.7×10^{-2} and 5.9×10^{-2} for Watt market and 2.0×10^{-6}, 4.8×10^{-2} and 5.4×10^{-2} for Mbukpa market.

2) *Cumulative Cancer Risk* (*CCR*)

Cumulative cancer risk for 8 Miles, Marian, Watt and Mbukpa markets were 9.5×10^{-2}, 8.9×10^{-2}, 1.1×10^{-1} and 1.0×10^{-1}.

4. Discussion

4.1. Total Heavy Metal Content of Crayfish Obtained from Major Markets in Calabar

Together with periwinkle, crayfish hold the promise of reducing protein deficiency in human diet in southern Nigeria. With the exception of carboxylates, all food nutrients are known to be present in it and is classified as a poly peptide consisting of about 36% - 45% protein (Okeke & Nwankwo, 2020). Whereas crayfish constitute an important part of human diet in the region, its quality and safety from environmental pollutants is of serious health concern. Lead, cadmium, mercury and arsenic occur naturally in the ecosystem. Their levels in the oil rich Niger Delta region of Nigeria are on the rise due to anthropogenic activities, posing threats to wild life and humans (Ayuba, 2012; Chinedu & Chukwuemeka, 2018; Ubiogoro & Adeyemo, 2017). Lead, mercury, arsenic and cadmium are non-essential elements. They have no known biological importance in living organisms and exhibits extreme toxicity even at low concentration. Though the metabolic function of arsenic is not well understood, it is suggested to be involved in the metabolism of methionine and in the regulation of gene expression (Kortei et al., 2020). To evaluate the safety of crayfish purchased from major markets in Calabar for human consumption, the concentration of these metals in edible tissues of the organism was first quantified (**Table 2**), then compared with global regulatory standards.

Lead is a highly toxic metal that has caused widespread environmental contamination and health problems around the world. Lead toxicity in living cell follows ionic mechanism and oxidative stress. The ionic mechanism is caused by its ability to replace bivalent cations such as Ca^{2+}, Mg^{2+} and Fe^{2+} and, monovalent cations like Na^{+}, which interrupts biological metabolism of the cell causing significant changes in biological processes. Oxidative stress in the cell is caused by imbalance between production of free radicals and generation of antioxidants to detoxify the reactive intermediate or to repair resulting damages (Jaishankar et al., 2014). Lead is known to interfere with the formation of hemoglobin, thus display anemia as a common symptom. Exposure to higher lead concentrations over time may produce permanent brain damage, kidney disfunction and several symptoms related to the central nervous system. Lead encephalopathy also displayed, is characterized by sleeplessness, and restlessness. Children are uniquely susceptible to lead poisoning as it interferes with the development of a child's brain and central nervous system. Children may be affected by learning and concentration deficit, behavioural disturbances, diminished intellectual capacity, reduced consciousness, coma and even death (Obasi & Akudinobi, 2020). The mean content of lead in crayfish (**Table 2**) purchased from each of the four markets was below Codex maximum level (ML) (FAO/WHO, 2015) of 0.30 Mg/kg and Commission of European Communities maximum levels for crustaceans of 0.5 Mg/kg (EC, 2015) all through the study. Codex maximum level for contaminants and toxins in food and feed is the maximum concentration of that substance recommended by the Codex Alimentarius commission to be legally permitted in that commodity. The average lead content of crayfish in the study was also below the Codex Committee on Food Additives maximum limit of 0.2 Mg/kg as well as US-EPA Health criteria for human health risk of carcinogen of

4 Mg/kg. Lead was not indicted in crayfish purchased from major markets in Calabar but the fact that there is no known concentration below which the metal appears to be safe, calls for serious concern. Lead concentrations ranging from 0.002 - 0.14 Mg/kg was reported by Waribo et al. (2019) for dried Crayfish sold in Creek Road market, Borokiri, Port Harcourt, Nigeria, is in agreement with the findings of this study. Higher concentrations ranging from 0.27 - 2.04 Mg/kg and 0.5 - 2.30 Mg/kg were reported for crayfish obtained from creeks in Rivers State, Nigeria and from Sabo market, Ile-Ife, Nigeria respectively (Elekima et al., 2020; Waribo, et al., 2019). A lower range (BDL-0.34) was reported for crayfish obtained from the great Qua River (Abraham et al., 2015). Lead concentrations ranging from 0.5 to 0.82 was reported for crayfish from selected Czech reservoirs (Kuklina et al., 2014). No significant difference lead concentration was observed between the four markets in this study suggesting a common fishing ground for the crayfish sold in the markets.

Average cadmium contents of crayfish recorded in this study (**Table 2**) was found to be below Codex maximum level (ML) (FAO/WHO, 2015) and Commission of European Communities maximum levels for crustaceans of 0.5 Mg/kg (EC, 2015) in June and July. The mean value of cadmium in August across all the markets was found to be higher than the maximum permissible limits. The fact that there was no significant difference in cadmium content between the markets as the case of lead, suggest that the crayfish may come from the same source. This finding is in corroboration with Ele and Nkang (2014). In the study of structure and efficiency of crayfish marketing in major markets in Calabar, Ele and Nkang reported that Nsidung beach, Calabar, is the major landing point for Crayfish in Cross River State and that traders from other markets purchase crayfish from Nsidung in large quantities for sale to customers within and outside the state. The source of crayfish at Nsidung beach was traced to Efiat, Bakasi and Oron which are major landing areas for fish and fishery product within the Cross River estuary. August is the peak of wet season in the Niger Delta region of Nigeria. The rainfalls are usually torrential with characteristics large surface run-off to the adjourning river and flood plans. water filtering into the estuary brings in nutrients and contaminants from the entire drainage basin (US-NOAA, 2017) and crayfish are known to be very sensitive to changing environmental conditions, hence their use as bio-monitors. This may account for the elevated cadmium concentrations observed in August. Cadmium is known to be extremely toxic. It is the seventh most toxic heavy metal in the ATSDR ranking (ATSDR, 2012). Cadmium content of crayfish in this study is a cause for serious concern because of its diverse toxic effects including teratogenicity, carcinogenicity, nephrotoxicity, endocrine and reproductive toxicity compounded by its long biological half-life (20 - 30 years in humans), low rate of excretion from the body and storage in soft tissues (Liver and Kidney) (Rani et al., 2014). Higher cadmium concentrations ranging from 0.17 - 1.94 Mg/kg and 0.4-1.40 Mg/kg were reported for crayfish obtained from creeks in Rivers State, Nigeria and from Sabo market, Ile-Ife, Nigeria respectively (Elekima et al., 2020; Waribo, et al., 2019). Cadmium concentration in crayfish obtained from the Great Qua River, Calabar, Nigeria was reported to be below detectable limits (Abraham et al., 2015). Higher cadmium concentrations ranging from 0.5 to 0.82 has also been reported for crayfish from selected Czech reservoirs (Kuklina et al., 2014).

In the aquatic environment, mercury exist mainly as metallic element, inorganic salts and organic compound, the different forms possessing different levels of bioavailability and toxicity. The three forms of mercury are easily taken up by organisms, transformed into methyl mercury within the organism and cannot be excreted easily. Biomagnification of mercury within the food chain is a well-known phenomenon (Jaishankar et al., 2014). Becoming increasingly more concentrated at successively higher trophic level makes the organism higher up in the food chain (e.g. man) more at risk. Eating contaminated fish and shellfish is the main source of methyl mercury exposure (WHO, 2007). Elemental mercury and methyl mercury are toxic to central and peripheral nervous system. Though the brain is the target organ for mercury, it can destroy any organ causing malfunctioning of nerves, kidney and muscles. It plays key roles in damaging protein structures and alter cellular functions. More still, mercury can pass from mother to fetus during pregnancy through the placenta and may affect the development of the central nervous system resulting to behavioural and cognitive impairment (Jaishankar et al., 2014). The mean content of mercury in crayfish (**Table 2**) purchased from each of the four markets was above Codex maximum level (ML) (FAO/WHO, 2015) of 0.50 Mg/kg and Commission of European Communities maximum levels for crustaceans of 0.5 Mg/kg (EC, 2015). The Joint FAO/WHO Expert committee on Food Additives (JECFA) in 2004 established a tolerable intake level of 1.6 μg/kg b.w per week for methyl mercury (which corresponds to 0.097 Mg/kg b.w for an adult of 60.7 kg considered in this study) in order to protect developing fetus from neurotoxic effects (JECFA, 2003; WHO, 2007). It follows therefore that crayfish in this study may not be safe for pregnant women as it is not protective of the developing fetus with respect to mercury poisoning. Lower mercury concentrations ranging from 0.001 - 0.004 Mg/kg and 0.06 - 0.18 Mg/kg were reported for crayfish obtained from creeks in Rivers State, Nigeria and from selected Czech reservoirs (Elekima et al., 2020; Kuklina et al., 2014). Higher mercury concentrations ranging from 4.744 - 5.136 Mg/kg was reported for crayfish obtained from Sabo market, Ile-Ife, Nigeria (Waribo et al., 2019).

Arsenic occurs in both organic and inorganic forms with the organic form being far more toxic. In human, approximately 60% - 90% of dietary inorganic arsenic is absorbed, transported to the liver, reduced to arsenite and then methylated. Majority of the ingested arsenic is rapidly excreted in the urine (Civantos et al., 1995). Average arsenic contents of crayfish recorded in this study (**Table 2**) was found to be below Codex maximum level (ML) (FAO/WHO, 2015) of 0.50 Mg/kg and Commission of European Communities maximum levels for crustaceans also of 0.5 Mg/kg (EC, 2015) through the study. Consumption of crayfish from the study area does not pose toxicological risk with respect to arsenic poisoning. Inorganic arsenic has been established as a human poison. Poisoning occurs with arsenic doses of 1 Mg/kg/day and above. Doses greater than 10 Mg/kg/day leads to gastrointestinal symptoms, encephalopathy and arsenicism (Civantos et al., 1995; NRC, 1999). Ingested inorganic arsenic is also associated with the risk of cancer (NRC, 1999). Similar concentrations of arsenic ranging from 0.01 - 0.11 were reported for crayfish obtained from creeks in Rivers State, Nigeria (Elekima et al., 2020). Higher arsenic concentration ranging from 1.428 - 8.989 Mg/kg was reported for crayfish obtained from the Great Qua River, Calabar, Nigeria (Abraham et al., 2015).

4.2. Evaluation of Dietary Intake and Hazard Potentials

4.2.1. Non-Carcinogenic Health Risk Assessment

1) *Estimated Daily Intake* (*EDI*)

Safe levels of lead, cadmium, mercury and arsenic through the consumption of crayfish purchased from major markets in Calabar was described using estimated daily intake. The EDI combines data on the concentration of the metals in crayfish and the measure of crayfish ingested daily (US-NAS, 2001). The EDI computed were compared to the Recommended Daily Intake (RDI) and the Tolerable Upper Intake Level (UL). RDI is the average daily intake of the metals that would likely meet the nutrient requirement of 97 to 98 percent healthy adults from 54 years of age upward and the tolerable upper intake level (UL) which is the highest level of daily intake which is likely to pose no risk of adverse effects for almost all individuals. The higher the EDI value above UL, the greater the chances of significant health problems (Guerra et al., 2012). Average EDI for each of the metal (**Table 3**) was found to be above the recommended daily intake level except for arsenic. The average estimated daily intake values for Pb and As were below the tolerable upper intake level indicating no risk to people's

Table 3. Estimated Daily Intake (Mg/kg b.w/day) of metals in Crayfish purchased from four major markets in Calabar metropolis.

	8 Miles Market				Marian Market				Watts Market				Mbukpa Market			
	Pb	Cd	Hg	As	Pb	Cd	Hg	As	Pb	Cd	Hg	As	Pb	Cd	Hg	As
June	0.018	0.105	0.131	0.024	0.012	0.081	0.140	0.030	0.018	0.083	0.137	0.033	0.012	0.105	0.119	0.039
July	0.012	0.086	0.164	0.030	0.012	0.098	0.140	0.030	0.012	0.092	0.128	0.039	0.015	0.108	0.125	0.030
August	0.033	0.194	0.173	0.036	0.033	0.200	0.188	0.042	0.042	0.194	0.164	0.045	0.045	0.170	0.164	0.039
Average	**0.021**	**0.128**	**0.156**	**0.030**	**0.019**	**0.126**	**0.156**	**0.034**	**0.024**	**0.123**	**0.143**	**0.039**	**0.024**	**0.128**	**0.136**	**0.036**
UL (Mg/day)	**0.240**	**0.064**	**1.6***	**1 - 3**	**0.240**	**0.064**	**1.6***	**1 - 3**	**0.240**	**0.064**	**1.6***	**1 - 3**	**0.240**	**0.064**	**1.6***	**1 - 3**
RDI (Mg/day)	**0.00**	**0.00**	**0.00**	**0.5 - 1**	**0.00**	**0.00**	**0.00**	**0.5 - 1**	**0.00**	**0.00**	**0.00**	**0.5 - 1**	**0.00**	**0.00**	**0.00**	**0.5 - 1**

* = µg/kg bw/week.

associated with intake of the metals via consumption of crayfish from the study area. On the other hand, the average EDI for cadmium and mercury were above the UL suggesting possible risk with respect to cadmium and mercury poisoning. The estimated daily metal intake computed in the study were expressed per kilogram body weight per day (Mg/kg b.w/day) so that for an average Nigerian adult of 60.7 kg^{-1} body weight considered in this study, the average EDI of say cadmium in crayfish from 8 miles, Marian, Watt and Mbukpa markets are equivalent to 0.128, 0.126, 0.123 and 0.128, respectively, which when multiplied by 60.7 gives 7.770, 7.648, 7.466 and 7.770 Mg/day, respectively. Based on the tolerable upper intake level, cadmium is seriously indicted in the study. Ideally neither children nor adult should have lead, cadmium and mercury in their bodies because it provides no physiological benefits. Consumption of crayfish from the study area poses considerable toxicological risk.

2) *Target Hazard Quotient* (*THQ*)

To evaluate the potential risk, pose by each metal, target hazard quotient which is the ratio of potential exposure to a chemical contaminant and reference oral dose was used (Guerra et al., 2012; Lanre-Iyanda & Adekunle, 2012). When the ratio is less than one (1), no risk is implied. The higher the value above unity the greater the risk. Exposure to the metals under study may occur through multiple routes. Target hazard quotient in this study considered only the exposure through consumption of crayfish from major markets in Calabar without considering other exposure routes. The average THQ of all the metals in the study were below 1.00 (**Table 4**).

3) *Hazard Index* (*HI*)

Hazard index is used to evaluate potential human health risk due to more than one contaminant (Guerra et al., 2012). It assumes that, the severity of the adverse effect of contaminants is proportional to the sum of the multiple contaminant's exposures. It also assumes target organs are linearly affected by similar working mechanism (Guerra et al., 2012). Potential health risk is implied when HI value is greater than unity. Even though there was no apparent risk when each metal was analyzed individually, the potential risk could be multiplied when all metals are considered together. The hazard index of crayfish purchased from each of

Table 4. Target hazard quotient and hazard index of metals in crayfish purchased from four major markets in Calabar metropolis.

Sampling Months	8 Miles Market					Marian Market					Watts Market					Mbukpa Market				
	Target Hazard Quotient				Hazard Index	Target Hazard Quotient				Hazard Index	Target Hazard Quotient				Hazard Index	Target Hazard Quotient				Hazard Index
	Pb	Cd	Hg	As		Pb	Cd	Hg	As		Pb	Cd	Hg	As		Pb	Cd	Hg	As	
June	0.005	0.104	0.437	0.079	**0.625**	0.003	0.080	0.467	0.100	**0.650**	0.005	0.083	0.457	0.110	**0.655**	0.005	0.104	0.398	0.130	**0.637**
July	0.003	0.086	0.546	0.100	**0.735**	0.003	0.098	0.466	0.100	**0.667**	0.003	0.092	0.427	0.130	**0.652**	0.007	0.107	0.417	0.100	**0.631**
August	0.009	0.194	0.577	0.119	**0.899**	0.009	0.200	0.629	0.139	**0.977**	0.012	0.193	0.547	0.139	**0.891**	0.013	0.170	0.546	0.130	**0.859**
Average	**0.006**	**0.128**	**0.520**	**0.099**	**0.753**	**0.005**	**0.126**	**0.521**	**0.113**	**0.765**	**0.007**	**0.123**	**0.477**	**0.126**	**0.733**	**0.008**	**0.127**	**0.454**	**0.120**	**0.709**

the four markets, for an adult of 60.7 kg body weight considered in the present study was found to be less than unity. The relative contributions to the aggregated risk posed by lead, cadmium, mercury and arsenic at each of the market was 0.80%, 17.00%, 69.06% and 13.15% for 8 Miles market, 0.65%, 16.47%, 68.10% and 14.77% for Marian market, 0.95%, 16.78%, 65.08% and 17.19% for Watt market, and 1.13%, 17.91%, 64.03% and 16.93% for Mbukpa market.

4.2.2. Carcinogenic Health Risk Assessment

1) *Incremental Lifetime Cancer Risk* (*ILCR*)

The acceptable risk level for carcinogens ranges from 10^{-4} at which the risk of developing cancer over a life time is 1 in 10,000 to 10^{-6} at which the risk of developing cancer over a life time is 1 in 1,000,000 (US-EPA, 2005). Cancer risks below 10^{-6} are considered negligible and above 10^{-4} are considered unacceptable. The cancer risks determined in this study (**Table 5**) were greater than the standard tolerable regulatory risk of the carcinogens (10^{-4}). In the International Agency for Research on Cancer (IARC) classification for carcinogenic agent, cadmium and arsenic are placed in group 1 which are definite human carcinogens, lead is in group 2B which are possible human carcinogens and mercury in group 3 which are not classified as to its carcinogenicity to humans probably because of evidence of its carcinogenicity to humans is insufficient (IARC, 2021). This implies that cadmium, arsenic and lead in crayfish poses significant carcinogenic risk.

2) *Cumulative Cancer Risk* (*CCR*)

The cumulative cancer risk due to exposure to cadmium, lead and arsenic via consumption of crayfish from each of the market under study (**Table 4**) was also found to be above the standard tolerable regulatory of risk the carcinogens (10^{-4}) indicating significant carcinogenic risk.

Table 5. Incremental Lifetime Cancer Risk (ILCR) and Cumulative Cancer Risk (CCR) of metals in Crayfish purchased from four major markets in Calabar metropolis.

Sampling Months	8 Miles Market					Marian Market					Watts Market					Mbukpa Market				
	ILCR				CCR	ILCR				CCR	ILCR				CCR	ILCR				CCR
	Pb	Cd	Hg	As		Pb	Cd	Hg	As		Pb	Cd	Hg	As		Pb	Cd	Hg	As	
June	1.8×10^{-6}	4.0×10^{-2}	-	3.6×10^{-2}	7.6×10^{-2}	1.0×10^{-6}	3.1×10^{-2}	-	4.5×10^{-2}	7.6×10^{-2}	1.5×10^{-6}	3.2×10^{-2}	-	5.0×10^{-2}	8.2×10^{-2}	1.0×10^{-2}	4.0×10^{-2}	-	5.9×10^{-2}	9.9×10^{-2}
July	1.0×10^{-6}	3.3×10^{-2}	-	4.5×10^{-2}	7.8×10^{-2}	1.0×10^{-6}	3.7×10^{-2}	-	4.5×10^{-2}	8.2×10^{-2}	1.0×10^{-6}	3.4×10^{-2}	-	5.9×10^{-2}	9.3×10^{-2}	1.3×10^{-6}	4.1×10^{-2}	-	4.5×10^{-2}	8.6×10^{-2}
August	2.8×10^{-6}	7.4×10^{-2}	-	5.4×10^{-2}	1.3×10^{-1}	2.8×10^{-6}	7.6×10^{-2}	-	6.3×10^{-2}	1.1×10^{-1}	3.6×10^{-6}	7.0×10^{-2}	-	6.8×10^{-2}	1.4×10^{-1}	3.8×10^{-6}	6.5×10^{-2}	-	5.9×10^{-2}	1.2×10^{-1}
Average	1.8×10^{-6}	4.9×10^{-2}	-	4.5×10^{-2}	9.5×10^{-2}	1.6×10^{-6}	4.8×10^{-2}	-	5.1×10^{-2}	8.9×10^{-2}	2.4×10^{-6}	4.7×10^{-2}	-	5.9×10^{-2}	1.1×10^{-1}	2.0×10^{-6}	4.8×10^{-2}	-	5.4×10^{-2}	1.0×10^{-1}

5. Conclusion

In view of the wide consumption of crayfish and the consequent increase in environmental pollution resulting from anthropogenic activities in the Niger Delta, monitoring the levels of these chemical substances in crayfish and other food fishes is essential for the evaluation of the quality of the aquatic environment given that the levels of the chemicals in fish usually reflect the levels found in the sediment and water of the environment from which they are sourced and most importantly to safeguard human health. Dietary intake and hazard potentials of lead, cadmium, mercury and arsenic exposure via consumption of crayfish from the area were evaluated. The concentrations (Mg kg^{-1}) of the metals were of the ranges 0.02 - 0.24, 0.14 - 0.86, 0.32 - 0.72, 0.04 - 0.19 for Pb, Cd, Hg and As respectively. The mean content of cadmium and mercury exceeded FAO/WHO and Commission of European Communities maximum levels (ML) for crustaceans but lead and arsenic contents were within the acceptable levels. Average EDI for each of the metal was found to be above the recommended daily intake level except for arsenic. The average estimated daily intake values for Cd and Hg were above the tolerable upper intake level (UL). The average Target Hazard Quotient (THQ) of all the metals and Hazard Index (HI) of all the markets were below 1.00. The Incremental Lifetime Cancer Risk (ILCR) of the metals was greater than the standard tolerable regulatory risk (10^{-4}) for carcinogens. Consumption of crayfish from major markets in Calabar could pose a range of carcinogenic and non-carcinogenic human health risk with respect to lead, cadmium and mercury poisoning in people eating at or above the fish ingestion rate. Periodic monitoring fish and fishery products from Niger Delta, Nigeria are here-by advocated.

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Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

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