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# Evaluation of Heavy Metal Pollution in Commonly Consumed Mollusc (*Crassostrea gasar*) from Elechi Creek, River State, Nigeria and the Health Risk Implications

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**Abstract**: Marine biotas are used to assess potential adverse human health risks associated with consuming protein-rich aquatic organisms. Heavy metal content of Mangrove oysters (*Crassostrea gasar*) was evaluated between January and June 2022 in 3 stations. Six heavy metals (copper, cadmium, zinc, lead, arsenic and iron) were determined using standard methods. Target Hazard Quotient (THQ) and Hazard Index (HI) were used for the non-carcinogenic assessment while Target Cancer Risk (TR) was used for the carcinogenic assessment of the potential human health risk of consuming the oysters. The heavy metal values recorded were Cu (473.2 – 596.7 mg/kg), Cd (2.33 – 3.84 mg/kg), 209.02 – 246.41 mg/kg), Pb (6.16 – 12.07 mg/kg), As (0.012 – 0.016 mg/kg) and Fe (1609.0 – 1846.0 mg/kg). All the heavy metals were above the acceptable limits except arsenic. Stations 2 and 3 had relatively higher values; attributed to anthropogenic activities. The THQ and HI values were less than 1 in all the metals and stations while TR for Pb and arsenic were within the negligible range in all the stations. However, Cd was unacceptable among the children in station 2. Station 3 had relatively higher values while the children were more vulnerable to both non-carcinogenic and carcinogenic risks. In conclusion, the consumption of oysters from Elechi Creek is considered safe based on acceptable levels of the THQ, HI and TR; though Cd-TR for children (Station 2) was unacceptable.

**Keywords:** Heavy metals, Mangrove oysters, Health risks, Hazard Quotient, Carcinogenic assessment.

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## 1. INTRODUCTION

Estuaries are transitional zone between rivers and marine environments found in coastal zones across the world (1). Due to intense socio-economic activities, estuaries are subjected to severe perturbation (2-4); serving as sinks for pollutants, including heavy metals. The constant mixture of fresh and saltwater in the estuaries provides for the remobilization of heavy metals (5). Heavy metals are discharged into the aquatic environment via natural and anthropogenic sources (6, 7).

In the aquatic environment, heavy metals are easily distributed and accumulated in the tissues of aquatic biota; leading to deleterious effects (8, 9). Aquatic organisms are rich in protein content, low in saturated fats and provide different health advantages (10). They are a ready source of nutrients for local residents (11). The nutritional content of seafood has increased its ever-increasing demand (12).

However, contamination of seafood especially by heavy metals elicits great interest because they can be accumulated in the surrounding environment (8, 13), which raises the issue of food safety globally. A number of marine organisms have been used as bioindicators in the evaluation of potential adverse human health risks associated with the consumption of contaminated marine biota (14-17).

Oysters are increasingly being studied as indicators of heavy metal pollution because of their wide distribution in coastal environments, susceptibility to pollution, abundance and ease of collection as well as sessile habit and low enzymatic activity level (4, 8, 9, 11, 18, 19).

In the Niger Delta, rivers have become targets for waste disposal due to their open and accessible nature (20). Agricultural, industrial, and municipal wastes are frequently discarded directly into rivers, turning them into convenient landfills (17, 21). Artisanal crude oil refineries have been reported to be a critical anthropogenic activity currently polluting the Niger Delta environment (22-24).

In view of the foregoing, there is a need to understand heavy metal dynamics and accumulation in oysters in Elechi Creek; bearing in mind that it is one of the commonest sources of protein in the area. The aim of this study is to evaluate the heavy metal content of mangrove oysters (*Crassostrea gasar*) and the potential non-carcinogenic and carcinogenic human health risks associated with its consumption.

## 2. MATERIALS AND METHODS

### 2.1. Description of Sample Stations

The study was carried out in Elechi Creek, Port Harcourt, Rivers State, Nigeria. It discharges into the Bonny Estuary and is brackish in nature. It extends from Eagle Island to the Illoabuchi Street waterfront. The creek had varied widths and was surrounded by mangrove trees. Some anthropogenic activities observed around the stations include industrial discharges, urbanization and stormwater runoff, agricultural activities, mangrove degradation, shipping and transportation, waste disposal, and industrial and construction activities.

Station one is located on a sand-Filled area known as Eagle Island (Latitude N04°47.149'; Longitude E006°58.958'). It is located around an abandoned artisanal refinery site. The dominant vegetation in the area is Nipa palm (*Nypa fruticans*) with scattered patches of white mangrove (*Laguncularia racemosa*) and red mangrove (*Rhizophora mangle*) and elephant grass (*Pennisetum purpureum*). Station two is located at the Sawmill area (Latitude N04°47.28'; Longitude E006°59.255'); about 2.14 km downstream of station 1. It is around an active artisanal refinery site.

The dominant vegetation is Nipa palm (*Nypa fruticans*). Station three is located in the Appa area (Latitude N04°47.047'; Longitude E006°59.362'); about 2.21 km downstream of station 2. It is located around crude oil and refined products storage areas used by illegal refiners. Nipa palm (*Nypa fruticans*) is also the dominant vegetation though a large expanse has been destroyed. A large stormwater canal also discharges into the area.

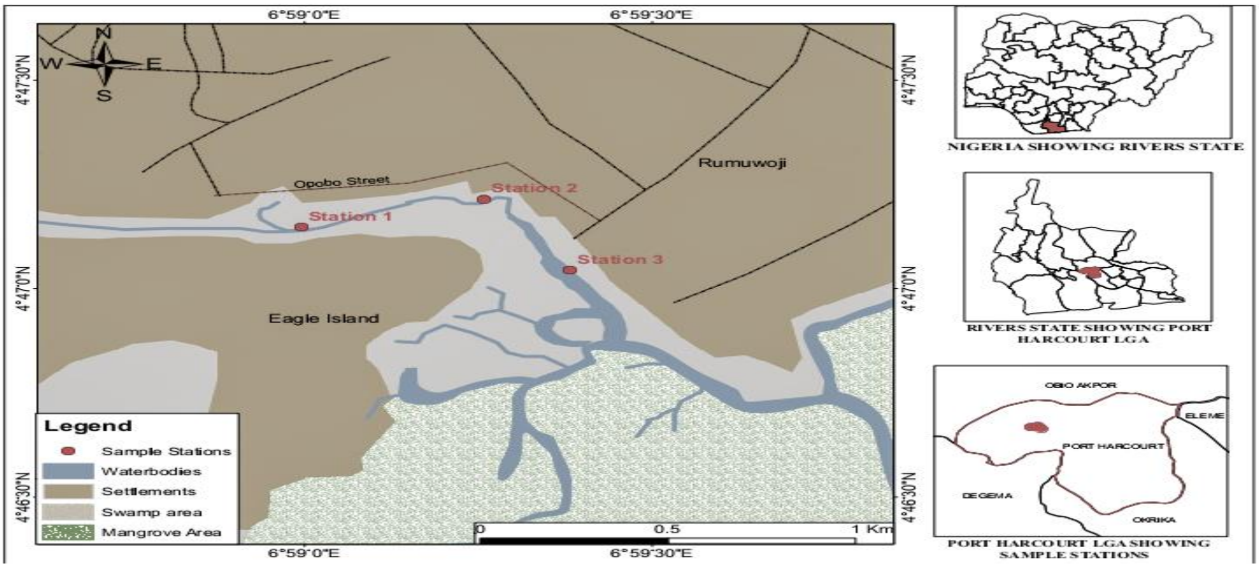


**Figure 1:** Showing the map of the study area and sampling stations.

### 2.2. Collection of Oyster Samples

Each station collected ten (10) Mangrove oysters (*Crassostrea gasar*). A total of 180 samples were collected between January and June 2022. The oysters were harvested from the prop roots of the mangrove tree during the low tide. The samples were immediately transported to the laboratory for analysis in an ice chest. The soft tissues from 8 to 10 individuals were dissected, dried, and stored in clean, clearly labelled plastic containers.

### 2.3. Sample Preparation and Digestion

The tissue samples, each weighing 0.5±0.01g, were placed straight into Teflon digestion containers that had been cleaned with acid. Each vessel received 10 ml of ultra-pure nitric acid, which was then heated to 100°C using an XT-9800 pre-treatment heater until nearly all the nitrogen dioxide was released.

In order to prepare the sample for microwave digestion, a 4 mL aliquot of concentrated $HNO_3$: HF (1:1 v/v) acid solution was added. Every digestion batch had a minimum of one reagent blank, one representative reference standard, and generally, one sample replication to assess homogeneity and procedure effectiveness.

There were three stages to microwave digestion: 1.5 MPa for 1 min, 1.0 MPa for 2 min, and 1.5 MPa for 3 min. The digested sample was transferred to a graduated plastic test tube and allowed for an hour to cool and the volume was made up to 100 mL with Milli-Q water (25, 26).

### 2.4. Quality Assurance and Control

After microwave digestion, each metal's certified reference materials (CRMs) from Sigma-Aldrich were employed for atomic absorption spectroscopy (AAS). The metals - cadmium (Cd), iron (Fe), arsenic (As), lead (Pb), zinc (Zn), and copper (Cu) were analysed in the oyster samples in triplicates. The apparatus was calibrated using atomic absorption standards for numerous dangerous metals that were buck-certified in order to create an analytical curve.

To avoid equipment drift, 10 samples were analysed before a reagent blank. According to calculations, the percentage recovery (%R) for metals was Fe (89.0%), Pb (98.7%), Cd (100%), As (99.6%), Zn (84.5%), and Cu (97.6%). An atomic absorption spectrophotometer (Model 210 VGP, Buck Scientific) was used to determine the metal contents in the samples.

### 2.5. Human Health Risk Assessment

The Non-Carcinogenic Risk Assessment and Individual Lifetime Cancer Risk were applied in this study. The Target hazard quotient (THQ) for each heavy metal was calculated in order to assess the potential health risk of consuming Oysters (*Crassostrea gasar*) collected from the study area (27). It was calculated using Equation 1:

$$THQ = \frac{ED \ x \ IR \ x \ EF \ x \ CW}{RfD \ x \ BW \ x \ AT} \ x \ 10^{-3} \quad (1)$$

Where ED is the Exposure duration – 70 years (adults) and 10 years (children); IR is the daily ingestion rate – 0.3 mg/kg/person/day (adults) and 0.15 mg/kg/person/day (children) (28); EF is the exposure frequency (365 days/year); CW is the concentration of respective heavy metal (mg/kg) in the oysters; RFD is the reference oral dose in mg/kg/day (0.001 for Cd, 0.004 for Pb, 0.3 for Zn, 0.0003 for As, 0.7 for Fe and 0.04 for Cu); BW is body weight - 70 kg (adult) and 25 kg (children) and AT (ED x EF) is the average time of the exposure – 25550 days (adult) and 3650 days (children) (29). THQ > 1 points to adverse non-carcinogenic effects that cannot be overlooked but acceptable levels are at HQ < 1.

### 2.6. Hazard Index (HI)

Hazard index (HI) is the cumulative potential for non-carcinogenic effects from more than one heavy metal through ingestion pathways and can be estimated from equation 2 (30).

$$HI = \sum_{i=1}^{n} THQ \quad (2)$$

Where HI is the hazard index for the overall toxic risk and n equals the total number of metals under consideration. If HI for non-carcinogenic adverse effects due to ingestion exposures is lower than one (HI < 1.0), then no chronic risks are expected to occur but if HI is greater than one (HI > 1.0), possible chronic risk arising from the ingestion exposures could manifest (31).

### 2.6. Carcinogenic Risk

Target Cancer Risk (TR) was used to determine the carcinogenic risk (8). Target cancer risk (TR) posed by the assessed heavy metals was determined with Equation 3 (35):

$$TR = \frac{ED \ x \ IR \ x \ EF \ x \ CW \ x \ C_{SF}}{BW \ x \ AT} \ x \ 10^{-3} \quad (3)$$

Where CSF is the Cancer Slope Factor while other input parameters have been previously defined in Equation 1. The acceptable range for carcinogenic risks is between $10^{-4}$ and $10^{-6}$ and values > $10^{-4}$ will likely result in cancer (8). The cancer slope factors (CSF) were Cd (6.3 mg/kg/day), Pb (0.0085 mg/kg/day) and As (1.5 mg/kg/day).

### 2.7. Statistical Analysis

One-way Analyses of variance (ANOVA) and descriptive statistics were carried out using SPSS version 16 while the Duncan Multiple Range Test was used to differentiate significant means at 0.05.

## 3. RESULTS AND DISCUSSION

**Concentrations of heavy metals in Oysters (*Crassostrea gasar*):** The mean concentrations of the heavy metals in the oysters in the different stations are presented in Table 1. Copper ranged from 473.2 – 596.7 mg/kg. The lowest value was recorded in station 1 while the highest was recorded in station 3. Station 1 was significantly ($p < 0.05$) lower than the others and all the values exceeded the limit (3.09 mg/kg) set by (32). The Copper values recorded in the oysters exceeded the limit (3.09 mg/kg) set by (32) by a wide margin. Copper is a critical metal that is easily taken up by aquatic organisms; which could be responsible for relatively high values recorded in the oysters (4). Copper is also a nutritional component of oysters (11) and has been reported to accumulate in oysters with zinc (4, 8).

Lower values were recorded in related studies. Mean values of 11.93 mg/kg (Mundaú/Manguaba lagoon complex) and 14.33 mg/kg (Meirim River) were recorded by (18) in Alagoas, Brazil, 0.97 mg/kg by (19) in Muar River, Johor, Malaysia, 0.34 – 1.16 mg/kg by (4) in Bonny Estuary, Nigeria and 3.80 mg/kg by (11) in Paranaguá Estuarine System, Brazil. The lowest value recorded in Station 1 and the highest in Station 3 could be attributed to anthropogenic activities especially artisanal refining activities (24, 23, 24). Station 1 was located in an abandoned artisanal refinery site while station 3 was located around crude oil and refined product storage site. Station 3 also received discharges from the activities upstream in Stations 1 and 2.

**Table 1:** Mean concentrations of the heavy metals in the oysters (mg/kg).

| Metals | Station 1 | Station 2 | Station 3 | IAEA* |
|---|---|---|---|---|
| Cu | 473.2±61.8[b] | 531.0±81.1[a] | 596.7±69.1[a] | 3.09 |
| Cd | 2.33±0.8[b] | 3.84±0.7[a] | 2.59±27 [b] | 0.18 |
| Zn | 215.44±19.9[a] | 209.02±19.3[a] | 246.41±20.4[a] | 66.4 |
| Pb | 6.16±2.9[a] | 8.51±2.7[ab] | 12.07±2.5[b] | 0.10 |
| As | 0.012±0.002[a] | 0.013±0.002[a] | 0.016±0.002[a] | 13.3 |
| Fe | 1609.0±130.2[b] | 1634.0±132.1[b] | 1846.0±216.4[a] | 146.0 |

* International Atomic Energy Agency (2003, 2022)

Cadmium ranged between 2.33 and 3.84 mg/kg. The lowest value was also recorded in station 1 while the highest was in station 2. Stations 1 and 3 were significantly ($p < 0.05$) lower than station 2 and all the values exceeded the limit (0.18 mg/kg) set by (32). Cadmium values also exceeded the limit (0.18 mg/kg) set by (32). Higher mean values of 4.65 mg/kg (Mundaú/Manguaba lagoon complex) and 4.21 mg/kg (Meirim River) were recorded by (18) in Alagoas, Brazil, while lower values: 1.29 mg/kg was recorded by (19) in Muar River, Johor, Malaysia, 0.005 – 0.040 mg/kg by (4) in Bonny Estuary, Nigeria and 0.16 mg/kg by (11) in Paranaguá Estuarine System, Brazil. The lowest and highest values recorded in stations 1 and 2 could be attributed to anthropogenic activities in the watershed as in Cu.

Zinc ranged from 209.02 – 246.41 mg/kg. The lowest value was recorded in station 2 while the highest was recorded in station 3. There was no significant difference in all the stations and all the values in the stations exceeded the limit (66.4 mg/kg) set by (32). However, Zinc values were higher than 66.4 mg/kg set by (32) with a wide margin and exhibited the same trend as copper because they undergo the same processes (11). Zn is also a natural component of oysters and high concentrations have also been reported with copper (4, 8, 11). Zinc is necessary for good health, but elevated concentrations can be harmful because excessive intake will lead to the suppression of the intake of copper and iron (18).

Higher mean values of 413.58 mg/kg (Mundaú/Manguaba lagoon complex) and 401.43 mg/kg (Meirim River) were recorded by (18) in Alagoas, Brazil, while lower values: 1.02 mg/kg was recorded by (19) in Muar River, Johor, Malaysia, 0.87 – 7.62 mg/kg by (4) in Bonny Estuary, Nigeria and 250.3 mg/kg by (11) in Paranaguá Estuarine System, Brazil. The highest value was also recorded in station 3, though the lowest was in station 2; attributed to anthropogenic impact.

Lead ranged from 6.16 – 12.07 mg/kg. The lowest value was recorded in station 1 while the highest was recorded in station 3. Station 3 was significantly ($p < 0.05$) different from station 1; though all values exceeded the limit (0.10 mg/kg) set by (32). However, Lead values were also higher than the 0.10 mg/kg set by (32). Lower values were recorded elsewhere. 0.52 mg/kg was recorded by (19) in Muar River, Johor, Malaysia and 0.017 – 0.24 mg/kg by (4) in Bonny Estuary, Nigeria. The lowest and highest values were also recorded in stations 1 and 3 respectively as observed in copper.

Arsenic ranged from 0.012– 0.016 mg/kg. The lowest value was also recorded in station 1 while the highest was recorded in station 3. All the values in the stations were within the limit (13.3 mg/kg) set by (32). There was no significant ($p > 0.05$) difference. Arsenic values were within the acceptable limit (13.3 mg/kg) set by (32). (8) recorded a higher value of 0.72 mg/kg in oysters in Hangzhou Bay, China and 0.96 mg/kg by (11) in Paranaguá Estuarine System, Brazil. The lowest and highest values were also recorded in stations 1 and 3 respectively as observed in copper and lead.

Iron ranged between 1609.0 and 1846.0 mg/kg. The lowest value was recorded in station 1 while the highest was recorded in station 3. Stations 1 and 2 were significantly ($p < 0.05$) lower than station 3 but all the values exceeded the limit (146.0 mg/kg) set by (32).

Iron values were higher than the limit (146.0 mg/kg) set by (32) with a wide range. It is the metal that recorded the highest concentration; attributed to anthropogenic impact and environmental stress (9). Lower mean values of 278.06 mg/kg (Mundaú/Manguaba lagoon complex) and 203.18 mg/kg (Meirim River) were recorded by (18) in Alagoas, Brazil, 0.56 mg/kg by (19) in Muar River, Johor, Malaysia and 2.44–227.72 mg/kg by (9) in Pattani Bay, Thailand. The lowest and highest values were also recorded in stations 1 and 3 respectively as observed in copper, lead and Arsenic.

**Health Risk Assessment:** The Target Hazard Quotients (THQs) of the heavy metals evaluated in *C. gasar* are presented in Table 2. All THQs for the heavy metals were less than 1. The THQs for children were generally higher than that of adults in all the metals and stations. The lowest Cd – THQ was recorded among adults (Station 1) while the highest was among children (Station 2) while Pb and Cu - THQs had the lowest values among adults (Station 1) and the highest among children (Station 3). On the other hand, the lowest Zn and Fe – THQs were recorded among adults (Stations 1 and 2) and the highest among children (Station 3). For As, the lowest THQ values were recorded in Station 1 (adult and children), stations 2 and 3 (adults) while the highest values (equal to the reference dose) were recorded among the children in Stations 2 and 3. All HI values were lower than 1; though values among the children were relatively higher and increased spatially from stations 1 to 3 (Table 2).

**Table 2:** Target Hazard quotients (THQs) of the heavy metals in *C. gasar*.

| Heavy metals | Station 1 | | Station 2 | | Station 3 | | Reference Dose |
|---|---|---|---|---|---|---|---|
| | Adult | Children | Adult | Children | Adult | Children | |
| Cd | 1.00E-02* | 1.40E-02 | 1.60E-02 | 2.30E-02** | 1.10E-02 | 1.60E-02 | 4.00E-02 |
| Pb | 7.00E-03* | 9.00E-03 | 9.00E-03 | 1.30E-02 | 1.30E-02 | 1.80E-02** | 1.00E-03 |
| Zn | 3.00E-03* | 4.00E-03 | 3.00E-03* | 4.00E-03 | 4.00E-03 | 5.00E-03** | 3.00E-01 |
| Fe | 1.00E-02* | 1.40E-02 | 1.00E-02* | 1.40E-02 | 1.10E-02 | 1.60E-02** | 4.00E-03 |
| As | 2.00E-04* | 2.00E-04* | 2.00E-04* | 3.00E-04** | 2.00E-04* | 3.00E-04** | 3.00E-04 |
| Cu | 5.10E-02* | 7.00E-02 | 6.00E-02 | 8.00E-02 | 6.00E-02 | 9.00E-02** | 7.00E-01 |
| **ΣTHQ (HI)** | **8.12E-02** | **1.11E-01** | **9.82E-02** | **1.34E-01** | **9.92E-02** | **1.45E-01** | |

**Key:** * = Lowest THQs; ** = Highest THQs

All THQs were less than 1; suggesting that the consumption of oysters from Elechi Creek would not cause any adverse effects (4, 19). However, (11) record THQs greater than 1 in some stations and arsenic; suggesting health risks in consuming the oysters from their study area. THQs should not be overlooked even when they are lower than 1 because cumulative effects could occur when combined with other exposure routes (33). However, when the value is greater or equal to the reference dose, there is a tendency that the population will experience health risks (29).

This trend was observed in Pb and Fe (adults and children in all the stations) and As (children in stations 2 and 3). The THQs for children were generally higher than that of adults in all the metals and stations. This could be attributed to their assimilation level (34). Children have been reported to have high metabolic rates which translates to high assimilation. (8) recorded THQs greater than 1 among children. The higher THQs in Station 3 could be attributed to the anthropogenic activities around the station. All HI values were also lower than 1 in all the stations and both adults and children as observed by (8). With HI < 1, it is unlikely that consumption of the oysters from Elechi Creek will have significant risks to human health (9). However, excessive consumption of oysters should be discouraged to prevent deleterious health risks arising from exposure to multiple heavy metals (9). (34) further reported that frequency of exposure is one of the factors that determine the extent of toxicity of heavy metals. (11) recorded hazard index values greater than one; an indication of potentially-high health risks, which was attributed to zinc and arsenic with high THQs. The TR values were used to assess the carcinogenic human health risks associated with the consumption of oysters from Elechi Creek.

**Carcinogenic Human Health Risk:**

The carcinogenic human health risks resulting from the consumption of oysters collected from Elechi Creek were determined using TR values. The TR values for the three carcinogens evaluated are presented in Table 3. The TR values for Pb and As were within the acceptable limits (1.00E-04 – 1.00E-06) among the adults and children in all the stations while Cd had a value greater than 1.00E-04 among the children in Station 2. Stations 2 and 3 had relatively higher values. The children's values were also higher in all metals and stations as observed in the non-carcinogenic assessment.

**Table 3:** Target Cancer Risk (TR) values of consuming *C. gasar*.

| Heavy metals | Station 1 | | Station 2 | | Station 3 | |
|---|---|---|---|---|---|---|
| | Adult | Children | Adult | Children | Adult | Children |
| Cd | 6.30E-05 | 8.80E-05 | 1.00E-04 | 1.50E-04 | 7.00E-05 | 9.80E-05 |
| Pb | 2.20E-07 | 3.10E-07 | 3.10E-07 | 4.30E-07 | 4.40E-07 | 6.20E-07 |
| As | 7.70E-08 | 1.10E-07 | 8.40E-08 | 1.20E-07 | 1.00E-07 | 1.40E-07 |

The TR values of 1.00E-04 – 1.00E-06 were considered acceptable. However, Cd had a value greater than 1.00E-04 among the children in Station 2; which is unacceptable (14). (9) reported that Cd is of carcinogenic concern among children. The TR values for Pb and As were lower than 1.00E-06 among the adults and children in all the stations; considered negligible (11). Stations 2 and 3 had relatively higher values attributed to anthropogenic impacts. Children were more susceptible in line with previous studies (8, 15).

## 4. CONCLUSION

All the heavy metals were above the acceptable limits except arsenic. Stations 2 and 3 had relatively higher values; attributed to anthropogenic activities. The THQ and HI values were less than 1 in all the metals and stations while TR for Pb and arsenic were within the negligible range in all the stations. TR - Cd was unacceptable among the children in station 2. Station 3 had relatively higher values while the children were more vulnerable to both non-carcinogenic and carcinogenic risks. The consumption of oysters from

Elechi Creek is considered safe based on acceptable levels of the THQ, HI and TR; though Cd-TR for children (Station 2) was unacceptable.

## 5. CONFLICT OF INTEREST

No potential conflicts of interest exist between the authors' authorship and the publishing, they disclose.

## 6. ACKNOWLEDGMENTS

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