

Evaluation of Water Quality and Heavy Metal Pollution in the Shoots and Roots of Aquatic Plants

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Hazard Index (HI)

Target Cancer Risk (TR)

Abstract

The safety of vegetables has been an issue of public concern in recent years. Thus, heavy metals (Cu, Cd, Mn, Pb, Zn and Fe) in the shoot and root of *Sacciolepis africana*, *Ceratophyllum demersum* and *Pistia stratiotes* in Owena reservoir, Nigeria were tested using Atomic Absorption Spectrophotometer while some Physico-chemical parameters of water were measured using multi parameter hand-held meter. The water quality result revealed that the average temperature, turbidity, conductivity, dissolved oxygen and pH were 27.77°C, 7.21NTU, 101.83Ω/S, 6.95mg/l and 6.55 respectively. The single factor pollution index showed that Temperature (0.93), DO (0.99) and pH (0.99) fell within the category of slight pollution while turbidity (1.20) and conductivity (1.02) fell within the medium pollution category. *Sacciolepis africana* bio-concentrated more heavy metals than *Ceratophyllum demersum* and *Pistia stratiotes*. The HRIs of Cu, Cd, Mn, Zn and Fe indicated no risk since all values were less than 1. However, there was a potential risk for Pb through the consumption of the plants despite that the HRI of the shoot of *Ceratophyllum demersum* (0.65) and root of *Pistia stratiotes* (0.46) were less than 1. The THQ and HI values for the root and shoot of all the vegetables were less than 1, indicating that they are unlikely to result in any chronic systemic risk-averse health effects.

Introduction

Vegetables are herbaceous plants whose parts are eaten as supporting food or main dishes and they may be aromatic, bitter or tasteless (Adedokun et al., 2016). A vegetable is a major part of the Nigerian diet and is very susceptible to environmental pollution due to the activities being practised in the area of cultivation. Plants absorb metals through their foliage or roots (Adesuyi et al., 2015). Heavy metals such as zinc (Zn), iron (Fe), copper (Cu), and manganese (Mn) are essential in plant nutrition; however, they often do not

significantly affect the plants' physiology. Heavy metals can have a very deleterious effect on the human body even in low concentrations because there is no effective mechanism for its excretion (Adedokun et al., 2016).

Nigeria has a variety of traditional vegetables that are consumed by various ethnic groups for different reasons. Humans are encouraged to consume more vegetables and fruits because they are good sources of fiber, vitamins and minerals (Adesuyi et al., 2015). However, plants contain both essential and toxic metals over a wide range of concentrations. The safety of vegetables and other food crops have been an issue of

public concern in recent years (Gyawali et al., 2011). It is, therefore, necessary to constantly quantify the water quality, heavy metals, and the safety of consumers of aquatic plant (Adesuyi et al., 2015 & 2016; Adedokun et al., 2016; Olawusi-Peters et al., 2019). Several studies have assessed the potential health risks associated with exposure to heavy metals through the consumption of vegetables (Cao et al., 2010; Wang et al., 2011; Ghosh et al., 2012). However, only very few studies have been done on the evaluation of potential health risks of heavy metals in consumers of aquatic plants. Therefore, this work determined some physicochemical parameters as well as the contamination levels of Cd, Cu, Zn, Fe, Mn and Pb in the three most available plant species (*Sacciolepis africana*, *Ceratophyllum demersum* and *Pistia stratiotes*) in the Owena reservoir for six months, and evaluated the health risk to the consumers of the plants as vegetables or herbs.

Materials and Methods

Study Area

This study was done from April to September 2017 in Owena reservoir of Ondo State, Nigeria which is

positioned between latitude $7^{\circ} 15'N$, Longitude $5^{\circ} 5'E$ and latitude $7^{\circ} 4'N$, longitude $4^{\circ} 47'E$ in western Nigeria (Figure 1). Owena Reservoir was formed as a result of taming the Owena River, a major River in Ondo State (Olaniyan, 2013). The reservoir is about 300m long and 9m in its deepest part with the capacity of approximately $600,000m^3$ and the catchments area controlled by the reservoir is $790km^2$.

Collection and Analysis of Water

Water samples were collected at a depth of approximately 30 cm below the water surface from the Owena reservoir. Water samples were collected into 20mL ultraclean HDPE (High-Density Polyethylene) bottles. The sampling bottles were rinsed with reservoir waters before the original samples were taken. The water samples were kept in clean stoppered plastic bottles and then kept refrigerated and transferred in an icebox to the laboratory for analysis (Ajibare et al., 2019). The temperature, pH, conductivity, turbidity and dissolved oxygen were measured using Hanna multi parameter hand-held meter (Model HI9828). Concentrations of studied parameters were determined according to the procedures laid down in APHA (1995).

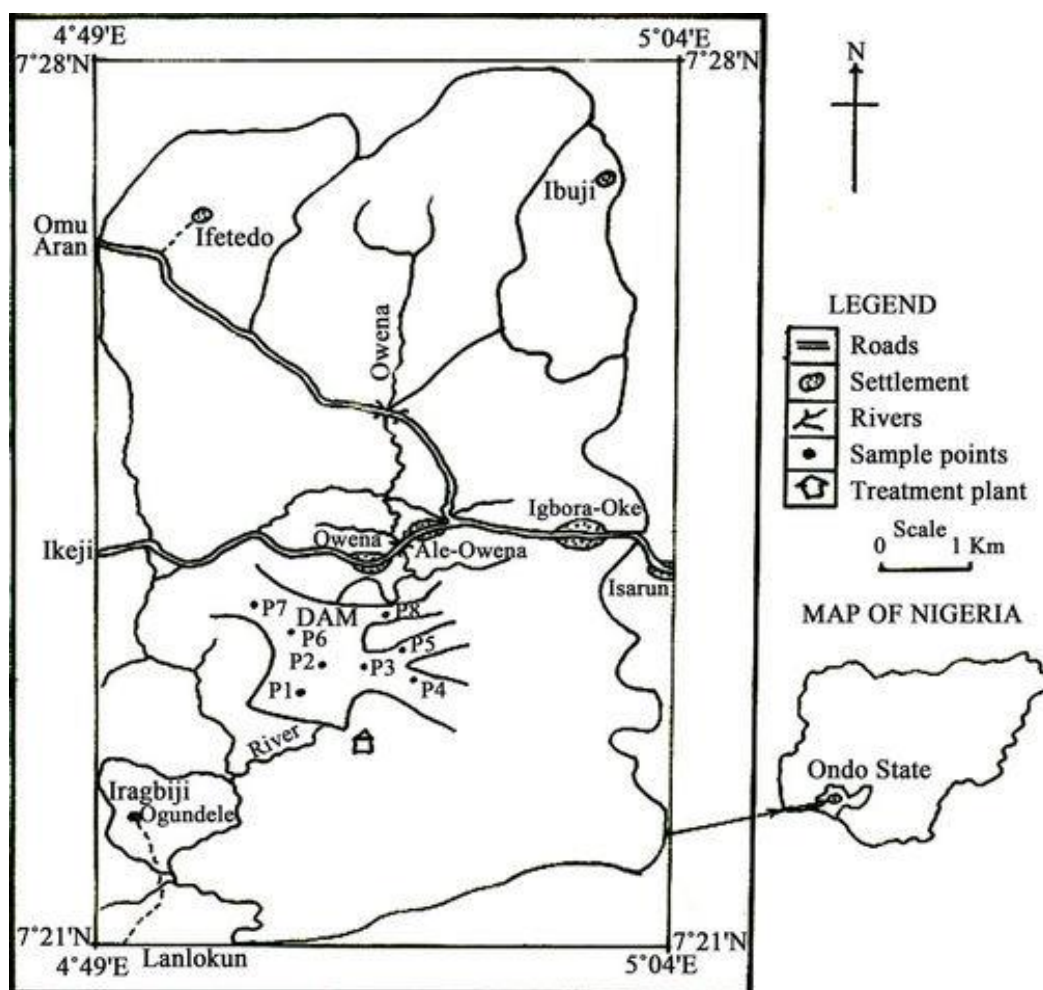


Figure 1. Map showing the study area.

The single-factor pollution index and comprehensive pollution index were used to determine the pollution status of the reservoir as follow:

- i. The single-factor pollution index was calculated as:

$$P_i = \frac{C_i}{S_i} \text{ (Yan et al., 2015)}$$

Where: P_i is the pollution index of parameter i ,
 C_i is the concentration of the parameter in water (mg/l)

S_i is the permissible limit for the parameter in water.

The single-factor pollution index (P_i) is classified into five grades, according to Li et al. (2010) and Ajibare et al., (2019) (Table 1).

- ii. The comprehensive pollution index (CPI) was calculated as:

$$CPI = \frac{1}{n} \sum_{i=1}^n \frac{C_i}{S_i} \text{ (Tao et al., 2011)}$$

Where: CPI = the comprehensive pollution index,

C_i = concentration of the parameter i (mg/l),

S_i = permissible limit for the parameter i in water

n = the number of analysed parameters.

CPI is classified according to Tao et al. (2011) into five water quality levels (Table 2).

Collection and Preparation of Plant Samples

Fresh samples of the three most available plants (*Sacciolepis africana*, *Ceratophyllum demersum* and *Pistia stratiotes*) were obtained from the Owena reservoir. The freshly collected vegetables were thoroughly washed with distilled water to remove any attachment (such as; soil, dust particles, unicellular organisms, etc.). They were dried with blotting paper and then by filter paper at room temperature to remove

surface water. The samples were properly tagged according to species in polythene bags and taken to the herbarium of CSP Department, of the Federal University of Technology, Akure for identification, and subsequently taken to the laboratory for heavy metal analyses. The root and shoot of each vegetable sample were used for analysis.

Estimation of Heavy Metals

The plants were chopped into small pieces and oven-dried at (55±1°C). They were then crushed into a fine powder using a porcelain mortar and pestle. The resulting powder was kept in an airtight polythene packet at 26±1°C before being digested. 0.5g of each sample was measured into a clean dried beaker (100ml), 10ml of the acidic mixture of HNO₃/HClO₄ in ratio 2:1 was then added to the sample for digestion (Olawusi-Peters et al., 2019). The samples were stirred with a glass rod so that they will be evenly distributed in the acid; the beaker was then placed on the digestion block in a fume cupboard for 2 hours at 150°C for digestion. The digested samples were then filtered into a 25ml volumetric flask and made to mark with deionised water. The digested samples were kept at 4°C before analysis (Adedokun et al., 2016). A Buck scientific atomic absorption spectrophotometer (Model 210VGP) was used for the determination of Iron (Fe), zinc (Zn), lead (Pb), cadmium (Cd), copper (Cu) and Manganese (Mn) (Olawusi-Peters et al., 2017).

Health Risk Assessment

The potential health risks of heavy metal through the consumption of analysed plants were assessed based on:

Daily Intake of Metals (DIM) was calculated to averagely estimate the daily metal loading into the body system of specified bodyweight of a consumer. This informed the relative Phyto-availability of metal.

Table 1. Standards for single-factor pollution index (P_i)

Pi	Pollution grades
<0.4	Non-pollution
0.4 - 1.0	Slight pollution
1.0 - 2.0	Medium polluted
2.0 - 5.0	Heavy polluted
>5.0	Serious polluted

Table 2. Classification of surface water quality based on CPI

Values	Water Quality Grades
<0.2	Cleanness
0.21 - 0.4	Sub-cleanness
0.41 - 1.0	Slight pollution
1.01 - 2.0	Moderate pollution
>2.01	Severe pollution

$$DIM = \frac{C_{\text{metal}} \times C_{\text{factor}} \times C_{\text{food intake}}}{\text{Average body weight}} \quad (\text{Omobepade et al., 2020})$$

Where C_{metal} is the heavy metal conc. in vegetables (mg/kg), C_{factor} is the conversion factor (0.085); $C_{\text{food intake}}$ is the daily intake of vegetables (65g/day) (Adedokun et al., 2016) while the average body weight used was 60kg for this study (Olawusi-Peters et al., 2019).

Health Risk Index (HRI) was calculated to give quantitative information on the risk posed by each contaminant to the health of the fish consumers

$$HRI = \frac{\text{Daily Intake of Metal (DIM)}}{\text{Reference Oral Dose (RfD)}} \quad (\text{Olawusi-Peters and Adejugalbe, 2020})$$

Where: Reference Oral Doses (RfD) is 0.040, 0.300, 0.700, 0.004, 0.001, and 0.140mg/kg/day for Cu, Zn, Fe, Pb, Cd, and Mn respectively (USEPA, 2017).

Target Hazard Quotient (THQ)

THQ was calculated to define the exposure duration and the non-carcinogenic risk within the period

$$THQ = \frac{EFr \times ED \times FIR \times M}{RfD \times BW \times ATn} \times 10^{-3} \quad (\text{Orosun et al., 2016})$$

Where; EFr is the exposure frequency (350 days/year); ED is the exposure duration (54 years, equivalent to the average life expectancy of the Nigerian population); FIR is the food ingestion rate (vegetable consumption values for southwestern adult Nigerian is 65g/person/day) (Adedokun et al., 2016); M is the metal concentration in the edible parts of vegetable (mg/kg); RfD is the oral reference dose; BW is the average body weight and ATn is the average exposure time for non-carcinogens (19710) (USEPA, 2017).

Note: If the THQ value is greater than 1 (i.e. $THQ > 1$), the exposure is likely to cause obvious adverse effects.

Hazard Index (HI)

The hazard index (HI) was calculated as:

$$HI = \sum_{n=1}^n THQ \quad (\text{Núñez et al., 2018})$$

Where; n=number of heavy metals examined, THQ= unit THQ of *ith* heavy metal

Target Cancer Risk (TR)

Target cancer risk (TR) was calculated to reveal the carcinogenic risk of the plants

$$TR = \frac{M \times FIR \times CPSo \times EF \times ED}{Bw \times ATn} \times 10^{-3} \quad (\text{USEPA, 2017})$$

Where; M, FIR, EF, ED, BW and ATn have already explained above. CPSo is the carcinogenic potency slope oral (mg/kg bw-day⁻¹). CPSo has not yet been established for Mn, Fe, Cu and Zn since they do not cause any carcinogenic effects (USEPA, 2017). Hence, TR values for the intake of only Pb and Cd were calculated to determine the carcinogenic risk. Their carcinogenic potency slope oral (CPSo) as obtained from the integrated risk information system database (USEPA, 2017) is given as 0.009 and 0.6 respectively. According to USEPA (2017) the TR categories are described as, if $TR \leq 10^{-6}$ = Low; 10^{-4} to 10^{-3} = moderate; 10^{-3} to 10^{-1} = high; $\geq 10^{-1}$ = very high.

Results

Physico-chemical Properties of Water

The result of the Physico-chemical properties of water is presented in Figure 2. The figure revealed that

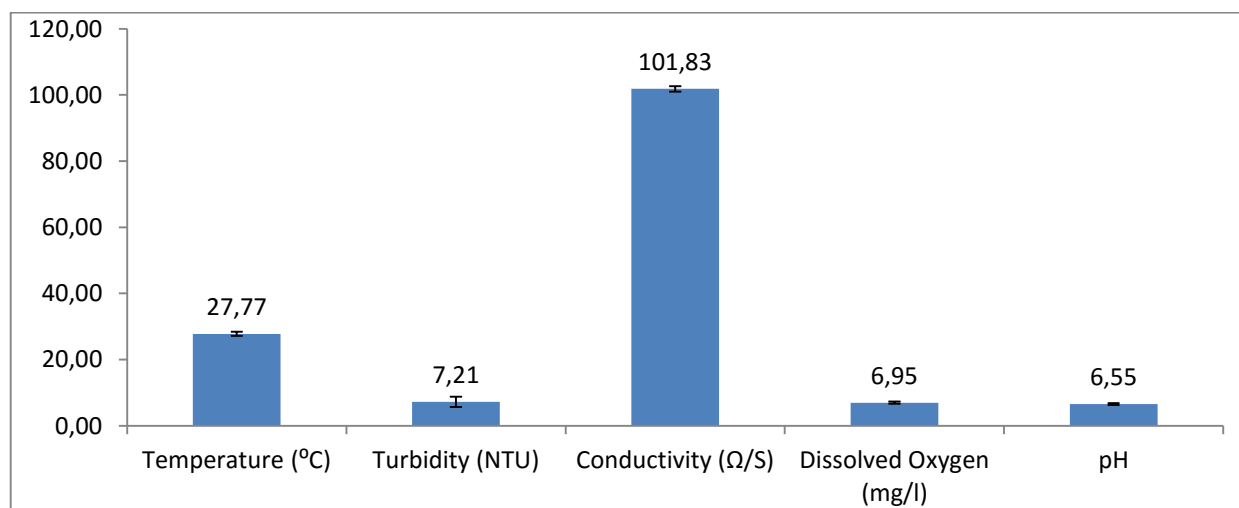


Figure 2. Physico-chemical properties of Owena reservoir Ondo State, Nigeria

the average Temperature, Turbidity, Conductivity, Dissolved Oxygen, and pH were 27.77°C, 7.21NTU, 101.83Ω/S, 6.95mg/l and 6.55 respectively. Also, the single factor pollution index and comprehensive pollution index (CPI) of Owena reservoir during this study is presented in Table 3 which shows that Temperature (0.93), Dissolved Oxygen (0.99) and pH (0.99) fell within the category of slight pollution while turbidity (1.20) and conductivity (1.02) fell within the medium pollution category. The comprehensive pollution index (1.03) revealed that the reservoir was moderately polluted.

Heavy Metals in the Plants

The concentration of heavy metals in the root and shoot of plants inhabiting the Owena reservoir is presented in Table 4. The table shows that the root of *Sacciolepis africana* (0.0042±0.00) bio-accumulated Cd significantly more than *Ceratophyllum demersum* (0.0027±0.00) and *Pistia stratiotes* (0.0032±0.00) while there was no significant difference ($P>0.05$) between the concentration of Cd recorded in the root of *Sacciolepis africana* (0.0047±0.00) and *Pistia stratiotes* (0.0036±0.00). A similar trend was observed in the root and shoot of the three plants for Mn, Cu and Pb. The table also revealed that no significant difference was observed in the concentrations recorded for the root of

the three plants while *Sacciolepis africana* had a significantly higher concentration in the shoot.

The Daily intake of metals (DIM) and Health Risk Index (HRI) of the consumers of the studied plant species is presented in Tables 5 and 6 respectively. The DIM for Cd, Zn, Fe and Cu were less than the RfD while Mn and Pb were slightly higher. The HRIs of Cd, Cu, Fe, Mn and Zn were all less than 1, which showed there was no risk of Cd, Cu, Fe, Mn and Zn in the root and shoot of the plants' species. The table further showed that there was a potential risk for Pb through the consumption of the plants despite that the HRI of the shoot of *Ceratophyllum demersum* (0.65) and root of *Pistia stratiotes* (0.46) were less than 1.

The THQ values of Cd, Pb, Zn, Mn, Fe and Cu due to vegetable consumption for the populace of the study area are listed in Table 7. The THQ values in the root and shoot showed that all the values were less than 1 (THQ<1). Similarly, the HI (Figure 3) for all the vegetables were less than 1 (HI<1) indicating that they are unlikely to result in any chronic systemic risk-averse health effects during a lifetime of exposure and would therefore be considered as acceptable. Moreover, the target cancer risk (TR) of the aquatic plants inhabiting the Owena reservoir is presented in Table 8 which shows that there was a low carcinogenic risk for all the plants since the TR values were less or equal to 10^{-6} (TR≤ 10^{-6}).

Table 3. Single-Factor Pollution Index and Comprehensive Pollution Index (CPI) of Owena reservoir Ondo State, Nigeria

Parameter	Single Factor Pollution Index	Interpretation
Temperature (°C)	0.93	Slight Pollution
Turbidity (NTU)	1.20	Medium Pollution
Conductivity (Ω/S)	1.02	Medium Pollution
Dissolved Oxygen (mg/l)	0.99	Slight Pollution
pH	0.99	Slight Pollution
Comprehensive Pollution Index	1.03	Moderate Pollution

Table 4. Heavy Metals in the Plants of Owena reservoir Ondo State, Nigeria

Part	Metal	<i>Ceratophyllum demersum</i>	<i>Pistia stratiotes</i>	<i>Sacciolepis africana</i>
Root	Cd (mg/kg)	0.0027±0.00 ^a	0.0032±0.00 ^a	0.0042±0.00 ^b
	Mn (mg/kg)	0.2209±0.11 ^a	0.2851±0.10 ^a	0.4471±0.19 ^b
	Cu (mg/kg)	0.3243±0.11 ^a	0.3717±0.08 ^a	0.4351±0.08 ^b
	Pb (mg/kg)	0.0808±0.03 ^c	0.0174±0.01 ^a	0.0556±0.04 ^b
	Fe (mg/kg)	0.7104±0.37 ^a	0.5882±0.29 ^a	2.2238±1.55 ^b
	Zn (mg/kg)	0.4740±0.22 ^a	0.3827±0.32 ^a	0.4766±0.31 ^a
Shoot	Cd (mg/kg)	0.0025±0.00 ^a	0.0036±0.00 ^b	0.0047±0.00 ^b
	Mn (mg/kg)	0.1319±0.07 ^a	0.2951±0.12 ^b	0.3145±0.10 ^b
	Cu (mg/kg)	0.2213±0.08 ^a	0.3615±0.10 ^b	0.4055±0.13 ^b
	Pb (mg/kg)	0.0247±0.01 ^a	0.1290±0.14 ^b	0.0468±0.03 ^b
	Fe (mg/kg)	0.9826±0.73 ^a	0.8306±0.57 ^a	2.5960±1.11 ^b
	Zn (mg/kg)	0.2234±0.10 ^a	0.2984±0.09 ^a	0.4853±0.18 ^b

Mean in the same row with the same superscripts are not significantly different ($P>0.05$)

Table 5. Daily intake of Metal (DIM) of Aquatic Plants inhabiting Owena Reservoir Ondo State, Nigeria

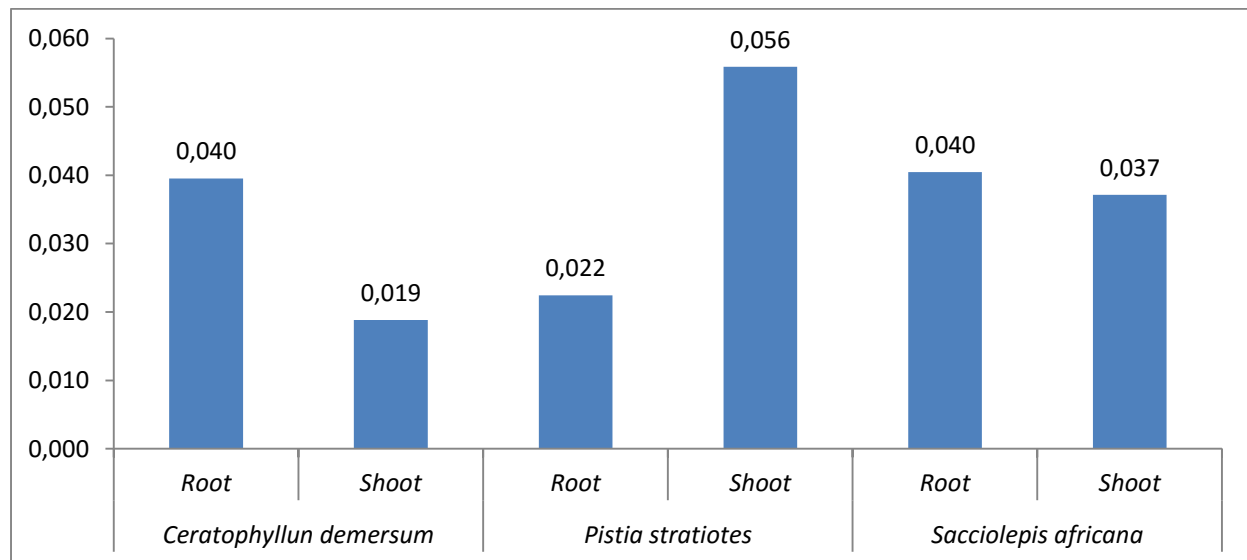
Metal	<i>Ceratophyllum demersum</i>		<i>Pistia stratiotes</i>		<i>Sacciolepis africana</i>	
	Root	Shoot	Root	Shoot	Root	Shoot
Cd	0.000	0.000	0.000	0.000	0.000	0.000
Mn	0.020	0.012	0.026	0.027	0.041	0.029
Cu	0.030	0.020	0.034	0.033	0.040	0.037
Pb	0.007	0.002	0.002	0.012	0.005	0.004
Fe	0.065	0.090	0.054	0.076	0.205	0.239
Zn	0.044	0.021	0.035	0.027	0.044	0.045

Table 6. Health Risks Index (HRI) of Aquatic Plants inhabiting Owena Reservoir Ondo State, Nigeria

Metal	<i>Ceratophyllum demersum</i>		<i>Pistia stratiotes</i>		<i>Sacciolepis africana</i>	
	Root	Shoot	Root	Shoot	Root	Shoot
Cd	0.25	0.23	0.29	0.33	0.39	0.43
Mn	0.15	0.09	0.19	0.19	0.29	0.21
Cu	0.75	0.51	0.86	0.83	1.00	0.93
Pb	2.13	0.65	0.46	3.39	1.46	1.23
Fe	0.09	0.13	0.08	0.11	0.29	0.34
Zn	0.15	0.07	0.12	0.09	0.15	0.15

Table 7. Target Hazard Quotient (THQ) of Aquatic Plants inhabiting Owena Reservoir Ondo State, Nigeria

Metal	<i>Ceratophyllum demersum</i>		<i>Pistia stratiotes</i>		<i>Sacciolepis africana</i>	
	Root	Shoot	Root	Shoot	Root	Shoot
Cd	0.003	0.003	0.003	0.004	0.004	0.005
Mn	0.002	0.001	0.002	0.002	0.003	0.002
Cu	0.008	0.006	0.010	0.009	0.011	0.011
Pb	0.024	0.007	0.005	0.038	0.016	0.014
Fe	0.001	0.001	0.001	0.001	0.003	0.004
Zn	0.002	0.001	0.001	0.001	0.002	0.002

**Figure 3.** Hazard Index (HI) of Aquatic Plants inhabiting Owena Reservoir Ondo State, Nigeria**Table 8.** Target Cancer Risk (TR) of Aquatic Plants inhabiting Owena Reservoir Ondo State, Nigeria

Metal	<i>Ceratophyllum demersum</i>		<i>Pistia stratiotes</i>		<i>Sacciolepis africana</i>	
	ROOT	SHOOT	ROOT	SHOOT	ROOT	SHOOT
Cd	1.666E-06	1.531E-06	1.974E-06	2.251E-06	2.63E-06	2.909E-06
Pb	7.557E-07	2.306E-07	1.626E-07	1.206E-06	5.19E-07	4.375E-07

Discussion

The determined physical and chemical variables affecting the metabolic rate of living organisms in the aquatic environment were within the optimal limits that have been previously documented for optimum production and growth of planktons and fish in freshwater bodies (Ajibare et al., 2019; Olawusi-Peters and Akinola, 2018). The conductivity and turbidity values indicated medium pollution and greatly influenced the quality of the reservoir. This could be linked to the presence of suspended particles due to discharges, runoffs and other human-mediated activities such as agriculture in and around the study area (Ajibare et al., 2019).

The metal concentrations observed in the plant samples (both root and shoot) were all within the permissible limit. For the concentrations of Cd in the plants, the mean values of the three species were 0.0025-0.0047mg/kg, which falls within the range observed in Jiangsu, China (Cao et al., 2010) and Gejiu City, Yunnan Province, China (Xiao et al., 2011). Also, the range of Pb concentrations (0.0174-0.1290mg/kg) in edible portions of vegetables was comparably lower than the values observed by Cao et al., (2010) and Asdeo & Loonker (2011), where the concentrations of Pb in vegetables were within the range of 2.32 – 5.76mg/kg.

The daily intake of heavy metals depends on the number of heavy metals in the edible portions of vegetables as well as the associated consumption rate. Moreover, body weight can influence the tolerance of pollutants. The DIMs of Mn and Pb were slightly above the RfDs while the DIMs of Cu, Fe, Zn and Cd were all far below the RfDs. This suggests that the consumption of vegetables posed a very negligible health risk to consumers. The HRI for Cd, Mn, Zn, Fe and Cu from this study were far lesser than 1 (HRI<1). Generally, HRI<1 means that the exposed population is safe of metals health risk while HRI>1 means otherwise (Olawusi-Peters et al., 2019). The consuming population is therefore not at any risk of Cd, Mn, Zn, Fe and Cu. However, Pb revealed potential health risks to consumers. Lead (Pb) particularly has impacts on children. It influences the nervous system, thereby slowing down the nervous response. This, in turn, influences learning abilities and behaviour (Omobepade et al., 2020).

The THQ is a ratio between the measured concentration and the oral reference dose, weighted by the length and frequency of exposure; amount ingested and body weight (Adedokun et al., 2016). In this study, the THQ of all examined metals was far less than 1 in all the plant species; hence, there is no health risk concern for their consumption. Higher THQ for Cd and Pb was reported by Zhou et al. (2016) in vegetable species planted in contaminated soils in Nanning, China. Similarly, the target cancer risk (TR) indicated that there was no carcinogenic risk for the consumers of the plants.

However, for special populations, such as pregnant women and allergic/sensitive individuals, the potential health risks of heavy metal via consumption of vegetables were likely to be higher. Also, since vegetables only accounted for some proportion of human diets, and other foods (such as seafood, grains and livestock) might contain heavy metals, (which could significantly increase the risk of ingestion of heavy metals), the potential health risks for the consumers of the studied plants may be higher than the results from this study. Therefore, considerable attention should be given to the health risk of heavy metals through several media.

Conclusion

This study showed that Owena reservoir is moderately polluted because the turbidity and conductivity of the reservoir had a medium deviation from the normal. The pollution of the water has also influenced the plants inhabiting the reservoir to have bio-concentrated Zn, Cu, Cd, Mn, Fe and Pb in concentrations below the permissible limits. The HRI observed in this study revealed that despite the low concentrations of the metals in the plants, there is a potential health risk of Pb through the consumption of *Ceratophyllum demersum*, *Pistia stratiotes* and *Sacciolepis africana* inhabiting the reservoir. THQs and HI in all the plants were far less than 1 in all the vegetable species. The TR also revealed that there was no carcinogenic risk in the consumption of the plants. However, the potential health risks for the consumers might be higher, if all other routes of entry of heavy metal are considered. It is therefore recommended that the health risks of other foods such as cereals, fishes, legumes, and cassava should be determined since vegetables only accounted for a part of human diets.

Ethical Statement

Not applicable.

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Author Contribution

This manuscript is a sole author manuscript. The author conceived, designed and performed the experiments; analyzed and interpreted the data; contributed reagents and materials, analysis tools or data, prepared, edited and reviewed the manuscript.

Conflict of Interest

The authors declare no conflict of interest.

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