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**Research** article

# Evaluation of human health risk assessment of potential toxic metals in commonly consumed crayfish (Palaemon hastatus) in Nigeria

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# ABSTRACT

Concentrations of potentially toxic metals were determined in crayfish (Palaemon hastatus) commonly consumed in Nigeria using Atomic Absorption Spectrophotometry. Results revealed that Fe had the highest mean value of  $18.88 \pm 0.10 \ \mu g/g$ , while Pb had the least (0.91  $\pm 0.01 \ \mu g/g$ ). Cluster analysis showed close inter-element relationships between the metals, indicating similar chemical properties and/or genetic origin. Correlation matrix indicated positive and significant correlations between Cr/Cd, Fe/Cd, Fe/Cd, Pb/Cd, Pb/Fe and Cu/Zn, establishing chemical affinity. Estimated daily intake, target hazard quotient and cancer risk showed that there was no health risk associated with the consumption of the crayfish. Relative risk showed that potential health risk could be attributed only to Cd level. The study concluded that consumption of the crayfish may not pose health risk to human health at the levels of the analysed metals, but should be consumed moderately to prevent bioaccumulation of the metals most importantly Cd.

# 1. Introduction

Crayfish is one of the healthy and available sea-foods in Nigeria and globally, and it is naturally packed with minerals, vitamins and other nutrients which are beneficial to health. Eating crayfish in moderation is the key to gaining all the essential nutrients it contains without worrying about the effects and health conditions that its high consumption might cause or trigger. The vitamins present in crayfish play an important role in the body too, as it helps in improving the general wellbeing of the body which includes the skin, eyes and more. Due to great nutrients contained in crayfish, it is indeed a great food to promote the overall body health but when taken in excess can lead to high blood pressure and cardiovascular diseases (Mehta and Hawkins, 1998).

However, pollution by toxic metals is a prevalent problem worldwide. These metals can become noxious when they exceed certain threshold concentrations. Crayfish readily accumulates toxic metals in tissues and also meet other criteria which make them suitable as bioindicators in the environment. As a result of the buildup effect of some toxic metals, especially through the food chain, there is the need to incessantly monitor their bioavailability. Through the analysis of metal concentrations in living organisms, it is possible to deduce the bioavailability and, by presumption, the level of environmental pollution by specific metals (Holdich et al., 2006).

In general, for all crayfish species, the concentration of metals in the environment is not sufficient to be a direct cause of death. Furthermore, crayfish are considered to be highly resistant to environmental metal contamination; nevertheless, some toxic metals might still get into the crayfish (Del Ramo et al., 1987; Roldan and Shivers, 1987; Chambers, 1995). The accumulation of metals in their tissues is dose- and time-dependent, and therefore may be reflective of the levels of metals in the environment (Antón et al., 2000; Rowe et al., 2001; Sánchez-López et al., 2004; Alcorlo et al., 2006; Schmitt et al., 2006; Allert et al., 2009).

The exposure of humans to the elevated levels of toxic metals present in crayfish might cause serious health risks to the consumers; hence, there is need to assess the elemental composition of the crayfish, its associated health risk as well as its potential impact on the environment.

# 2. Materials and methods

# 2.1. Sample collection and preparation

Ten (10) crayfish samples were collected from Sabo market in Ile-Ife, Osun State, Nigeria. To avoid contamination during sampling, transportation and storage, the crayfish samples were kept in polyethylene bags and taken immediately to the laboratory for analysis. The crayfish samples were pulverized to aid digestion using agate mortar. Each 0.5 g

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of the ground samples was digested in a 125 mL Teflon bomb with 3 mL of concentrated 68 % HNO<sub>3</sub> (BDH Laboratory Supplies Poole Bh 15 1 TD, England) and 3 mL 50 %  $H_2O_2$  (CDH (P) Ltd, India). The digestion was carried out using a thermostat hotplate at 70 °C under a fume hood. The reagents used are Analar grade reagents. After digestion, the bomb was allowed to cool to room temperature for a few hours. The digested sample was poured into a 50 mL volumetric flask, and double-distilled water was used to rinse the Teflon bomb into the volumetric flask; the flask was filled to the mark with double-distilled water (Ogner et al., 1991).

# 2.2. Elemental analysis

The elemental analysis of the crayfish samples was performed using Atomic Absorption Spectrophotometer (AAS) Model Alpha Star 4 (ChemTech Analytical) at the Centre for Energy Research and Development (CERD), Obafemi Awolowo University, Ile-Ife, Nigeria. The instrument was operated in accordance with the instrument's handbook while calibration was done using a mixed calibration standard solution prepared from the pure British Drug House (BDH) Analar grade salt of each metal.

#### 2.3. Quality assurance

The accuracy of the procedure employed was assured by spiking two 0.5g portions of each of the crayfish samples for recovery analysis. One portion was spiked with 5mL of 5.0  $\mu$ g/g standard mixtures of the heavy metal solution while the other portion which serves as control was left unspiked. Both the spiked and unspiked portions were separately digested. The percentage recovery (%) of the heavy metals was determined by comparing the concentrated values of each metal from spiked and unspiked samples using the expression:

$$[\mathbf{A} - \mathbf{A}']/\mathbf{B} \times 100 \tag{1}$$

Where

A = Heavy metal concentration in spiked crayfish samples

A' = Heavy metal concentration in unspiked crayfish samples and B = the amount of heavy metal used for spiking (Oyewole and Adebiyi, 2017).

#### 2.4. Data treatment

For the interpretation of the data, the following statistical methods were used: Descriptive statistics (mean, range, standard deviation) were performed in addition to pollution index to ascertain the contamination of the samples. Cluster analysis was carried out to determine the interelement clustering relationship. To assess the association between the analyzed metals, correlations of the heavy metals were determined between the different matrices. Human health risk assessment such as estimated daily intake, target hazard quotient, cancer risk and relative risk were carried out to determine the health risks associated with exposure to the samples.

# 2.4.1. Pollution index (PI)

The elemental concentrations of the crayfish samples were subjected to statistical analysis to determine the PI of the elements. The PI is the quotient of the concentration of the element x in the sample to the maximum permissible level of the element.

$$PI(X) = \frac{\text{Metal concentration in the sample}}{\text{Permissible limit or background value}}$$
(2)

It is agreed in principle that if the value of PI of an element is greater than 1.0, it implies that the contamination of the sample by the element is

high and may be toxic at the level it is present in the sample (Fasasi and Obiajunwa, 2000).

#### 2.4.2. Estimated daily intake (EDI)

Estimated daily intake (EDI) of heavy metals via ingestion route (crayfish) was calculated using Eq. (3)

$$EDI = \frac{C \times IR \times EF \times ED}{BW \times AT}$$
(3)

Assumptions for the health risk calculations were;

- a. Ingested dose is equal to the absorbed pollutant dose (USEPA, 1989).
- b. Cooking has no effect on the pollutants (Cooper et al., 1991).
- c. The average body weight of a Nigerian adult is assumed to be 70 kg.
- d. The average body weight of a Nigerian child is assumed to be 16 kg.

Where: EF = exposure frequency (360 days year<sup>-1</sup>); ED = exposure duration (70 years for adults), equivalent to the average lifetime; IR = ingestion rate (kg person<sup>-1</sup> day<sup>-1</sup>), (0.02kg person<sup>-1</sup> day<sup>-1</sup> for adults); C = metal concentration in crayfish (mg kg<sup>-1</sup>); BW = average body weight (kg), (70 kg for adults, 16 kg for children); AT = average exposure time for noncarcinogens (365 days year<sup>-1</sup>×ED), total THQ in this study was treated as the arithmetic sum of the individual metal THQ values (USEPA, 2015).

# 2.4.3. Target hazard quotient (THQ)

The THQ, the ratio of the exposure dose to the reference dose (RfD), represents the risk of noncarcinogenic effects. If it is less than 1, exposure level is less than the RfD. This indicates the daily exposure at this level is unlikely to cause adverse effects during a person's lifetime, and vice versa. The dose calculations were performed using standard assumptions from the integrated USEPA risk analysis (USEPA, 2000).

The target hazard quotient (THQ) was finally calculated using Eq. (4).

$$THQ = \frac{EDI}{RfD}$$
(4)

In this study, the total THQ was expressed as the arithmetic sum of the individual metal THQ values according to the method of Chien et al. (2002):

Total THQ (TTHQ) = THQ (toxicant 1) + THQ (toxicant 2) +  $\dots$  THQ (toxicant n) (5)

#### 2.4.4. Cancer risk (CR)

The CR over a lifetime of exposure to Pb was estimated using the cancer slope factor according to Eq. (6) (Peng et al., 2016; Shaheen et al., 2016):

$$CR = \frac{EF \times ED \times IR \times CF \times C \times CSF}{BW \times AT} \times 10^{-3}$$
(6)

where CSF is the cancer slope factor (mg/kg/day), while the other parameters have been defined previously. The US Environmental Protection Agency set an acceptable lifetime carcinogenic risk of  $10^{-5}$  (Saha et al., 2016).

#### 2.4.5. Relative risk (RR)

Yu et al. (2014) defined the RR of contaminants for both carcinogenic and non-carcinogenic effects, which can be helpful to recognize the most harmful contaminants. The RR equation is as follows:

$$RR = \frac{C}{RfD}$$
(7)

where all parameters have been previously defined (Yu et al., 2014).

#### 3. Results and discussion

#### 3.1. Results of recovery analysis

Table 1 shows the analytical result for calibration curve and percentage recovery for the analyzed heavy metals in the crayfish samples. The percentage recovery (%) of the heavy metals was determined by comparing the concentrated values of each metal from spiked and unspiked samples. The recoveries of metals in spiked sample were between 83.00 – 93.00%. Since the mean percentage recoveries for all analyte were within an acceptable range (70%–110%), this gives credence to the reliability of the results of this study.

#### 3.2. Elemental characterization

The range and mean levels of the analyzed metals (Fe, Zn, Mn, Pb, Cu, Cd and Cr) in the crayfish samples are shown in Table 2. It is observed that the crayfish samples contain relatively low levels of the analyzed metals. The results also reveal that Fe is the most abundant element with a range of 11.80–31.00 µg/g, mean value of 18.88  $\pm$  0.10 µg/g and coefficient of variation of 0.52 %, while Pb has the least concentration with a range of 0.40–1.40 µg/g, mean value of 0.91  $\pm$  0.01 µg/g and coefficient of variation of 1.10%. The order of decreasing mean values is as follow: Fe > Zn > Cu > Cr > Mn > Cd > Pb.

**Cadmium:** Cadmium concentration in this study ranges from 0.50 – 2.30  $\mu$ g/g. Cadmium is a toxic metal and there is no evidence of its essentiality to humans. The mean concentrations of Cd in this present study is higher than those reported by Hothem et al. (2007) and lower than those reported by Gherardi et al. (2002) in similar studies. Cadmium concentration in this study is also higher than the maximum permissible concentration set by World Health Organization/United States Environmental Protection Agency. This gives a cause for concern as Cd is known to be hazardous to human health at elevated level.

**Chromium:** The concentration of Chromium in this study ranges from  $1.80 - 5.90 \mu g/g$ . The mean concentrations of Cr in this present study is higher than those reported by Hothem et al. (2007) and lower than those reported by Bruno et al. (2006). Chromium concentration in this study is also higher than the maximum permissible concentration set by World Health Organization/United States Environmental Protection Agency. This also calls for concern as Cr is known to be hazardous to human health at elevated level.

**Copper:** In this present study, the concentration of Cu ranges from  $8.10 - 10.80 \ \mu$ g/g. Copper is required in normal carbohydrate and lipid metabolism and blood formation (Hambidge et al., 1987). Its mean concentrations in this present study is however higher than those reported by Mackevičienė (2002) and lower than those reported by Hothem et al. (2007). The concentration of copper in this study is lower than the maximum permissible concentration set by World Health Organization/United States Environmental Protection Agency.

**Zinc:** The concentration of Zinc in this study ranges from  $11.30 - 23.60 \ \mu\text{g/g}$ . Zinc is involved in glucose and lipid metabolism; hormone function and wound healing and it also helps in proper hair growth (Hambidge et al., 1987). The mean concentrations of Zinc in this present study is higher than those reported by Madden et al. (1991) and lower than those reported by Bruno et al. (2006). Zinc concentration in this

Table 1. Analytical results for calibration curve and percentage recovery (% R) for the analyzed metals.

Metal	Amount spiked (µg/g)	Amount recovered (µg/g)	% Recovery
Pb	5.00	4.65	93.00
Cu	5.00	4.26	85.20
Zn	5.00	4.35	87.00
Fe	5.00	4.15	83.00

study is lower than the maximum permissible concentration set by World Health Organization/United States Environmental Protection Agency.

**Iron:** In this present study, the concentration of Fe ranges from 11.80 – 31.00  $\mu$ g/g. Iron is required in the production of red blood corpuscles, oxygen transportation and the functioning of many enzymes in the organism. It also plays a significant role on vitamin A and iodine metabolism (Allen, 2002). Its mean concentrations in this present study is however higher than those reported by Hothem et al. (2007). The concentration of copper in this study is higher than the maximum permissible concentration set by World Health Organization/United States Environmental Protection Agency.

Lead: In this present study, the concentration of Lead ranges from  $0.40 - 1.40 \mu g/g$ . Its mean concentrations in this present study is however higher than those reported by Hothem et al. (2007) and lower than those reported by Bruno et al. (2006). The concentration of lead in this study is lower than the maximum permissible concentration set by World Health Organization/United States Environmental Protection Agency.

**Manganese:** In this present study, the concentration of Manganese ranges from  $1.20 - 2.80 \mu g/g$ . Manganese is required in bone formation and also in fat and carbohydrate metabolism. Its mean concentrations in this present study is however higher than those reported by Hothem et al. (2007). The concentration of copper in this study is higher than the maximum permissible concentration set by World Health Organization/United States Environmental Protection Agency.

# 3.3. Pollution index (PI) of the heavy metals in the crayfish samples

The PI of the heavy metals in the crayfish samples was carried out to determine the extent of contamination/pollution associated with the crayfish samples. The PI values of the metals are presented in Table 3. The results of the PI show that the crayfish samples are contaminated with Cd, Cr, Fe and Mn as the PI  $^{>}$  1. Other analyzed metals such as Zn, Pb and Cu have PI values less than 1. This suggests that the crayfish samples are clear of contamination by these metals.

#### 3.4. Cluster analysis (CA)

The hierarchical CA was used to determine the relationship among the various heavy metals using Euclidean distance as measure of similarity. This was performed using Statistical Package for Social Scientist (SPSS). Figure 1 shows the results of CA of the analyzed metals in the samples.

Cluster analysis grouped the heavy metals into clusters on the basis of similarities within a group and dissimilarities between different groups. Parameters belonging to the same cluster are likely to have originated from a common source and/or similar chemical properties. The CA results indicates three groups namely: A (Fe), B (Zn and Cu) and C (Cr, Mn, Pb and Cd) which showed closest inter-element clustering, indicating similar chemical properties such as variable oxidation state etc; it may also be due to similar genetic origin or source.

# 3.5. Correlation matrix analysis (CMA)

Correlation coefficient measures the strength of the linear relationship between any two variables on a scale of -1 (perfect inverse relation) through 0 (no relation) to +1 (perfect sympathetic relation). In this study, the raw data was used in calculating the correlation coefficient using the Microsoft Excel computation software package.

Table 4 shows the CMA results of the analyzed metals in the crayfish samples; positive and strong significant correlations exist between Cr/Cd, Fe/Cd, Fe/Cd, Pb/Cd, Pb/Fe and Cu/Zn, while strong and negative correlation exists between Zn/Cr. Strong and significant positive correlations indicated chemical affinity, similar genetic origin and/or common background levels in the samples, whereas the negative correlation could indicate that the metals originated from different sources or have non-chemical similarity.

# Table 2. Elemental concentrations of the crayfish samples ( $\mu g/g$ ).

Element ( $\mu g/g$ )SP1SP2SP3SP4SP5SP6SP7SP8SP9SP10RangeMean $\pm$ SDCV (%)Cd $2.00 \pm$ $0.082.30 \pm0.060.040.50 \pm0.041.00 \pm0.061.90 \pm0.082.30 \pm0.060.60 \pm0.040.70 \pm0.041.00 \pm0.061.30 \pm0.063.840.06Cr5.40 \pm0.045.90 \pm0.011.90 \pm0.032.30 \pm0.065.70 \pm0.061.80 \pm0.063.20 \pm0.042.30 \pm0.043.61 \pm1.180 \pm1.101.10Fe24.60 \pm0.0130.60 \pm0.1111.80 \pm0.0312.40 \pm0.0125.60 \pm0.0131.00 \pm0.0213.00 \pm13.40 \pm11.80 -13.00 \pm18.88 \pm0.52Zn11.30 \pm0.1213.60 \pm0.1211.80 \pm0.0313.40 \pm0.0113.40 \pm12.80 \pm13.00 \pm13.00 \pm11.80 -13.00 \pm18.88 \pm0.52Zn11.30 \pm0.1213.60 \pm0.0711.80 \pm0.0711.40 \pm0.0613.40 \pm13.20 \pm13.00 \pm13.90 \pm11.80 -13.80 \pm15.72 \pm0.06Pb0.90 \pm0.070.070.110.060.070.090.090.090.080.060.080.090.040.06Pb0.90 \pm0.010.20 \pm0.010.50 \pm0.011.20 \pm0.011.00 \pm0.020.40 -$														
$ \begin{array}{c c c c c c c c c c c c c c c c c c c $	Element (µg/g)	SP1	SP2	SP3	SP4	SP5	SP6	SP7	SP8	SP9	SP10	Range	$\text{Mean}\pm\text{SD}$	CV (%)
$ \begin{array}{cccccccccccccccccccccccccccccccccccc$	Cd	$\begin{array}{c} \textbf{2.00} \pm \\ \textbf{0.08} \end{array}$	$\begin{array}{c} 2.30 \ \pm \\ 0.06 \end{array}$	0.70 ± 0.04	$\begin{array}{c} 0.50 \ \pm \\ 0.06 \end{array}$	$\begin{array}{c} 1.00 \ \pm \\ 0.08 \end{array}$	$\begin{array}{c} 1.90 \ \pm \\ 0.03 \end{array}$	$\begin{array}{c} 2.30 \ \pm \\ 0.06 \end{array}$	0.60 ± 0.03	0.70 ± 0.04	$\begin{array}{c} 1.00 \ \pm \\ 0.02 \end{array}$	0.50– 2.30	$\begin{array}{c} 1.30 \ \pm \\ 0.05 \end{array}$	3.84
$ \begin{array}{cccccccccccccccccccccccccccccccccccc$	Cr	$\begin{array}{c} \textbf{5.40} \pm \\ \textbf{0.04} \end{array}$	$\begin{array}{c} 5.90 \ \pm \\ 0.01 \end{array}$	$\begin{array}{c} 1.90 \ \pm \\ 0.03 \end{array}$	$\begin{array}{c} \textbf{2.90} \pm \\ \textbf{0.03} \end{array}$	$\begin{array}{c} \textbf{2.30} \pm \\ \textbf{0.09} \end{array}$	$\begin{array}{c} 5.00 \ \pm \\ 0.10 \end{array}$	$\begin{array}{c} 5.70 \ \pm \\ 0.07 \end{array}$	$\begin{array}{c} 1.80 \ \pm \\ 0.06 \end{array}$	$\begin{array}{c} 3.20 \ \pm \\ 0.01 \end{array}$	$\begin{array}{c} 2.00 \ \pm \\ 0.04 \end{array}$	1.80– 5.90	$\begin{array}{c} \textbf{3.61} \pm \\ \textbf{0.04} \end{array}$	1.10
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$	Fe	$\begin{array}{c} 24.60 \pm \\ 0.60 \end{array}$	$\begin{array}{c} 30.60 \pm \\ 0.01 \end{array}$	$\begin{array}{c} 13.60 \pm \\ 0.12 \end{array}$	$\begin{array}{c} 11.80 \pm \\ 0.08 \end{array}$	$\begin{array}{c} 12.40 \pm \\ 0.01 \end{array}$	$\begin{array}{c} 25.60 \pm \\ 0.08 \end{array}$	$\begin{array}{c} 31.00 \pm \\ 0.02 \end{array}$	$\begin{array}{c} 13.40 \pm \\ 0.08 \end{array}$	$\begin{array}{c} 12.80 \pm \\ 0.01 \end{array}$	$\begin{array}{c} 13.00 \pm \\ 0.04 \end{array}$	11.80– 31.00	$\begin{array}{c} 18.88 \pm \\ 0.10 \end{array}$	0.52
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$	Zn	$\begin{array}{c} 11.30 \pm \\ 0.16 \end{array}$	$\begin{array}{c} 15.30 \pm \\ 0.08 \end{array}$	$\begin{array}{c} 23.60 \pm \\ 0.07 \end{array}$	$\begin{array}{c} 16.60 \pm \\ 0.11 \end{array}$	$\begin{array}{c} 17.80 \pm \\ 0.06 \end{array}$	$\begin{array}{c} 11.40 \pm \\ 0.07 \end{array}$	$\begin{array}{c} 13.30 \pm \\ 0.09 \end{array}$	$\begin{array}{c} 17.20 \pm \\ 0.09 \end{array}$	$\begin{array}{c} 13.90 \pm \\ 0.08 \end{array}$	$\begin{array}{c} 16.80 \pm \\ 0.08 \end{array}$	11.30– 23.60	$\begin{array}{c} 15.72 \pm \\ 0.08 \end{array}$	0.50
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$	РЬ	$\begin{array}{c} \textbf{0.90} \pm \\ \textbf{0.01} \end{array}$	$\begin{array}{c} 1.20 \ \pm \\ 0.01 \end{array}$	$\begin{array}{c} \textbf{0.90} \ \pm \\ \textbf{0.01} \end{array}$	$\begin{array}{c} 0.40 \ \pm \\ 0.02 \end{array}$	$\begin{array}{c} 0.50 \ \pm \\ 0.01 \end{array}$	$\begin{array}{c} 1.20 \ \pm \\ 0.01 \end{array}$	$\begin{array}{c} 1.40 \ \pm \\ 0.01 \end{array}$	$\begin{array}{c} 1.00 \ \pm \\ 0.03 \end{array}$	$\begin{array}{c} 0.60 \ \pm \\ 0.02 \end{array}$	$\begin{array}{c} 1.00 \ \pm \\ 0.01 \end{array}$	0.40– 1.40	$\begin{array}{c} \textbf{0.91} \pm \\ \textbf{0.01} \end{array}$	1.10
$ \begin{array}{cccccccccccccccccccccccccccccccccccc$	Cu	$\begin{array}{c} 8.10 \ \pm \\ 0.10 \end{array}$	$\begin{array}{c}\textbf{9.10} \pm \\ \textbf{0.07} \end{array}$	$\begin{array}{c} 10.60 \pm \\ 0.07 \end{array}$	$\begin{array}{c} 8.40 \ \pm \\ 0.08 \end{array}$	$\begin{array}{c} 8.80 \ \pm \\ 0.06 \end{array}$	$\begin{array}{c} 8.40 \ \pm \\ 0.09 \end{array}$	$\begin{array}{c} 9.50 \ \pm \\ 0.07 \end{array}$	$\begin{array}{c} 10.80 \pm \\ 0.07 \end{array}$	$\begin{array}{c} 8.60 \ \pm \\ 0.11 \end{array}$	$\begin{array}{c} 9.10 \ \pm \\ 0.11 \end{array}$	8.10– 10.80	$\begin{array}{c} 9.14 \ \pm \\ 0.08 \end{array}$	0.88
	Mn	$\begin{array}{c} 2.60 \pm \\ 0.03 \end{array}$	$\begin{array}{c} 2.00 \pm \\ 0.02 \end{array}$	$\begin{array}{c} 1.90 \pm \\ 0.03 \end{array}$	$\begin{array}{c} 1.30 \pm \\ 0.01 \end{array}$	$\begin{array}{c} 1.70 \pm \\ 0.02 \end{array}$	$\begin{array}{c} 2.80 \pm \\ 0.03 \end{array}$	$\begin{array}{c} 1.90 \pm \\ 0.04 \end{array}$	$\begin{array}{c} 1.90 \pm \\ 0.01 \end{array}$	$\begin{array}{c} 1.20 \pm \\ 0.03 \end{array}$	$\begin{array}{c} 2.00 \pm \\ 0.03 \end{array}$	1.20– 2.80	$\begin{array}{c} 1.93 \pm \\ 0.05 \end{array}$	2.59

SD = standard deviation, CV = coefficient of variation.

#### 3.6. Comparison of the total elemental concentrations with similar studies

The comparison of the concentrations of the analysed metals in this study with similar studies is presented in Table 5. The metal concentrations reported in this study was lower than that reported by Akpan et al. (2009) in the assessment of potential toxic elements pollution of marine environment using African cuttlefish (*Sepia bertholoti*) from the Gulf of

Guinea as an environmental bioindicator. Zinc and Mn concentrations in this study were higher than those reported by Sabir et al. (2003) in the effect of environmental pollution of quality of meat in District Bagh, Azad Kashmir, while Cu and Fe concentrations were lower than the result reported by Sabir et al. (2003). Zinc and Cu concentrations in this study

Heavy meta	al Mean	concer	tratic	m (i	וס/ס]	WHO/	USEPA	nermissil	ole I	limit	PI
samples.											
Table 3.	Pollution	index	(PI)	of	the	analyzed	heavy	metals	in	the	crayfish

Heavy metal	Mean concentration (µg/g)	WHO/USEPA permissible minit	PI
Cd	1.30	1	1.30
Cr	3.61	1	3.61
Fe	18.88	0.5	37.76
Zn	15.72	100	0.16
Pb	0.91	2	0.46
Cu	9.14	30	0.30
Mn	1 93	1	1.93

Table 4.	Correlation	matrix	of the	analyzed	metals in	1 the	cravfish	samples.
Tuble 4.	Gonciation	maun	or uic	anarvaca	metals n	I UIC	CIUVIDII	sumpres.

	Cd	Cr	Fe	Zn	Pb	Си	Mn
Cd	1						
Cr	0.92	1					
Fe	0.97	0.94	1				
Zn	-0.61	-0.73	-0.58	1			
Pb	0.73	0.56	0.78	-0.29	1		
Cu	-0.30	-0.46	-0.19	0.68	0.28	1	
Mn	0.62	0.46	0.57	-0.40	0.62	-0.12	1

 $n=10,\,r\geq 0.63$  at 95% confidence interval.

Bold indicates strong and significant correlation.

# Dendrogram using Average Linkage (Between Groups)





Table 5. Comparison of the total elemental concentration in this study with similar studies.

Element (µg/g)	This study	Sabir et al., (2003)	Akpan et al., (2009)	Kuklina et al., (2014)
Cd	1.30	NDT	NDT	$0.13\pm0.08$
Cr	3.61	NDT	NDT	$\textbf{0.99} \pm \textbf{0.84}$
Fe	18.88	1475–1525 (1500)	72.50–700 (247)	NDT
Zn	15.72	ND - 1.00 (0.25)	49.50–79.10 (65.90)	$128.23\pm44.33$
Pb	0.91	NDT	NDT	<sup>&lt;</sup> 0.50
Cu	9.14	61.00-68.00 (65.00)	47.20-81.20 (62.80)	$55.97 \pm 14.07$
Mn	1.93	N.D – 1.00 (0.50)	0.58–13.00 (7.07)	NDT
		1.1.00		

ND = not detected; NDT = not determined.

Table 6. Estimated Target Hazard Quotients (THQ) for analyzed metals from crayfish consumption by adults.

Heavy metal (µg/g)	EDI	RfD	THQ
Cd	0.00036	0.001	0.36634
Cr	0.00101	0.003	0.33910
Fe	0.00532	0.300	0.01773
Zn	0.00442	0.300	0.01476
Pb	0.00025	0.004	0.06411
Cu	0.00257	0.040	0.06439
Mn	0.00054	0.140	0.00388
TTHQ			0.87032

EDI = estimated daily intake, RfD = oral reference dose, THQ = target hazard quotient and TTHQ = total target hazard quotient.

 Table 7. Estimated Target Hazard Quotients (THQ) for analyzed metals from crayfish consumption by children.

Heavy metal (µg/g)	EDI	RfD	THQ
Cd	0.00160	0.001	1.60274
Cr	0.00445	0.003	1.48356
Fe	0.02327	0.300	0.07758
Zn	0.01938	0.300	0.06460
Pb	0.00112	0.004	0.28047
Cu	0.01126	0.040	0.28171
Mn	0.00237	0.140	0.01699
TTHQ			3.80768

EDI = estimated daily intake, RfD = oral reference dose, THQ = target hazard quotient and TTHQ = total target hazard quotient.

were lower than that reported by Kuklina et al. (2014) in the accumulation of heavy metals in crayfish and fish from selected Czech reservoirs, while Cd, Cr and Pb in this study were higher than the one reported by Kuklina et al. (2014). These variations could be attributed to the differences in the species investigated and the differences in the levels of metal pollution of the studied environments.

## 4. Human health risk assessment

# 4.1. Estimation of estimated daily intake (EDI) and target hazard quotient (THQ)

The oral reference dose based on the recommendations of United States Environmental Protection Agency (USEPA) is presented in Table 6. For the adults, the estimated daily intake for Cd, Cr, Fe, Zn, Pb, Cu and Mn are 0.00036, 0.00101, 0.00532, 0.00442, 0.00025, 0.00257 and 0.00054 respectively. For the children, the estimated daily intake for Cd, Cr, Fe, Zn, Pb, Cu and Mn are 0.00160, 0.00445, 0.02327, 0.01938, 0.00112, 0.01126 and 0.00237 respectively. These results showed that

Table 8. Estimated	Cancer	Risk	and	Relative	Risks	for	analyzed	metals	fron
crayfish consumption	n.								

Heavy metal (ug/g)	CSE	CR	RR (%)
			10.00
Ca	-	-	42.09
Cr	-	-	38.96
Fe	-	-	2.03
Zn	-	-	1.69
Pb	$8.5  imes 10^{-3}$	$2.179 \times 10^{-8}a$	7.36
		$9.536 \times 10^{-6}$ b	
Cu	-	-	7.39
Mn	-	-	0.44
CSE — concor clope factor	CP - concorrick	PP - rolativo rick a - ad	lulte and b _

CSF = cancer slope factor, CR = cancer risk, RR = relative risk, a = adults and b = children.

the EDI for the investigated metals were lower than the RfD (oral reference dose). These indicated that the intake of the crayfish might not have an adverse effect on the health of the populace consuming it. In both cases for the adults and children, the target hazard quotient (THQ) is according to the order: Cd  $^{\circ}$  Cr  $^{\circ}$  Cu  $^{\circ}$  Pb  $^{\circ}$  Fe  $^{\circ}$  Zn  $^{\circ}$  Mn. The THQ of each metal from the ingesting of crayfish was generally less than 1. This suggests that the populace consuming this crayfish would not experience significant health risks from the intake of individual metals (see Table 7).

# 4.2. Estimation of the cancer risk (CR) for Lead and relative risk

The estimated factors of the cancer risk for Pb and relative risk are presented in Table 8. Based on the result of this study, the CR factor for Pb over a lifetime of exposure through contaminated crayfish consumption by adults and children are  $2.179 \times 10^{-8}$  and  $9.536 \times 10^{-8}$  respectively. The tolerable value of lifetime carcinogenic risk set by USEPA is  $10^{-5}$ . The cancer risk factor of Pb obtained in this study is lower than the set tolerable limit, indicating that the consumption of the crayfish might not pose carcinogenic risk for Pb.

The non-carcinogenic relative risk (RR) values for the consumption of contaminated crayfish for all the metals is of the order: Cd  $^{\circ}$  Cr  $^{\circ}$  Cu  $^{\circ}$  Pb  $^{\circ}$  Fe  $^{\circ}$  Zn  $^{\circ}$  Mn. The contribution of Cd is 42.09 %, while that of Mn is 0.44 %. The highest concern of crayfish consumption is related to Cd.

# 5. Conclusion and recommendation

The study investigated some selected potential toxic metals and their associated health risks to humans. The pollution index values of the metals showed that Cd, Cr, Fe and Mn have values higher than 1 which indicated a level of contamination for the cravfish samples. Correlation matrix and cluster analysis indicated significant relationship between the analyzed metals which suggested similar sources and/or genetic origin for the metals. The estimated daily intake was less than the oral reference dose for both adults and children. The target hazard quotient was also less than 1. The cancer risk was also less than the set tolerable limit. The health risk assessment showed that the concentrations of the investigated potential toxic metals will not pose threat to the health of the consumers. However, based on the results obtained for the relative risk, among the considered metals, the main risk for human health can be related to the amount of Cd. Due to the possible accumulation of this metal to toxic levels, it is recommended that the crayfish should be consumed at moderate amount.

# Declarations

#### Author contribution statement

Festus Mayowa Adebiyi: Conceived and designed the experiments; Contributed reagents, materials, analysis tools or data; Wrote the paper. Odunayo Timothy Ore: Performed the experiments; Analyzed and interpreted the data; Contributed reagents, materials, analysis tools or data; Wrote the paper.

Isaiah Olayemi Ogunjimi: Performed the experiments; Contributed reagents, materials, analysis tools or data; Wrote the paper.

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#### Competing interest statement

The authors declare no conflict of interest.

# Additional information

No additional information is available for this paper.

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