

Investigation of Estimated Daily Intake of Toxic Metals and Potential Health Risks in *Oreochromis noliticus* (Tilapia Fish)

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Abstract. *Oreochromis noliticus* (Tilapia fish) is one of the major sources of protein and its readily available in all the markets, Agboyi river is one of the major outlet of Tilapia fish in the southwest Nigeria this was the motivation for this study. Pb, Cr, Mn, Cd, Zn and Ni were identified from *Oreochromis noliticus* (Tilapia fish). Objectives: This study aimed to quantify the level of heavy metals and determine the health risk index (HRI) level using estimated daily intake (EDI) of *Oreochromis noliticus* in samples collected. Methodology: *Oreochromis noliticus* samples were collected (samples D1 to D3) from Agboyi river a fresh water river, and dried then the edible tissues were separated from the other parts of the fish, this was then digested for a Perkin-Elmer model 306 Atomic Absorption spectrophotometer (AAS) for the analysis to ascertain the levels of concentration of the metals identified in the samples. Results: From the samples D1 to D8, for Pb concentrations is the highest in sample D8 with the value of 7.32 ± 0.65 mg/kg and lowest in the sample D6 with the value of 0.02 ± 0.33 mg/kg. The Ni concentration is highest in sample D5 with value 2.72 ± 0.03 mg/kg, lowest in sample D6, D8 and not detectable (ND) in sample D1. Zn concentration is highest in sample D3 with the mean concentration of 62.99 ± 0.05 mg/kg and lowest in sample D6 with a value of 0.74 ± 0.14 mg/kg. Cr mean concentration in D3 and D6 are 8.51 ± 0.54 mg/kg and 0.02 ± 0.63 mg/kg respectively the highest and the lowest in all the samples. Whereas, Mn mean concentration are 9.51 ± 0.42 mg/kg and 0.73 ± 0.07 mg/kg in sample D4 and D7 respectively and this implies that sample D4 has the highest while sample D7 has the lowest values of Mn. Cd has the highest mean concentration values in D1 and the lowest in D6 with values of 5.76 ± 0.66 mg/kg and 0.01 ± 0.68 mg/kg respectively. These values were used to compute the EDI for both children and adults using each sample per heavy metal identified. The EDI for the children is within the range of 0.012–0.638 mg/kgBw/day and for the adults within the range of 0.002–0.193 mg/kgBw/day. The health risk index (HRI) greater than

unity in samples D1 and D3, whereas in other samples is less than unity. The targeted cancer risk (TCR) was estimated in Pb and Ni obtained results were less than USEPA 2010 recommended values. Conclusion: From the obtained results, the HRI obtained from the samples collected did not exceed the standard guideline values, so therefore; the estimated hazard index (HI) suggests that it safe to consume fish from Agboyi River.

KEY WORDS: Toxic metals, Non-carcinogens, Estimated daily intake, Fish, Health risk index, Cancer.

1 Introduction:

Pollution of aquatic environment and ecosystems with heavy metals has become a worldwide problem and of significant concern since metals are indestructible, majorities of them have toxic effects on every living organisms they come in contact with [1–3]. Aquatic lives are exposed to high levels of pollution from metal that comes from various areas such as industrial waste, commercial waste and domestic wastes. These heavy metals enter rivers and lakes from a various sources which include rocks, soils directly exposed to surface water, and in addition to the discharge of various treated and untreated liquid wastes [4,5]. Environmental pollution and aquatic life vulnerability to heavy metal contaminants may primarily be due to exposure to anthropogenic sources, such as agricultural, domestic pesticides, fertilizers, incinerator emissions, municipal or local waste emissions, and including smelting and mining operations [6]. Metals may be absorbed by fish skin through dermal contact or accumulate in the gills through inhaling [7], Fish are sources of protein as a class of food to man. They are widely found in aquatic environment and consumed in every part of the world. Aquatic ecosystems contaminations by heavy metals have been studied and the data bank is increasing with every new studies coming up from different areas. Divesting effects of the heavy metal contamination on aquatic life have found its ways to human body from the food chains. Studies have shown that metal pollution places a leading row in environmental pollution, thus making metals of particular concern due to its toxic effects and also its ability to bioaccumulation in aquatic life. Fish species has been used as bio-indicators in many research carried out on heavy metal pollution in the environment [8, 9]. Fish is low in cholesterol and contains all essential amino acids and is estimated to provide roughly 60% of the world's protein requirements, with 60% of the developing world obtaining more than 30% of their animal protein from fish [10]. Heavy metals can build up in fish and proliferate throughout the food web, causing health problems in humans, such as cardiovascular, renal, and neurological disorders [11]. Accumulation of heavy metals in an aquatic life's have direct consequences on human begin and the ecosystem. These bio indicators are testified in the form of their changes such as biochemical, physiological or behavioral due

to exposure and bioavailability of pollutants that are presence in the surrounding environment [12]. Monitoring contamination of fish allows us to identify any toxic contaminants in fish that may be dangerous to consumers, and proper action can be taken to protect public health and the environment [13]. Therefore, it is significant to study the bioaccumulation of heavy metals in fishes of various types to ensure that heavy metals content are not transmitted to human through consumption of fish [14]. However, in this research work *Clarias gariepinus* (Catfish) and *Tilapia* fish will be used to assess the health risk of contaminating fishes with toxic metals, by identifying and quantifying the heavy metals that may be the present in the Fishes caught from Agboyi River.

2 Material and Methods

2.1 Sampling location

Agboyi River is one of the tributary of Ogun River into Lagos Lagoon then into the Atlantic Ocean. Most of the fishes caught here ends up in Ketu- Lagos one of the major towns with large fish market in Lagos.

There are different fishes caught from the Agboyi River but the two prominent ones are the *Tilapia* fish. Thus Figure 1 shows the sampling location for this work. These types of fishes are generally found in the market and near all the food resultant of this major town in Lagos.

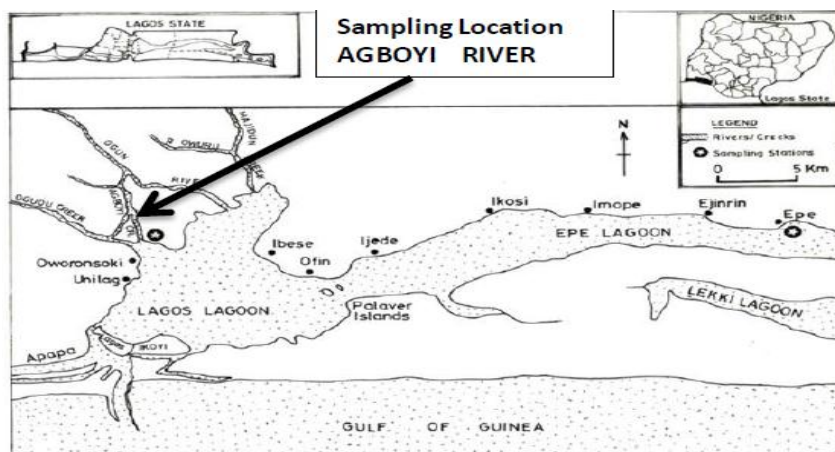


Figure 1. Map showing Agboyi River (Obtained from Lagos State Survey office Ikeja).

2.2 Sample collection

Tilapia fish used in this work were selected based on their popularity in local diets and availability in Agboyi River. Two healthy large sizes adult of each of these fishes (Figure 2a and 2b), per collection points were obtained from the river as indicated in figure1 above designated sampling points. The fishes were collected twice per day, the early morning harvest and the early evening harvest just as the fishermen are getting to the river bank.



Figure 2. Samples *Oreochromis niloticus* (Tilapia fish).

2.3 Sample preparation and measurements

To obtain a representative sample, composites were prepared by taking the edible tissues (fillet) of the three fish samples at each sampling site. The fish samples were oven dried at 105°C, until it reaches constant weight. Each dried sample was then ground into a fine powder using porcelain mortar and pestle, and thereafter all powdered tissues were kept in desiccators prior to further chemical analysis. The powdered fish samples were thoroughly homogenized before subjecting them to digestion and were digested using concentrated nitric acid and hydrogen peroxide (1:1) v/v as recommended by FAO. 1.00 g of dried and powdered fish samples was weighted and transferred into 250 mL round bottled flask and the mixture of 10 mL of concentrated HNO₃ (70%) and 10 mL of H₂O₂ (30%) was added. The flask was covered with a watch glass and left aside until the initial vigorous reactions occur. Then, the samples were heated on a hot plate gradually to 130°C until dissolution the volume reduces to 3–4 mL. Then the samples were allowed to cool, filtered and diluted to 50 mL in volumetric flask with deionized water. The digests were kept in plastic bottles and later analyzed for selected heavy metal using a Perkin-Elmer model 306 Atomic Absorption spectrophotometer (AAS).

2.4 Human health risk assessment in both *Oreochromis nolticus*

The human health risk assessment in fish through metal ingestion can be evaluated using the following parameters: estimated daily intake of metals (EDI), Hazard Quotient (HQ) and the health Risk index (HRI). The EDI of potentially toxic elements (PTE) is directly proportional to the concentrations of PTE in the fish and daily fish consumption. Furthermore, human body weight has an important effect on the tolerance to contaminants [15]. Table 1 shows the tolerable limit of the heavy metals as prescribed by some international regulatory bodies

Table 1. Parameters for the health risk estimations of heavy metals

Parameters	Units	Children	Adults
Average Bodyweight(ABW)	kg	15	65
Exposure frequency (EF)	Days/year	365	365
Exposure duration (ED)	Years	10	70
Ingestion rate (IR)	mg/day	200	100
Average time	Days/year		
For carcinogenic		365×70	365×70
For non-carcinogenic		$365 \times ED$	$365 \times ED$

2.4.1 Estimation of Daily Intake (EDI) of the metals in fish

The EDI of the heavy metals through ingestion by consumption of both the *Oreochromis nolticus* by human being may be estimated using the equation below [16]:

$$EDI = \frac{C \times RI}{ABW},$$

where C is the mean concentration of heavy metals in the samples collected (mg/kg wet weight); RI is the average rate ingestion of metals per person taken to be 100×10^{-3} kg/day for adult and 200×10^{-3} kg/day [17, 18] and ABW is the average body weight of the consumers 200g for adult and 100 g for children.

2.4.2 Target Hazard Quotient (THQ)

Considering the non-carcinogenic, the THQ may be estimated as recommended by USEPA in the relationship below [19]:

$$THQ = \frac{EF \times DI \times RI \times Cf \times C}{ABW \times AT_n \times Rfd} \times 10^{-3}.$$

EF is the exposure frequency (365 days/year), DI is the exposure duration (30 years for non-cancer risk, as used by USEPA [21]), RI is the ingestion rate (75 g/person/day) (BBS, 2011), Cf is the conversion factor (0.208) to convert

fresh weight (FW) to dry weight (DW) given that 79% is the moisture content in fish, C is the heavy metal concentration in fish (mg/kg ww),

ABW is the average body weight (bw) (65 kg) and ATn is the average exposure time for non-carcinogens (EF×DI) (365 days/year for 30 years), as used characterizing no cancer risk [20]. The oral reference dose (RfD) of the metal (is an estimate of the daily exposure to which the human population may be continuously exposed over a lifetime without an appreciable risk of deleterious effects) and ORf mg kg⁻¹day⁻¹ were used for analysis according to USEPA [21, 22].

2.4.3 Health Risk Index (HRI)

HI from THQs is expressed as the sum of the hazard quotients [23]

$$HI = \sum_{1}^{6} THQ_{(K)},$$

where THQ is the Targeted Quotient and K is the individual metal identified in the sample. The recommended values of HRI may be less than unity or greater than unity.

2.4.4 Target Cancer Risk (TCR)

TCR has been used to indicate carcinogenic risks. This is provided by USEPA 2011 [21] risk-based concentration chart.

$$TCR = \frac{EF \times EDI \times RI \times C \times CPSo}{ABW \times ATc} \times 10^{-3},$$

where C is the metal concentration (μg/g), IR is the ingestion rate (g/day), CPSo is the carcinogenic potency slope for oral route (mg/kg bw/day)⁻¹ and ATc is the averaging time of carcinogens.

Table 2. Reference dose and cancer slope for heavy metals

Element	Reference dose RfD (mg/kgBw/day)	Cancer slope factor (mg/kg bw/day) ⁻¹	References
Zn	3 × 10 ⁻¹	—	
Mn	0.046	—	[24]
Pb	3.5 × 10 ⁻³	8.5 × 10 ⁻³	[25]
Ni	2 × 10 ⁻²	9.10 × 10 ⁻¹	[24, 26]
Cd	1 × 10 ⁻³	—	
Cr	3 × 10 ⁻⁴	—	USEPA IRIS [21, 22]

3 Results

The mean concentrations of the identified metal from each sample from D1 to D8 were obtained from the AAS analysis and recorded the results is as illustrated in Table 3 below. From the obtained concentrations of the heavy metals in each of the samples D1 to D8, for Pb concentrations is the highest in sample D8 the value is 7.32 ± 0.65 mg/kg and lowest in the sample D6 with the value of 0.02 ± 0.33 mg/kg.

Table 3. Heavy metals Concentrations in Tilapia fish (mg/kg)

Sample	Concentration of heavy metals in Tilapia fish (mg/kg)					
	Pb	Ni	Zn	Cr	Mn	Cd
D1	0.73 ± 0.21	ND	23.96 ± 0.01	5.47 ± 0.36	3.61 ± 0.99	5.76 ± 0.66
D2	2.43 ± 0.18	1.25 ± 0.11	32.57 ± 0.97	3.12 ± 0.73	7.79 ± 0.87	0.66 ± 0.27
D3	6.79 ± 0.01	0.74 ± 0.82	62.99 ± 0.05	8.51 ± 0.54	2.17 ± 0.10	2.85 ± 0.43
D4	0.58 ± 0.59	2.09 ± 0.91	3.74 ± 0.67	2.86 ± 0.23	9.51 ± 0.42	1.04 ± 0.06
D5	0.39 ± 0.09	2.72 ± 0.03	7.87 ± 0.11	0.33 ± 0.37	8.31 ± 0.77	0.09 ± 0.57
D6	0.02 ± 0.33	0.03 ± 0.72	0.74 ± 0.14	0.02 ± 0.63	0.11 ± 0.82	0.01 ± 0.68
D7	2.16 ± 0.07	ND	0.81 ± 0.83	0.06 ± 0.98	0.73 ± 0.07	0.51 ± 0.64
D8	7.32 ± 0.65	0.03 ± 0.87	25.22 ± 0.11	2.01 ± 0.05	1.06 ± 0.89	4.02 ± 0.45

ND implies Not Detectable

The Ni concentration is highest in sample D5 with value 2.72 ± 0.03 mg/kg, lowest in sample D6, D8 and not detectable in sample D1. For Zn, the highest is in sample D3 with the mean concentration of 62.99 ± 0.05 mg/kg and lowest in sample D6 with a value of 0.74 ± 0.14 mg/kg.

Table 4. Recommended tolerable values of some heavy metals in fish (mg/kg)

Organization	Heavy metals						References
	Pb	Ni	Zn	Cr	Mn	Cd	
UNEP	0.3		5		0.02	0.3	27
IAEA-407	0.12	0.6		0.73		0.18	28
TFC	0.2					0.05	29
DIRECTIVE 2005/78/EC	0.2					0.5	30
FAO/WHO	0.5		5	0.05	5.5	0.5	31

Cr mean concentration in D3 and D6 are of the value 8.51 ± 0.54 mg/kg and 0.02 ± 0.63 mg/kg which are respectively the highest and the lowest in all the samples. Whereas, Mn mean concentration are 9.51 ± 0.42 mg/kg and 0.73 ± 0.07 mg/kg in sample D4 and D7 respectively and this implies that sample D4 has the highest while sample D7 has the lowest values of Mn. Cd has the highest mean concentration values in D1 and the lowest in D6 with values of $5.76 \pm$

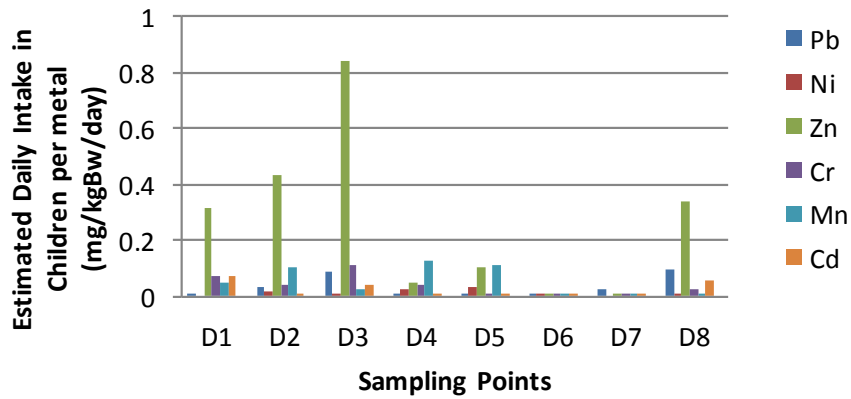


Figure 3. The estimated daily intake of heavy metals in children per sample per metal.

0.66 mg/kg and 0.01 ± 0.68 mg/kg respectively in all the samples. The mean values were compared with the heavy metal recommended tolerable values in fish by different organizations as in Table 4 above. The estimated daily intake of the detected heavy metals in *Oreochromis niloticus* was analyzed for the health impact in both the adults and the children using the relationship in Section 2.4.1. The obtained results are as illustrated in Figure 3 for the children and Figure 4 for adults.

Figure 3 above shows that Children consuming 200 mg of fish per day will have estimated daily intake of each metal detected from the sample D1 to D8. Zn intake per day in Children is the highest in samples D3, D2, D8 and D1, while Cr in take per day is the highest in samples D3, D1 D2 and D4. While that of the adults is also illustrated in Figure 4 below. The adults are assumed to consume

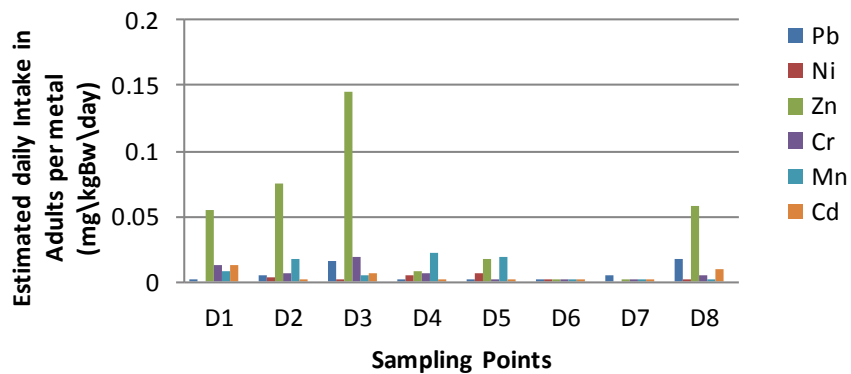


Figure 4. The estimated daily intake of heavy metals in adults per sample per metal.

Investigation of Estimated Daily Intake of Toxic Metals and ...

100mg of fish per day, thus the estimated heavy metal intake per day per metal in each sample for D1 to D8.

The comparison between the total EDI for the children and adults are illustrated in Table 5, In general Children have more total daily intake than the adults in each of the samples collected. These values of total EDI will used to analyze the health risk factor in each of the samples.

Table 5. Comparison of health impact parameters of heavy metals in Adults and children

Sample	Total EDI (mg/(kgBw/day))		Total THQ	
	Adults	Children	Adults	Children
D1	0.091	0.527	0.835	1.014
D2	0.111	0.638	0.414	0.689
D3	0.193	1.121	1.047	1.056
D4	0.046	0.264	0.379	0.641
D5	0.045	0.263	0.056	0.444
D6	0.002	0.012	0.003	0.01
D7	0.099	0.057	0.046	0.158
D8	0.091	0.529	0.044	0.935

Carcinogenic risks estimated may defined as the incremental probability of an individual to develop cancer over a lifetime exposure to potential carcinogens [21] these values were estimated based on the relationship in 2.4.4 as provided by USEPA 2011 report [21] on risk based chat for selected metals. Thus, the target cancer risk TCR has been used to indicate carcinogenic risks in both the adults and the children as illustrated in Table 6 below.

Table 6. Comparison of TCR in adults and children from Pb and Ni

Samples	Target Cancer Risk			
	Pb		Ni	
	Adults	Children	Adults	Children
D1	9.58E-10	5.81E-08	0	0
D2	3.86E-09	2.34E-07	1.99E-09	1.2E-07
D3	1.90E-08	1.15E-06	2.07E-09	1.25E-07
D4	3.82E-10	2.32E-08	1.38E-09	8.35E-08
D5	2.55E-10	1.55E-08	1.78E-09	1.08E-07
D6	6.18E-13	3.75E-11	9.27E-13	5.62E-11
D7	3.06E-10	1.86E-08	0	0
D8	9.64E-09	5.85E-07	3.95E-11	2.4E-09

4 Discussions

The results of this work indicates that the mean concentrations of each of the heavy metal detected are for Pb the mean concentrations is in the range of 7.32 to 0.02 mg/kg; Ni with mean concentrations in the range of 2.72 to ND; for Zn the range is 25.22 to 0.81 mg/kg; Cr with the range of 8.51 to 0.02 mg/kg; Mn has the mean concentrations of 9.51 to 0.11 mg/kg and Cd with the range of 5.76 to 0.01 mg/kg. The daily recommended tolerable values of heavy metals in fish as show in Table ?? indicates that nearly of the samples D1 to D8 have exceeded the recommended values. All of the heavy metals in samples D1 to D8 exceeded the recommended tolerable values by FAO/WHO or IAEA. In estimating the daily intake of these heavy metals in fish in every dish of an adult and children, it was assumed that the children will consumed 200 g while the adult 100 g per day. With this we obtained the total EDI for both the children and the adults as indicated in Table 5. From all the samples D1 to D8, the total EDI in children range from 0.012 to 1.121 mg/kgBw/day this is higher than that of the adult which is in the range 0.002 to 0.193 mg/kgBw/day and that is in each of the samples, indicating that children may be at higher risk of metal poisoning in fish harvested from Agboyi river which is the main source of fishing in the locality and most of these fishes are taken to the larger market away from the area. Health risks assessment of the samples we analyzed the total THQ obtained from each of the samples as indicated in Table 5 show that the total targeted quotient is less than unity in all the samples expect samples D3 for both the adults and the children, so also sample D1 for the children which implies that the HRI greater than one the D1 and D3 resulting in higher risk of heavy metal poisoning in those samples. While other samples THQ values are less than unity indicating that they may be less health risk to both the adults and children that consumed the fish from the sampling areas. The TCR values for Pb and Ni were estimated based on available carcinogenic potency slope factors from USEPA risk based report; hence for samples D1 to D8 is in the range of 1.90×10^{-08} to 6.18×10^{-13} in adults and for children in the range 1.15×10^{-06} to 3.75×10^{-11} from Pb. While from Ni in samples D1 to D8 is in the range of $0-9.27 \times 10^{-13}$ in adults and in the range of 0 to 5.62×10^{-11} . As report in USEPA 2010 analysis, values of TCR lower than 10^{-6} is negligible and greater than 10^{-4} is unacceptable thus, from this study the TCR values of Pb and Ni were lower than 10^{-6} hence they are considered negligible.

5 Conclusions

In this study Pb, Ni, Cr, Zn, Cd, and Mn, have been identified in *Oreochromis noliticus* (Tilapia fish). The EDI in both children and adults were computed (as illustrated in Figures 3 and 4) in each of the samples collected and the results show the EDI of the children in each sample as follows:

Investigation of Estimated Daily Intake of Toxic Metals and ...

- D1: Zn >Cd >Cr >Mn >Pb >Ni;
- D2: Zn >Mn >Cr >Pb >Ni >Cd;
- D3: Zn >Cr >Pb >Cd >Mn >Ni;
- D4: Mn >Zn >Cr >Ni >Cd >Pb;
- D5: Mn >Zn >Ni >Pb >Cd >Cr;
- D6: Zn >Pb >Cr >Cd >Mn >Ni;
- D7: Pb >Zn >Cr >Mn >Cd >Ni;
- D8: Zn >Pb >Cd >Cr >Mn >Ni.

This indicates the level of heavy metals intakes in each of the samples as the children consumes 100 grams of Tilapia fish per day. For the adults the trends is the same but with an increase in amount consumed by 100 grams that it the adult will consume 200grams of Tilapia fish per day from each of the samples. However, the THQ values were less than unity indicating that the health risk may not be significant immediately on both the children and the adults that consumed the Tilapia fish from the sampling areas. Nevertheless; the long time effects may post health risks due to heavy metal bioaccumulation in both the children and adults as indicated in the samples of the fish. HRI >1 indicates the level is hazardous as can be observed in some of the samples D1 and D3 whereas in other samples HRI <1 implies less risk in the consumption of the Tilapia fish. Values of TCR due to exposure children and adults to Pb and Ni the values obtained from this study were small and therefore we can conclude that it is safe to take *Oreochromis noliticus* (Tilapia fish) from Agboyi River in Southwest Nigeria

6 Recommendations

It is recommended that the consumptions of *Oreochromis noliticus* (Tilapia fish) from Agboyi River in Southwest Nigeria are safe. Nevertheless; periodic study of the Agboyi River Tilapia fish recommended and more sampling collections is needed in the area.

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