

## Research Article

# Heavy metals concentration profile of an aquatic environment and health implications of human exposure to fish and prawn species from an urban river (Densu)

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### Abstract:

Fish is a good source of protein; however, certain anthropogenic activities can contaminate their habitat with elevated heavy metals levels. In this study, copper, lead, mercury, cadmium, and arsenic in fish tissue, water and sediment were determined using PerkinElmer PINAAcle 900T Graphite AAS. Standard indices in human health risk assessment were used to estimate non-carcinogenic implications associated with consuming *Clarias batrachus*, *Clarias gariepinus*, *Hemichromis fasciatus*, *Chrysichthys nigrodigitatus*, and *Macrobrachium rosenbergii* from Densu River. Heavy metal concentration levels recorded in November 2017 were in the order of surface water < pelagic fish < benthic fishes < sediments. Cadmium and Lead levels in all investigated fish tissues exceeded FAO/WHO recommended standard. Pb, Cd, and Hg mean concentration levels in the water exceeded the WHO threshold level of 0.01, 0.003, and 0.001mg/kg, respectively. Concentration level of all sediment samples was below the USEPA set limit for analyzed heavy metals. From the correlation analysis, *Hemichromis fasciatus* was identified as an applicable bioindicator for assessing heavy metal pollution because it correlated with water and sediment significantly. Principal component analysis ascribed heavy metal pollution in Densu River to anthropogenic activities along the river. The interpretation of estimated daily intake computation showed that the content of individual heavy metals in the fishes is not likely to endanger the health of the consumers. However, the recorded hazard index for *Clarias gariepinus*, *Clarias batracus*, and *Macrobrachium rosenbergii* exceeded one (HI>1), an indication of a non-cancer risk to consumers.

**Keywords:** Densu River, Fish, Health risk assessment, Sediment, Source identification.

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## Introduction

Contamination of natural ecosystems and the environment as a whole can be attributed to the constant unsustainable utilization of heavy metals, mainly for industrial purposes (Chen *et al.*, 2015; Borrell *et al.*, 2016; Mohanta *et al.*, 2020). It is established that any deviation from the baseline situation in an ecosystem can lead to an unpleasant reaction in the system. The global extent of the deleterious adverse impact of Cd, As, Hg, Pb, and Cu on ecology prompted the United States Environmental Protection Agency (USEPA) to enlist them as primary concern pollutants that need to be strictly controlled (Abrahams 2002, Rodrigues *et al.*, 2013). So heavy metals are of significant health concern globally due to their impact on biological organisms, humans, and the aquatic ecosystem in general (USEPA 2000, Karadede and Unlü, 2000; Gao *et al.*, 2016). Heavy metals are naturally toxic, accumulative, bioavailable, and non-biodegradable (Ruilian *et al.*, 2008). Several heavy metals, including Pb, As, Cd, and Hg, are also soluble in fat and have the ability to cross biological barriers (Kibria, 2016). These capabilities make heavy metals harmful to both human health and the immediate ecosystem because of their biological amplification in the food web (Kibria 2016, Wang *et al.*, 2012). An in-situ fish species may serve as a bioindicator for determining metal levels in an aquatic environment. The feeding pattern, age, size, elimination kinetics among fish species, and prevailing conditions in

both water and sediments in an aquatic ecosystem affect heavy metal accumulation in fish tissues (Farkas *et al.*, 2003; Kelly *et al.*, 2008; Biswas *et al.*, 2012). Consumption of contaminated water resources by human, plant, and a host of living organisms have profound health implications. For example cadmium is persistent and so can be stored in muscles, lobsters, and fish causing seafood poisoning in humans. Victims of such poisoning end up with diseases and cancer of the bone, heart, kidney, etc. (Kibria *et al.*, 2010). Elevated levels of Pb in the body deteriorate immune, circulatory, endocrine, and enzyme systems (Chen *et al.*, 2015). Therefore, heavy metals, inclusive of As, Cd, Pb, Cu, and Hg, are classified as toxic, and maximum acceptable levels are set for human consumption (Karadede and Unlü, 2000; Zhao *et al.*, 2012).

In Ghana and many other developing countries, contamination of heavy metals in water originates mainly from anthropogenic sources, such as fertilizer application, mining, industrial activities, and indiscriminate discarding of waste with metal residues (Mohanta *et al.*, 2020). In the case of the Densu River, intensive human activities and rapid industrialization are some of the many factors that lead to contamination of the river (Ansah-Asare, 2001). The river provides both drinking water and fish resources to residents of the western part of Accra (Ansah-Asare, 2001). The river contributed significantly to the national fish

production stock in the past. As a result of the increase in population and urbanization, portions of the river are encroached and used for residential, industrial, and agricultural activities, which have impacted the river with various waste effluents that contain some levels of heavy metals (Ansah-Asare, 2001; Kuma and Ashley, 2008).

It should be noted that most metals are naturally non-biodegradable and therefore remain mobile across different environmental media due to their high level of persistency and mobility. In many cases, in aquatic settings, surface sediment, relative to water, acts as a sink to store heavy metals (Turkmen *et al.*, 2005; Chen *et al.*, 2015; Li *et al.*, 2016). Suspended particulates settling on river beds, under certain conditions in water bodies, have the capacity to absorb released heavy metals into the medium (Dhanakumar *et al.*, 2015; Ke *et al.*, 2017). Sediments provide food and home for fish resources and other benthic plants. For this reason, a proper sedimentary analysis is relevant to a comprehensive study of heavy metals in the entire water body.

This present study aims at (1) determining the level and distribution of As, Cd, Pb, Cu, and Hg in water, sediment, and five species of fish and prawn; (2) identifying the relationship between heavy metal concentration in fish and prawn species, water and sediments; (3) identifying sources of contaminants in river Densu; and finally (4) conducting a human health risk assessment for consuming five species of fish and prawn (*Clarias batrachus*,

*Clarias gariepinus*,  
*Hemichromis fasciatus*,  
*Chrysichthys nigrodigitatus*, and  
*Macrobrachium rosenbergii*) from the Densu River.

## Materials and methods

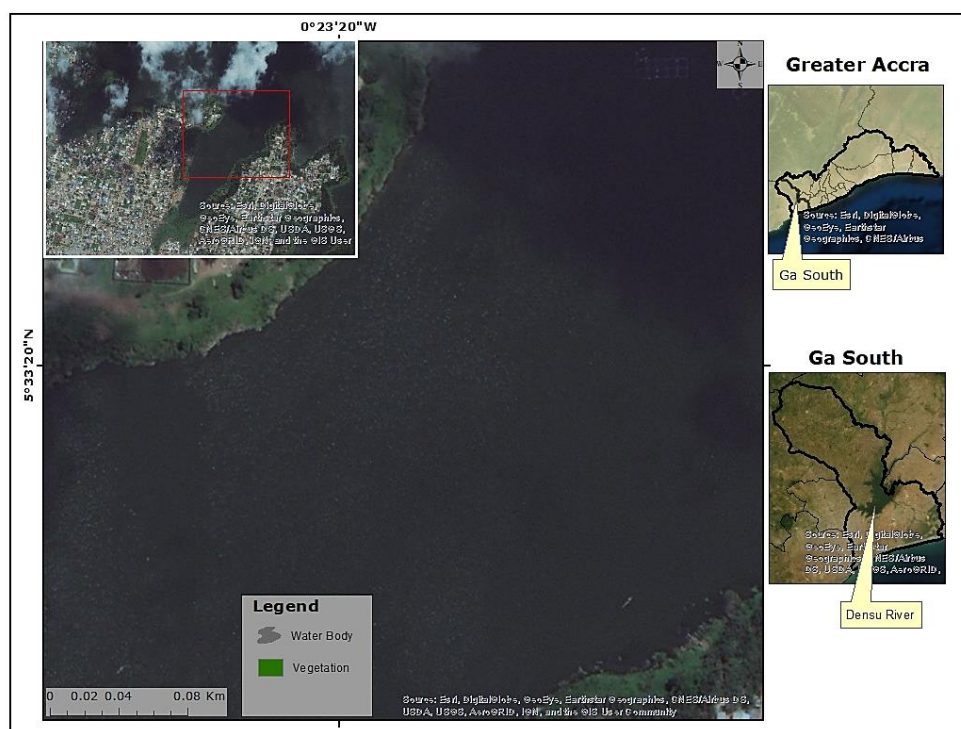
### Study site

Densu River is situated within coastal savannah, as shown in figure 1. The geography of the area is undulating and exhibits two contrasting rainfall patterns (rain season and dry season). The average water level of the river is 14.33m, and its storage capacity is  $113.5 \times 10^6 \text{ m}^3$  (25000 MG), covering an area of 20.5km<sup>2</sup> (Kuma and Ashley 2008). Weija reservoir covers 1183.167Km<sup>2</sup> with a projected inflow of 315,000m<sup>3</sup> per day and 40,500m<sup>3</sup> per day. As expected, a large quantity of water from the Densu River is consumed by a section of Ghanaians (Kuma and Ashley 2008).

### Fish sample collection and preparation

A total of 50 fish and prawns of five different species (*Clarias batrachus*, *Clarias gariepinus*, *Hemichromis fasciatus*, *Chrysichthys nigrodigitatus*, and *Macrobrachium rosenbergii*) were sampled from commercial catches made by local fishermen at the landing site in November 2017. The fish samples were immediately placed in clean rubber bags, kept on ice in a closed ice-chest, and were transferred to the laboratory. At the laboratory, measurements were taken for fresh weight and length of each fish sample to evaluate the coefficient of condition (K) using the

equation  $K = \frac{100W}{L^3}$ , where  $W$  is the fresh weight of fish in grams, and  $L$  is fork length in cm.



**Figure 1: Study map showing the Densu River in the Greater Accra Region, Ghana.**

The collected fishes were then tagged and stored at  $-18^{\circ}\text{C}$  in a deep freezer before preparing them for chemical analysis. On the day of chemical analysis, collected fishes were brought out of the freezer to allow them to thaw at room temperature. Distilled water was used in washing the samples, after which they were placed on tissue paper to dry. The scales and skins were taken off before the muscles were removed from each sample into a separate clean container. One gram of each muscle was accurately weighed and homogenized separately. Each homogenized fish sample was digested following the method described by (Akagi and Nishimura, 1991), as

reported by (Gyimah *et al.*, 2018). Briefly, 2ml of distilled water, 5ml nitric acid, and 3ml sulphuric acid were added to the sample, followed by heating on a heating mantle at  $85^{\circ}\text{C}$  for 30 min. Complete digestion was ascertained when the solution turned into a clear light yellow. The solution was allowed to cool at room temperature. The sample solution was then passed through a Whatman  $0.45\mu\text{m}$  paper filter into a 100ml volumetric flask, and the filtrate was diluted with distilled water to the 50ml mark. The entire filtrate was stored in labeled sterile plastic containers at  $4^{\circ}\text{C}$  and analyzed within 30 days.

### *Sediment and water sampling procedures*

Ekman grab was used in collecting a total of 20 sediment samples. A hand-held global positioning system (GPS) was used to geo-reference the sampling position on Densu River. Sampling sites were located on the river, and a total number of subsamples of surface sediment was taken from the site (n=6). An air-sealed rubber container was then used to store the sampled sediments. Samples were then air-dried at room temperature, powdered, and sieved with a 2 $\mu$ m mesh size to remove relatively larger size particles. A 4ml perchloric acid and 3ml concentrated nitric acid was mixed with 1g of each dried sediment samples before digesting the content. The content was heated at 75–80°C for a period between 1-2 hours on a heating mantle until a clear solution was obtained. Before analysis, all of the content was filtered through a Whatman 0.45 $\mu$ m paper filter into a 100ml volumetric flask, and the filtrate was diluted with distilled water to the 50ml mark.

Twenty samples of water were collected using a 1.0L polythene bag. The labeled bottle was rinsed three times before it was immersed in the river at a depth of 10cm. The content was acidified with 2mL HNO<sub>3</sub> before transporting to the lab. Acid digestion was performed using EPA method 3005 as reported by (Gyimah *et al.*, 2018). Shortly, the content was transferred into a 100mL Pyrex beaker, after which 1mL of HCl and 2mL of HNO<sub>3</sub> was added to 50ml of acidified

water, which was heated till the volume dropped to 30mL. The solution was filtered through a Whatman 0.45 $\mu$ m filter paper into a 50mL flask. The content was topped up with distilled water to the 50ml mark.

Finally, to ascertain the accuracy of the analytical method certified reference materials (CRMs) for sediment (SRM, 1944, New York Waterway sediment), water (Certified Reference Material ISE 999) and fish (DORM-3, fish protein, the National Research Council, Canada) were used.

### *Heavy metal analysis*

The concentration of metals (As, Cd, Pb, Cu, and Hg) in fish, water, and sediment samples was analyzed using Perkin Elmer PINAccel 900T Graphite AAS. The analyses of Cd, Pb, As, and Cu in all samples were performed using a Hollow cathode lamp (HCL) as described in Akoto *et al.* (2016). Mercury analysis was done with an electrodeless discharge lamp (EDL) coupled with an aflow injection assemble system (FIAS), as reported in Gyimah *et al.* (2018). The AAS calibration was based on a linear five-point calibration curve for which the coefficient of the calibration was greater than 0.999. Replicated analyses were done for all samples with a relative standard deviation (RDS) less than 0.04, an indication of good precision. For quality control, blanks and duplicate samples were analyzed after every 10 sample analysis. Replicated analyses of the reference materials exhibited a good accuracy

( $RSD \leq 0.03$ ), and the recovery rates ranged from 95% to 120% for all of them. Detection limits (DL) were determined using elemental standards in dilute aqueous solution. At 98% confidence level, detection level in mg/L for arsenic, cadmium, lead, and copper were 0.002, 0.001, 0.001, and 0.004 respectively. The detection limit of mercury was 2.0 pg total mercury.

Reagents used in the work were of analytical grade purchased from Sinopharm Chemical Reagent Co., Ltd. (Shanghai, China) and Yongfeng Chemical Reagent Co., Ltd. (Jiangsu, China).

#### *Health risk analyses of fish and prawn*

The health risk calculations were performed using mean values and standard assumptions from USEPA risk analysis unless otherwise stated (USEPA, 2000). Estimated daily intake (EDI), a function of heavy metal levels, food consumption, and the bodyweight of fish, was computed so that the level of heavy metal exposure through fish consumption could be assessed. To assess the level of human exposure to metal-contaminated fish, these assumptions were held-the ingested dose was equivalent to the absorbed pollutant dose, and the contaminants in fish are not affected by the method of cooking. The level of exposure to metals was assessed for adults only with the assumption that if it has adverse effects on adults, children could also be adversely affected (USEPA, 2000).

The EDI of heavy metals for adults was calculated as follows:

$$EDI = \frac{C \times Ccons}{BW} \quad (1)$$

where C is concentration of heavy metals in fish (mg/kg wet weight), Ccons is average daily consumption of fish in Ghana (68.5 g/day, Korateng *et al.*, 2006), and Bw represents average body weight of Ghanaian adults (65 kg, Biritwum *et al.*, 2005). Based on results of EDI computation, the level of human exposure to metals via food ingestion was determined by comparing EDI values for individual metals in fish species to oral reference dose (RfD,  $Hg = 1.6 \times 10^{-4} \text{ mg kg}^{-1} \text{ day}^{-1}$ ,  $Cu = 4.0 \times 10^{-2} \text{ mg kg}^{-1} \text{ day}^{-1}$ ,  $Cd = 1.0 \times 10^{-3} \text{ mg kg}^{-1} \text{ day}^{-1}$ ,  $As = 3 \times 10^{-4} \text{ mg kg}^{-1} \text{ day}^{-1}$ ,  $Pb = 4 \times 10^{-3} \text{ mg kg}^{-1} \text{ day}^{-1}$ , USEPA, 2009).

The hazard quotient (HQ) was computed using Equation (2) to assess non-carcinogenic risk for each of the metals. The final obtained value, after computation, determined health risk resulting from exposure to one metal. In this study, HQ comparison was fixed at 0.20, hence HQ values  $< 0.20$  indicated no potential adverse health effect, while HQ values  $> 0.20$  indicated a potential adverse health effect.

$$HQ = \frac{EDI}{Rfd} \quad (1)$$

Where Rfd represents reference dose.

According to reports from Hallenbeck and Cunningham (1986) and Madden (2003), the probable consequent interactive effect of pollutants on humans usually result from exposure to one or more pollutants. Therefore,

based on the HQ, human risk assessment for non-carcinogenic effects due to exposures to two or more metals within an individual fish species was calculated.  $HI > 1$  indicated a high chance of a non-carcinogenic effect on human health occurring, whereas  $HI < 1$  indicated no occurrence of non-carcinogenic effects on human health resulting from exposure to heavy metals concentrations in individual fish species. The formula for calculating HI for each fish species is expressed below:

$$HI_{\text{fish } i} = HQ_1 + HQ_2 + \dots + HQ_n \quad (2)$$

#### Statistical analysis

STATA statistical package software (version 15, IBM, Chicago, IL, USA) was used to statistically analyze the collected data. Existence of statistically significant differences in heavy metals among fish species was tested using one-factor analysis of variance (ANOVA). Statistical significance was accepted at  $p < 0.05$ . A Difference between fish species was assessed by Duncan's multiple range test. Standard deviation and mean for different parameters were calculated statistically, at significant level of 0.05 and 0.01. Pearson correlation analysis was used to identify relationship between fish species and the aquatic environment (water and sediments). Principal component analysis was performed on the data to explore plausible sources of pollution from heavy metals. The data was transformed

by means of the Z scores and sum of variance of the factor coefficients was maximized using Varimax rotation.

## Results

### *Heavy Metals Distribution in surface water and sediments*

Table 1 shows mean concentration of metals in collected water and sediment samples from Densu River. Range and mean concentration of Cu, Pb, Cd, As and Hg in water were recorded as  $0.013 \pm 0.00 \text{ mg/kg}$  (Below detection limit -  $0.131 \text{ mg/kg}$ ),  $0.24 \pm 0.04 \text{ mg/kg}$  ( $0.005$ - $0.83 \text{ mg/kg}$ ),  $0.096 \pm 0.07 \text{ mg/kg}$  ( $0.002$ - $0.81 \text{ mg/kg}$ ),  $0.007 \pm 0.00 \text{ mg/kg}$  (BDL- $0.12 \text{ mg/kg}$ ) and  $0.003 \pm 0.00 \text{ mg/kg}$  (BDL- $0.18 \text{ mg/kg}$ ) respectively. Mean concentration level of Pb, Cd and Hg exceeded WHO threshold level set for Pb ( $0.01 \text{ mg/kg}$ ), Cd ( $0.003 \text{ mg/kg}$ ) and Hg ( $0.001 \text{ mg/kg}$ ) in water. Whereas, mean concentrations of Cu and As were below WHO potability limits of  $1.3 \text{ mg/kg}$  and  $0.01 \text{ mg/kg}$ . The highest metal concentration in sediment was recorded for Cu ( $6.72$ - $26.49 \text{ mg/kg}$ ) with a mean value of  $18.42 \pm 2.62 \text{ mg/kg}$ . This was followed by Pb and Cd with mean levels of  $2.25 \pm 0.19 \text{ mg/kg}$  ( $0.186$ - $4.32 \text{ mg/kg}$ ) and  $1.28 \pm 0.11 \text{ mg/kg}$  ( $0.153$ - $2.170 \text{ mg/kg}$ ), respectively. Least heavy metal concentration in sediment, however, were recorded for As and Hg with mean values of  $0.95 \pm 0.04 \text{ mg/kg}$  ( $0.09$ - $1.875 \text{ mg/kg}$ ) and  $0.38 \pm 0.01 \text{ mg/kg}$  ( $0.038$ - $0.423 \text{ mg/kg}$ ), respectively.

**Table 1: Heavy metals in surface water and sediment (mg/kg) of Densu River. SD: Standard deviation.**

Metal	Water		Sediment	
	Range	Mean $\pm$ SD (n=20)	Range	Mean $\pm$ SD (n=6)
Cu	BDL-0.131	0.013 $\pm$ 00	6.72–26.49	18.42 $\pm$ 2.62
Pb	0.005-0.83	0.240 $\pm$ 0.04	0.186–4.32	2.25 $\pm$ 0.19
Cd	0.002-0.81	0.096 $\pm$ 0.07	0.153–2.170	1.28 $\pm$ 0.11
As	BDL-0.12	0.007 $\pm$ 00	0.09–1.875	0.95 $\pm$ 0.04
Hg	BDL-0.18	0.003 $\pm$ 00	0.038–0.423	0.38 $\pm$ 0.01

#### Level of heavy metals in fish and prawns

The results obtained in the present study for examined fish species are summarized in Table 2. Concentrations varied from 0.01mg/kg wet wt. in *Macrobrachium rosenbergii* and *Hemichromis fasciatus* to 0.1mg/kg wet wt. in *Clarias batrachus* and *Clarias gariepinus* for Hg. For Pb from 1.1mg/kg wet wt. in *Chrysichthys*

*snigrodigitatus* to 2.1mg/kg wet wt. in *Clarias gariepinus*. For Cd from 0.14 mg/kg wet wt. in *Hemichromis fasciatus* to 0.67mg/kg wet wt. in *Clarias gariepinus*. For Cu From 0.45mg/kg wet wt.in *Hemichromis fasciatus* to 1.32mg/kg wet wt.in *Clarias gariepinus*. For As from 0.02mg/kg wet wt. in *Chrysichthys nigrodigitatus* to 0.18mg/kg wet wt. in *Clarias gariepinus*.

**Table 2: Metal concentration in fish species (mg/kg) in Densu River. \*Significant ( $p < 0.05$ ), different letters (a, b) indicate means that are statistically different, SD: standard deviation.**

Variable	Hg	Pb	Cd	Cu	As
	Mean $\pm$ SD (n=50)				
<i>Macrobrachium rosenbergii</i>	0.01 $\pm$ 0.01 <sup>a</sup>	1.4 $\pm$ 0.06 <sup>a</sup>	0.32 $\pm$ 0.28 <sup>a</sup>	1.20 $\pm$ 0.56 <sup>a</sup>	0.04 $\pm$ 0.01 <sup>a</sup>
<i>Chrysichthys snigrodigitatus</i>	0.03 $\pm$ 0.02 <sup>a</sup>	1.1 $\pm$ 0.10 <sup>a</sup>	0.28 $\pm$ 0.04 <sup>a</sup>	1.31 $\pm$ 0.13 <sup>ab</sup>	0.02 $\pm$ 0.02 <sup>a</sup>
<i>Clarias batrachus</i>	0.1 $\pm$ 0.03 <sup>a</sup>	1.5 $\pm$ 0.15 <sup>a</sup>	0.48 $\pm$ 0.07 <sup>a</sup>	1.22 $\pm$ 0.14 <sup>a</sup>	0.13 $\pm$ 0.06 <sup>a</sup>
<i>Clarias gariepinus</i>	0.1 $\pm$ 0.02 <sup>b</sup>	2.1 $\pm$ 0.15 <sup>a</sup>	0.67 $\pm$ 0.02 <sup>a</sup>	1.32 $\pm$ 0.06 <sup>ab</sup>	0.18 $\pm$ 0.02 <sup>a</sup>
<i>Hemichromis fasciatus</i>	0.01 $\pm$ 0.01 <sup>a</sup>	1.2 $\pm$ 0.15 <sup>a</sup>	0.14 $\pm$ 0.01 <sup>a</sup>	0.45 $\pm$ 0.15 <sup>a</sup>	0.06 $\pm$ 0.01 <sup>a</sup>
p –value	0.042*	0.282	0.128	0.032*	0.327

#### Relationship between heavy metals in fish species, sediments and surface water

Based on their different feeding pattern and ecological niche, the five species of prawn and fish were grouped into benthic (*Clarias gariepinus*, *Clarias batrachus*, *Chrysichthys nigrodigitatus*,

and *Macrobrachium rosenbergii*) and pelagic (*Hemichromis fasciatus*). Therefore, the mean concentration of heavy metals in fish, sediments, and surface water in ascending order are surface water < pelagic fish < benthic fishes < sediments.



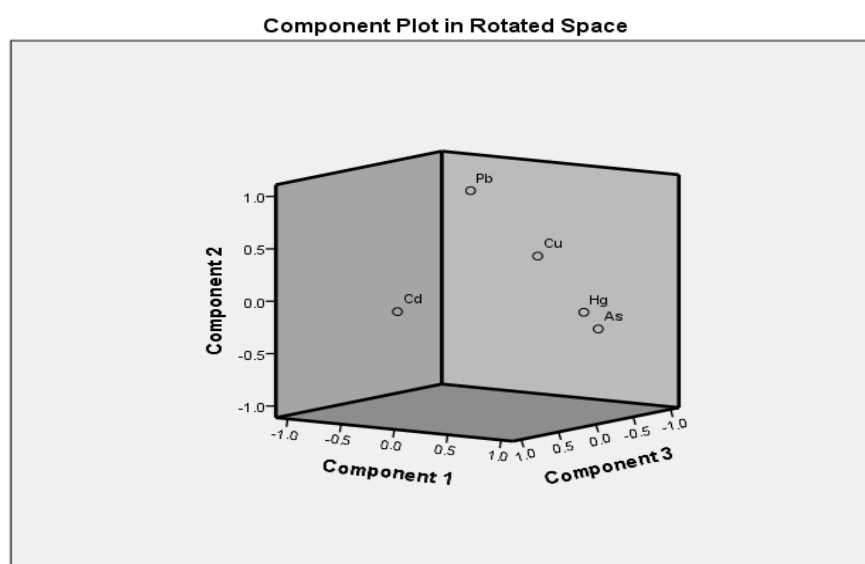
**Table 3: Correlation analysis of heavy metal concentration in fish, water, and sediment. Statistically, at significance levels 0.05\*(2 tailed); 0.01\*\*(2 tailed).**

	<i>Macrobrachium rosenbergii</i>	<i>Chrysichthys nigrodigitatus</i>	<i>Clarias batrachus</i>	<i>Clarias gariepinus</i>	<i>Hemichromis fasciatus</i>
Water	-0.201	r = -0.293	r = -0.361	r = -0.227	r = 0.583*
Sediments	0.582*	r = 0.308	r = 0.774**	r = 0.821**	r = 0.532*

### Sources of heavy metal contamination in sediment

Principal component analysis (PCA) was utilized in the identification of origins of heavy metal contamination. The data used for analysis were scaled and normalized through the Z scale transformation (Amankwaa *et al.*, 2020). Based on eigenvalues, three

extracted principal components are displayed in Fig. 2. The Sum of the first three PC axes is explained as 98.738% of the total variance, which is clearly expressed in Table 4. The extraction communalities were very high, indicating that PC1, PC2, and PC3 represented contaminants (As, Cu, Pb, Hg, and Cd) well.

**Figure 2: Factor loadings of rotated components of As, Cd, Hg, Cu, and Pb in sediments of Densu River.****Table 4: Component matrix of factor analysis of heavy metals in the Densu River. Factor loadings in bold are statistically significant (value>0.7).**

Variable	Communalities (extraction)	PC 1	PC2	PC3
Cu	1.000	<b>0.795</b>	0.511	0.327
As	0.999	<b>0.852</b>	-0.284	-0.402
Hg	0.996	<b>0.986</b>	-0.056	-0.017
Pb	0.968	-0.078	<b>0.996</b>	-0.021
Cd	0.975	-0.051	-0.009	<b>0.996</b>
Eigenvalue		2.358	1.548	1.031
% Variance		47.167	30.961	20.611
Cum % Variance		47.167	78.128	98.738

*Human health risk analyses of heavy metals in fish*

Estimated daily intake of Pb, As, Hg, Cd, and Cu via ingestion of fish from Densu River is displayed in Table 5. Estimated daily intake of the five metals recorded values lower than their

respective RfD's for consumers. This outcome indicates that average consumers of any fish species from Densu River are unlikely to suffer complications related to exposure (through ingestion) to the metals.

**Table 5: Estimated daily intake (EDI) of heavy metals in selected fish species in the Densu River.**

Fish species	Heavy metal				
	As	Pb	Cu	Hg	Cd
<i>Macrobrachium rosenbergii</i>	4.54E-05	1.50E-03	1.36E-03	1.93E-05	3.64E-04
<i>Chrysichthys nigrodigitatus</i>	2.27E-05	1.20E-03	1.49E-03	3.52E-05	3.18E-04
<i>Clarias batrachus</i>	1.48E-04	1.70E-03	1.39E-03	1.27E-04	5.46E-04
<i>Clarias gariepinus</i>	2.05E-04	2.30E-03	1.50E-03	1.27E-04	7.62E-04
<i>Hemichromis fasciatus</i>	6.82E-05	1.30E-03	5.12E-04	1.36E-05	2.73E-04
<b>RfD(USEPA 2009)</b>	3.00E-04	4.00E-03	4.00E-02	1.60E-04	1.00E-3

The human risk from fish consumption is summarized in Table 6. The results showed that the hazard quotient ranged from 0.01 for Cu to 0.80 for Hg in the investigated fish species. In all fish species, hazard quotient (HQ) for Cd and Pb was higher than the HQ safety limit of 0.20 (HQ>0.20), which is an indication that Cd and Pb levels within individual fish species posed harmful health effects to consumers. Also, HQ values for As in *Clarias batrachus*, *Clarias gariepinus*, and *Hemichromis*

*fasciatus* exceeded the 0.20 safety limit, indicating that the concentration level of arsenic in the fish species may harm the health of the consumers. *Chrysichthys nigrodigitatus*, *Clarias gariepinus*, and *Clarias batrachus* had HQ values above 0.20 for Hg, which could potentially harm consumers. It should be noted that the concentration of Cu in all fish species was lower than the HQ safety limit of 0.20, expressing no potential health effect.

**Table 6: Hazard quotients (HQ) of heavy metals in selected fish species in the Densu River.**

Fish species	Heavy metal					
	As	Pb	Cu	Hg	Cd	HI
<i>Macrobrachium rosenbergii</i>	0.15	0.38	0.03	0.12	0.36	1.04
<i>Chrysichthys nigrodigitatus</i>	0.08	0.30	0.04	0.22	0.31	0.95
<i>Clarias batrachus</i>	0.49	0.43	0.03	0.80	0.55	2.29
<i>Clarias gariepinus</i>	0.68	0.58	0.04	0.80	0.76	2.85
<i>Hemichromis fasciatus</i>	0.23	0.33	0.01	0.09	0.27	0.92

Consumers should, however, be advised to eat the four fish species (*Clarias batrachus*, *Clarias gariepinus*,

*Hemichromis fasciatus*, and *Chrysichthys nigrodigitatus*) in moderation to avoid possible harmful

health effects that may result from the accumulation of As, Pb, Hg, and Cd. Furthermore, since humans are mostly exposed to more than one metal pollutant at a given time, the hazard index (HI) was computed for metals within each fish species. The results revealed that *Chrysichthys nigrodigitatus* and *Hemichromis fasciatus* obtained 0.95 and 0.92, respectively. This means consumers of the two fish species will not suffer from non-carcinogenic health risks due to the ingestion of more than one metal. However, *Clarias gariepinus*, *Clarias batrachus*, and *Macrobrachium rosenbergii* showed HI values of 2.85, 2.29, and 1.04, respectively. These outcomes indicate that consumers of the three fish species are highly likely to suffer from a non-carcinogenic health risk due to ingestion of two or more metals. Relative contributions of As, Pb, Cu, Hg, and Cd to HI obtained from *Clarias gariepinus* were 23.9%, 20.2%, 1.3%, 27.9%, and 26.7%, respectively. Relative contributions for *Clarias batrachus* were 21.5%, 18.5%, 1.5%, 34.7% and 23.8, for *Macrobrachium rosenbergii* were 14.5%, 35.9%, 3.3%, 11.5% and 34.8%, for *Chrysichthys nigrodigitatus* were 8.4%, 31.6%, 4.2%, 23.2% and 32.6%, for *Hemichromis fasciatus* were 25%, 35.9%, 1.1%, 9.8% and 29.3%. The HI followed the order of *Clarias gariepinus* > *Clarias batrachus* > *Macrobrachium rosenbergii* > *Chrysichthys nigrodigitatus* > *Hemichromis fasciatus*.

## Discussion

FAO/WHO does not have a maximum level set for As in fish. However, regulation limits indicate that Cu, Pb, Cd and Hg should not be above 40, 0.30, 0.20 and 0.5 mg/kg wet wt., respectively (FAO/WHO, 2011). Pb and Cd in all fish and prawn species in this study were above FAO/WHO limit for human consumption. The excesses maybe attributed to the level of mobility, persistence and bioavailability of Cd and Pb in tissues of some fish and prawn species. This is demonstrated by high Cd and Pb levels reported in *Alosa caspiensis* which is also a type of catfish species from middle Black Sea, Turkey (Tüzen, 2003). Similar results in Hg and Pb was reported by Mol *et al.* (2010) in *Silurus triostegus* and *Acanthobrama marmid* in Ataturk Lake, Turkey. From the study, high levels of Hg and Pb in tissues were attributed to petroleum leakage in the lake. *Clarias gariepinus* is reported to have a high level of Cd in the tissue collected from the Densu River in a different work (Anim *et al.*, 2011). Hülya *et al.* (2004) also reported high levels of heavy metal contamination in *S. triostegus*. Nzeve *et al.* (2014) detected a high level of Pb in *Clarias gariepinus* from Masinga Reservoir, Kenya. Dhanakumar *et al.* (2015) also found Cd and Pb levels to be high in fish from three major reservoirs of Cauvery delta region, India. Very high concentrations of Cd and Pb in fish from similar work are also reported by Chandra Sekhar *et al.* (2004) in India's largest freshwater lake known as Kolleru Lake. The high level

of heavy metals in fish species is attributed to local industrial and unchecked agricultural activities.

Surface water has a large surface area that is vulnerable to heavy metal pollution. However, contamination level is usually low because pollutants adhere to suspended particles, which can then settle in the bottom of the river. This may account for a low mean concentration of heavy metals in surface water and a high concentration level of heavy metals in sediment. Certain factors that may influence the high concentration of sedimentary metals are the weathering cycle and composition of the underlying rock. Mean concentrations in benthic fishes were higher than in pelagic fish. It should be noted that the levels of Cd and Pb in four omnivorous species of benthic fish (*Clarias gariepinus*, *Clarias batrachus*, *Chrysichthys nigrodigitatus*, and *Macrobrachium rosenbergii*) in this study were significantly high. These omnivores fishes are classified as bottom feeders and are at a high trophic level (Bruton, 1979; Ramesh and Kiran, 2016). They feed in particular on and around benthic plants located on sediments of the water bottom and mostly traverse the middle-lower column of the water where they are exposed to suspended particulates to prey on organic detritus, worms, insects, fish larvae, shrimps, and small fishes (Bruton 1979; Ramesh and Kiran, 2016). These cycles may increase bioaccumulation factors of the fish species for heavy metal uptake (especially Pb, Cd, and other metals

that are mobile and bioavailable), although metal levels in surface water and sediments are relatively low. On the other hand, *Hemichromis fasciatus*, a pelagic piscivore, recorded low mean concentration, probably due to its size, living and feeding pattern. This species feeds primarily on small fishes and mostly on the surface and mid-column of the water. Statistically, at significance levels 0.05 (2 tailed) and 0.01 (2 tailed), there was significant correlation between fish species and aquatic environment (water and sediment), such as *Clarias gariepinus* ( $r=0.821$ ), *Clarias batrachus* ( $r=0.774$ ), *Macrobrachium rosenbergii* ( $r=0.582$ ) and sediments. However, the absence of significant correlation between benthic fish *Chrysichthys nigrodigitatus* and sediments may be due to variations in their ability to accumulate heavy metals. Finally, it should be noted that there was a significant correlation between *Hemichromis fasciatus* and surface water ( $r=0.583$ ) and sediment ( $r=0.532$ ) because of living and feeding pattern of the fish species (Table 3). This indicates that *Hemichromis fasciatus* can be used as a bioindicator to determine the level of heavy metal pollution in Densu River.

From the principal component analysis, PC1, representing the first component, explained 47.167% of the variance and was significantly loaded with Cu, As, and Hg, which indicates that these metals originated from one source, mainly from anthropogenic activities, such as industrial effluent, electronic waste discharge, and

agricultural activities. The results obtained in this study reflect a large amount of wastewater rampantly discharged from industrial production and agricultural activities around the Densu River (Ansah-Asare, 2001). PCA was used to identify Chongqing and Yibin as anthropogenic sources of industrial waste, E-waste, chemicals, and automobile waste, which influenced the levels of As, Cu, Pb, Cd, Hg, and Zn in surface sediments of the upper Yangtze River, China (Yi *et al.*, 2017). PC2 expressed 30.961% of the variance and demonstrated strong positive ( $>0.7$ ) loading for Pb. The presence of Pb might be attributed to the use of lead-based paint, cars, and electronic batteries (Steinnes, 2013). Pb can also be attributed to vehicle emissions, as Saghatelian (2004) showed that vehicle emission was the principal source of Pb in Yerevan, the capital of Armenia. The same PC2 is reported by a study that identified anthropogenic pollution source as a significant contributor of lead, which augmented the level of As content in sediments along the banks of Bogacayi River in Turkey (Yalcin *et al.*, 2015). The third principal component (PC3), which explained a variance percentage of 20.611, was significantly loaded with Cd. Cd principally originates from tires, plating, and batteries, which is associated with activities of local car repair shops called “magazine” in local parlance. Ke *et al.* (2017) also utilized PCA in the identification of humans as a source of pollution. In general, PCA revealed that industrial sources and

agricultural activities are the main sources for As, Cu, Hg, Pb, and Cd in the Densu River.

Prolonged consumption of *Clarias gariepinus*, *Clarias batrachus*, and *Macrobrachium rosenbergii* may pose a higher health risk for consumers due to elevated levels of As, Pb, Hg, and Cd. Consumers are likely to suffer health complications, such as bone cancer and damage to the human nervous system, immune system, and kidney, which are associated with exposure to As, Cd, Pb, and Hg.

In conclusion, *Hemichromis fasciatus* was identified as a bioindicator for assessing the level of heavy metal pollution in the riverine. Results from the Human health risk analyses revealed that consumers of *Clarias gariepinus*, *Clarias batrachus* and *Macrobrachium rosenbergii* may suffer from a combined health effect by ingesting more than one heavy metal in fish species since their HI was recorded to be higher than one ( $HI > 1$ ). The principal component analysis identified anthropogenic activities as the main source of heavy metal contamination in the river. To safeguard the consumption of contaminated fish in the Densu River and to ensure a sustainable ecosystem, anthropogenic activities around the riverine body should be monitored regularly to reduce the heavy metal release into the river. Laws must be enacted and enforced to bar residents from encroaching the buffer zones of the river. Finally, future works on the ecological risk and environmental

impact assessment of the riverine body are highly recommended.

**Ethical statement:** Edible tissues from commercially harvested fish species caught by local fishermen were used in this present work.

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