Bioaccumulation of zinc, lead, cadmium in water hyacinth, hippo grass and papyrus reed as water quality indicator in River Kisat in Kisumu County, Kenya.

Mareri B.^{1*}, Kitur E.¹, Obade P.¹

^{1*,1,1} Department of Environmental Sciences and Education, Kenyatta University,
P.O Box 43844-0100, Nairobi, Kenya.

*Corresponding author's email: brenda.mareri@gmail.com

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ABSTRACT

River Kisat in Kenya is increasingly being polluted serving as a sink for waste receiving effluent from sewerage, industrial waste and agricultural pesticide residues leading to heavy metal contamination. This has led to the contamination of the aquatic ecosystem and water used by communities for consumption and farming. Little information is available on the bio-indicative aspects of water quality using bioaccumulation factor by macrophytes. This study aimed at determining concentrations of Zn, Pb and Cd in water, *Eichhornia crassipes* (water hyacinth), *Vossia cuspidata* (hippo grass), *Cyperus papyrus* (papyrus reed) and bioaccumulation factors (BAF) in River Kisat, Kisumu County. This was determined following laboratory standard procedures using Atomic Absorption Spectrophotometer (AAS). Results showed mean concentrations (mg/L) of Cd (Below detectable limit), Zn (0.2 -0.15) and Pb (0.03-0.03) in water. Mean values were within allowable WHO drinking water limits except for Pb which was above acceptable limits. Zn recorded highest accumulated levels in *E. crassipes* (72.43mg/L), *C. papyrus* (70.13mg/L) and *V. cuspidata* (71.03mg/L). *E. crassipes* was observed to bioaccumulate the highest concentration of the heavy metals with *C. papyrus* bioaccumulating the lowest (*E. crassipes* > *V. cuspidata* > *C. papyrus*). Pearson matrix correlation analysis showed positive significant correlations ($r = 0.621$; $p = 0.027$) between Zn and Pb in water and the macrophytes which reflected a common source of pollution and indication of accumulation. BAF recorded for all macrophytes ranged from 2.45 to 6.85, above value of 1, indicating significant bioaccumulation for Zn and Pb. The findings from this study showed

higher concentrations of Zn and Pb in the macrophytes in comparison to the water in River Kisat with BAF values >1 indicating bioaccumulation which represented poor water quality. The study recommends local authorities employ monitoring processes with incorporation of BAF as a water quality indicator where low metal concentrations in water are recorded, introduce penalties for polluters and sensitize local inhabitants on detrimental health effects of heavy metal and avoid use of River Kisat water for agricultural activities.

Keywords: Bioaccumulation, macrophytes, water quality, heavy metals, River Kisat.

INTRODUCTION

Direct and indirect discharge of pollutants into water bodies with limited treatment to remove pollutants leads to water pollution. Pollution of water bodies directly affects the organisms living in these water bodies and affects all natural biological communities. Toxic substances such as heavy metals are pollutants that accumulate in the ecosystem easily (An et al., 2015). Anthropogenic activities for example pesticide production, processing of metals, mining, electroplating and energy generation geothermally can lead to global heavy metal contamination (Peligro et al., 2016). Heavy metals exist in wastewater in different chemical forms hence their removal is difficult. Their non-biodegradable nature allows them to pass through various trophic levels and hence persist and accumulate in biota (Gall et al., 2015).

In Africa, Asia and Europe sources of metals are excessive fertilizer and pesticide use, waste, weathering rocks which contribute almost 50% total causes of pollution in aquatic systems (Qiaoqiao et al., 2020). Large river basins also face threats of pollution. Surface water heavy metal pollution has evolved from single to mixed pollution. City sewerage systems and factory untreated waste add large amounts of pollutants into river systems that serve local populations (Sinha & Loganathan, 2015). Human exposure to heavy metals occurs in various ways through food, water, air pollution or occupational exposure. Iron and manganese are examples of essential metals for biochemical and physiological body activities but elevated levels cause negative health effects. Heavy metals are toxic even at low levels. Long term intake of unsafe levels of heavy metals through foods may result in disruption of many biological and biochemical processes in humans. Consumption of water or food with high levels of zinc can lead to extensive health problems as stomach inflammations, skin cancers and anaemia. Lead and cadmium have been shown to affect reproductive systems, hepatic, endocrine, immune and gastrointestinal systems in both adults and children leading to lifelong illnesses or death (Jamshaid et al., 2018).

Macrophytes act as key accumulators of heavy metals by passively and absorbing actively therefore playing a key aspect of the wetland geochemistry (Vodyanitskii & Shoba, 2015). Some species of macrophytes can accumulate heavy metals to exceed the content in their ecosystem and are considered hyper accumulators. These species therefore can be used as indicators of environmental hazards and indicate the bioaccumulation potential of the species which in turn can evaluate persistence and toxic nature of metals (Singh et al., 2017). Trace elements in their ionic form can enter food chains in the ecosystem due to their persistence in nature. This in turn leads to their accumulation and contamination of biotic factors. Bioaccumulation factor determines the quotient of a metal in a plant to its initial content in the environment, specifically in water.

Monitoring of metal pollution has focused on metal concentrations in water, biota and sediments but dependence on water analysis is not a fully accurate method to identify metal inputs in aquatic systems (Turkmen et al., 2006). Determination of metal concentrations in aquatic organisms and their bioaccumulation factor is key in evaluating quality of water systems (Bastami et al., 2015). Macrophytes can be used as indicators of quality in the aquatic environment through by their uptake of heavy metals from the environment even at low concentrations (Matache et al., 2013).

Agricultural land is under heightened pressure in Kenya

and settlements by inhabitants has led to water catchment area destruction. There are many human activities documented that have impacted the state of Lake Victoria catchments such as Nyando, Nzoia, Yala, and Sondu-Miri (Mutuku et al., 2014; Onger et al., 2009). River Kisat receives wastewater, industrial and municipal wastes from anthropogenic activities from Kisumu city. The river flows into Lake Victoria basin through areas that have similar geological characteristics. The rainfall received in the area is moderate and most activities carried out include small scale maize and sugarcane farming as well as fishing. The dominant macrophyte species in River Kisat are *Eichhornia crassipes*, *Vossia cuspidata* and *Cyperus papyrus*. The river is an important source of water for consumption and for irrigation of agricultural activities. Human activities and natural or geological disturbances such as soil erosion, siltation, industrial effluents and rainfall can affect the deposits of organic and inorganic load in River Kisat aquatic ecosystem (Lalah et al., 2008). The river is polluted by effluent from the municipality, discharge from Kisat Wastewater Treatment Plant (KWTP), untreated waste from Nyalenda Waste Stabilization Ponds (NWSP), industrial effluent discharge from cosmetic industries in the locality, solid waste dumped from Obunga slums settlement, waste oil from informal car services garages and disposal of used molasses from informal alcohol brewing sites (Chamula et al., 2013).

The main challenge facing River Kisat is the extensive pollution of the river with heavy metals which affects the biota such as the macrophytes and poses a major health risk to the inhabitants of the area and their livelihood. Bioaccumulation status of the dominant macrophytes in River Kisat is unknown and has not been studied which is a threat to biodiversity and to communities consuming crops planted using contaminated water and using macrophytes as livestock feed. Aquatic vegetation in the Lake Victoria region is harvested for many purposes and many parts of the vegetation such as *Cyperus papyrus* plant can be utilized for fodder for animals. Besides *C. papyrus*, other wetland plant species such as *Phragmites* spp., *Eichhornia crassipes* are harvested for consumption by locals (herbs and medicinal plants) as well as fodder for animals kept in the homestead (Van Dam & Kipkemboi, 2016). Bioaccumulation factor can be utilized to assess the effects of heavy metal pollution on biodiversity and more so assess the extent of water quality in River Kisat providing time integrated information on metal presence.

The research problem addressed in this study is the environmental impact that heavy metal pollution has on the biota and potential social impact in the region in River Kisat. The aim of this study was to determine the levels of heavy metals in River Kisat water and in macrophytes and

to establish the bioaccumulation factors between water and macrophytes as an indicator of water quality and heavy metal pollution.

MATERIALS AND METHODOLOGY

Study area

River Kisat is situated in Kisumu Bay with coordinates of latitudes 0°18'S to 0°04'N and longitudes 34°43'E to 35°30'E (Figure 1). Located near the river source was sampling site K1, 3.5km from the river mouth located at the Kisat bridge characterized by disposal of waste oil from informal car-services garages and used molasses from local brewing. Site K2 was located 1km from K1, 2.5km from the river mouth, receiving effluent from the Kisat wastewater sewage and treatment plant area located in an Industrial area. Site K3 was located 1km away from K2 and 1.5km away from the river mouth in the Obunga slums with solid wastes dumped from informal settlements and various small cosmetic industries dumping waste into the River. K4 was located 500 metres away from the outlet of River Kisat into Lake Victoria and 1km away from K3 where local inhabitants practice agricultural activities with heavy point source pollution from agricultural activities due to pesticide and fertilizer use and run off.

Figure 1: Location of River Kisat in Lake Victoria Basin and selected sampling sites.

Water and macrophyte sample collection

Water sampling was done according to the procedures described by Ndimele and Johnson (2012). Water samples from all four sampling sites were collected at a depth of approximately 0.3 meters below the water surface and approximately 1.5 metres away from the riverbank at 3

different points on both sides of the riverbank using a 2-litre plastic Van Dorn water sampler and transferred into 500ml polypropylene bottles. Three droplets of Nitric acid (HNO_3) were added to the samples to reduce pH to <2 to ensure no precipitation and adsorption of metal ions to the polypropylene walls of the sample bottles. Samples were stored in a cool box at 4°C in order to halt the chemical and biological activities from occurring post sampling prior to analysis.

Three plant species were selected namely *Eichhornia crassipes*, *Vossia cuspidata* and *Cyperus papyrus* due to their dominance in the River Kisat ecosystem. Three composite samples of each of the macrophytes from the above-ground section (culms and stems) of *C. papyrus* and *V. cuspidata* and below ground section (submerged stolon sections) of *E. crassipes* were taken from each site. Plants were handpicked wearing nitrile gloves and were thoroughly washed at the sampling site using the sample water until appeared to be clean. Each plant sample was wrapped in foil to preserve the sample and placed in plastic airtight bags. Bags were sealed, labelled and stored at 4°C to wait for analysis.

Digestion of water samples

Digestion of water samples was done using concentrated Nitric acid (HNO_3) and Sulphuric (H_2SO_4) acid. Concentrated nitric acid (HNO_3) (10ml) and concentrated Sulphuric acid (H_2SO_4) (5ml) was added to 50ml of the water sample, heated to boil in a block digester until turbidity and color was clear. This was allowed to cool and the digested material was sieved through filter paper (0.45µm, whatman paper) into a flask and distilled water used to make the digest to the 100ml mark. Final filtrate was put in 100ml plastic bottles for heavy metals analysis. Analysis was done using Atomic Absorption Spectrometer (Spectra AA-10 model). The blank solutions were aspirated along with the digested samples.

Digestion of macrophytes samples

Macrophyte samples were placed in an oven and dried for 48 hours at 90°C to constant weight. Dried sample was crushed using pestle and mortar. Homogenized composite powder sample for each sample group was prepared by grinding crushed sample using stainless steel grinder. Digestion of the sample was done by adding 10ml of concentrated HNO_3 to 2g of the finely ground homogenized powder, heated at 80°C for 6 hours until a transparent color appeared of the sample. Digested mixture was evaporated to dryness and residue dissolved in 10ml of 1 N HNO_3 . This was filled to the 100ml with double deionized water in a volumetric flask and passed through a 0.45µm filter paper and transferred to 100ml polypropylene containers for metal analysis using the AAS.

Data analysis

Data was subjected to ANOVA. Significance was accepted at $p \leq 0.05$. Post-hoc Tukey's HSD test (HSD) test was used to separate the means. Pearson's correlation analysis established relationship between mean concentrations in water and plants. The bioaccumulation factor (BAF) was calculated as follows:

$$\text{BAF} = \frac{\text{Metal content in plant (mg/L)}}{\text{Metal content in water (mg/L)}}$$

RESULTS AND DISCUSSION

Zn, Pb, Cd concentrations in water

The mean Zn concentration levels are presented in Table 1. Zn concentration levels recorded were within the WHO set limits for water for human consumption. The elevated levels of Zn at K1, river source could be attributed to sewage and industrial effluent from the surrounding area (Table 1). This could be due to Zn^{2+} ions being more bioavailable in suitable conditions of pH and temperature. The introduction of Zinc in water can be done naturally through mineral erosion from soil and rocks or through anthropogenic activities such as manufacturing of steel products, fertilizer leachate, effluents from the municipal and the destruction of waste materials through burning (Tomno et al., 2020). In Dar es Salaam, a study conducted on River Msimbazi polluted by leakage of effluents from waste dumps, abattoirs, and domestic wastewater from septic tanks in Temeke municipality recorded Zinc levels of 0.99 – 1.26mg/L which were slightly higher than what was recorded in this study (Kacholi et al., 2018).

The mean Pb concentration levels in water are presented in Table 1. Pb concentration levels were above the WHO set limits for drinking water except for site K3 which was within WHO limits. The levels of Pb recorded in River Kisat could be attributed to the presence of various industries dealing in cosmetics and motor vehicle repairs that drained effluent directly into the river before treatment. Sources of Pb pollution include industrial effluents, pesticides, fertilizers, sewage sludge and the mining or smelting of Pb ore which has severe effects on plant and animal life and functions (Malar et al., 2016). A study on the Yacuambi River in Ecuador by Villa-Achupallas et al. (2018) was found polluted by gold metal mining activities and recorded mean Pb levels of 0.113mg/L which were comparable to levels recorded in this study.

In this study Cd levels were below the detection limit (Table 1) which indicates that there is minimal cadmium pollution in the river and possibly in the surrounding localities. Cadmium can be introduced into the aquatic water systems as a result of anthropogenic activities for

example the application of fertilizer, mining or smelting of metal ores and also from natural occurrences such as rock weathering. A study by Njuguna et al. (2017) on Cd concentrations in Nairobi River polluted by human and industrial waste from Korogocho slums recorded mean values of 0.0043mg/L which were low levels comparable to the below detectable limits recorded in this study.

Concentrations of Pb, Zn, Cd in macrophytes

The mean Zn, Pb and Cd concentration levels in *E. crassipes* are presented in Table 2. From this study the results showed that *Eichhornia crassipes* is a good bioaccumulator of Zn and Pb. *E. crassipes* a free floating macrophytes was observed to bioaccumulate the highest concentration of the three heavy metals (Table 2). *E. crassipes* has a greater capacity to remove metals from their environment as these species grow in abundance (Radu et al., 2017). Aquatic macrophytes of free floating nature such as water hyacinth (*Eichhornia crassipes*) has high potential and capability to remove heavy metals from their ecosystem. They actively transport heavy metals through their root system to the stems and leaves (Gunathilake et al., 2018). Mean varied levels of metals recorded from *Cyperus papyrus* are summarized in Table 2. From the results *C. papyrus* accumulated high concentration of Zinc at the river source which could be attributed to untreated wastewater released into River Kisat (Table 2). *Cyperus papyrus* has been used in the phytoremediation of metals in water systems and has exhibited capacity in the retention of heavy metals such as Cu, Pb, Zn and Iron (Mbemba et al., 2019). The mean Zn, Pb and Cd concentration levels in *V. cuspidata* for all the sampling sites are presented in Table 2. Cd levels were below detectable limit. The concentration of Zn and Pb in *V. cuspidata* is an indicator that the plant can bioaccumulate heavy metals (Table 2). The bioaccumulation of heavy metals in most emergent plants such as *V. cuspidata* is restricted due to the upward transport mechanism which is the movement of compounds towards the tips of the plant extremities hence the minimal levels of metals (Outa et al., 2020).

The upward or acropetal mechanism of emergent plants to transport compounds to plant extremities such as stems and leaves restricts movement of compounds which provides insight into the subsided levels of heavy metals in the leaves and stems of *V. cuspidata* and *C. papyrus* in comparison to *E. crassipes* (Sayer et al., 2018). Another study done by Ndeda and Manohar (2014) in Nairobi Dam showed that free floating macrophytes had greater accumulation ability values of Cu, Cd and Pb compared to emergent species. This concludes that free floating macrophytes have better ability to bioaccumulate heavy metals than emergent macrophytes.

Table 1: Mean values of Zn, Pb and Cd (mg/L) in water from River Kisat

Element/Sample Site	K1	K2	K3	K4	WHO Standards 3 RD Edition (mg/L)
Zn (mg/L)	0.22±0.04 ^a	0.12± 0.07 ^a	0.13 ± 0.13 ^a	0.15± 0.07 ^a	3
Range	0.22-0.22	0.11 - 0.13	0.15 - 0.13	0.16 - 0.14	
Cd (mg/L)	ND	ND	ND	ND	0.003
Pb (mg/L)	0.03±0.01 ^a	0.02 ±0.04 ^a	0.01±0.01 ^a	0.03± 0.05 ^a	0.01
Range	0.02– 0.02	0.02 – 0.01	0.01 – 0.01	0.03 – 0.01	

Note: Different letter superscripts = significant difference at $p < 0.05$. ND = below detectable limit

Table 2: Mean values of Zn, Pb and Cd (mg/L) in macrophytes

Plant Species	Sample Site	K1	K2	K3	K4
<i>Vossia cuspidata</i>	Zinc(mg/L)	71.03±0.26	78.95±0.05	34.79±0.50	31.50±0.40
	Range	70.03 – 71.04	54.34 – 79.04	24.12 – 35.92	24.56 – 33.72
	Lead(mg/L)	11.01 ±0.14	6.23±0.07	2.94±0.23	3.74±0.14
	Range	9.43 – 12.56	5.34 – 6.78	1.92 – 3.66	3.55 – 4.12
<i>Cyperus papyrus</i>	Cadmium(mg/L)	ND	ND	ND	ND
	Zinc(mg/L)	70.13 ± 0.40	79.10 ± 0.05	32.01 ± 0.50	30.95 ± 0.26
	Range	70.01 – 70.14	78.23 – 79.11	30.33 – 32.04	30.85 – 30.95
	Lead(mg/L)	10.45 ± 0.07	6.18± 0.23	3.21 ± 0.07	3.64 ± 0.14
<i>Eichhornia crassipes</i>	Range	10.33- 10.45	6.14 – 6.19	3.19 – 3.22	3.62 – 3.64
	Cadmium(mg/L)	ND	ND	ND	ND
	Zinc(mg/L)	72.43 ± 0.40	79.07 ± 0.05	34.63 ± 0.51	31.51 ± 0.26
	Range	71.40 – 72.33	78.03 – 79.15	33.41 – 34.65	30.15 – 31.50
	Lead(mg/L)	11.04 ± 0.23	6.33 ± 0.07	3.54 ± 0.14	3.78 ± 0.07
	Range	11.01 – 11.05	6.22 – 6.34	3.30 – 3.54	3.62 – 3.78
	Cadmium(mg/L)	ND	ND	ND	ND

Note: * = significant difference at $p < 0.05$, ND = below detectable limit

Relationships between water and macrophytes metals concentrations and BAF

Pearson correlation was determined to measure the relation and linear dependence between concentration of metals in water and macrophytes. The concentration of Zn and Pb in *E. crassipes* showed positive significant correlation with Zn and Pb in water ($r = 0.621$; $p = 0.027$) and ($r = 0.358$; $p = 0.023$) respectively (Table 3). The concentration of Zn, Pb in *C. papyrus* showed positive significant correlation with Zn and Pb levels in water ($r = 0.592$; $p = 0.027$), ($r = 0.456$; $p = 0.015$) respectively (Table 3). Zn and Pb concentrations in *V. cuspidata* showed weak positive correlations with Zn and Pb in water ($r = 0.153$; $p = 0.033$), ($r = 0.248$; $p = 0.036$) (Table 3).

Table 3: r values of correlation between water and macrophytes in River Kisat

	Zn –Water	Pb- Water
Zn– <i>E.crassipes</i>	.621*	.384
Pb– <i>E.crassipes</i>	.347	.358*
Zn– <i>C.papyrus</i>	.592*	.006
Pb– <i>C.papyrus</i>	-.057	.456*
Zn– <i>V.cuspidata</i>	.153*	-.293
Pb– <i>V.cuspidata</i>	-.059	.248*
Zn–Water	.194	.265
Pb–Water	.146	.299

Note: * = Correlation significance

The noted significant correlations among the heavy metals reflected a common source of occurrence and an indication of similar accumulation pathways for subsequent accumulation in macrophytes species.

Bioaccumulation factor was calculated to determine the extent of heavy metal bioaccumulation by the macrophytes and indicate the quality of water. The bioaccumulation factor (BAF) of heavy metals in the macrophyte species and River Kisat water are presented in Table 4. The results indicated no significant difference in the BAF values between the different sampling sites ($p = 0.619$; $df = 36$) (Table 4). The 3 macrophyte species in River Kisat

recorded BAF values >1 for Pb and Zn (Table 4). The high BAF values for Pb and Zn indicate that all 3 plant species were able to bioaccumulate Pb and Zn from River Kisat (Table 4). The ability and efficiency of plants to accumulate heavy metals in their plant tissues to levels higher than the surrounding environment exceeding the amounts of heavy metals in their aquatic ecosystem indicates extensive heavy metal pollution (Singh et al., 2017). These BAF values were evidence that Zn and Pb were bioaccumulated in the macrophytes and River Kisat water was contaminated with high enough levels of Pb and Zn to lead to bioaccumulation.

Table 4: Bioaccumulation Factors of heavy metals in *E. crassipes*, *V. cuspidata* and *C. papyrus* from River Kisat

Macrophytes		BAF Values		
	Sampling sites	Zinc	Lead	Cadmium
<i>Eichhornia crassipes</i>	K1	3.04	2.45	-
	K2	1.54	3.49	-
	K3	3.65	3.56	-
	K4	4.76	6.79	-
<i>Cyperus papyrus</i>	K1	3.14	2.59	-
	K2	1.54	3.58	-
	K3	3.95	3.93	-
	K4	4.84	7.04	-
<i>Vossia cuspidata</i>	K1	3.10	2.46	-
	K2	1.54	3.55	-
	K3	3.64	4.29	-
	K4	4.76	6.85	-

Note: * = significant difference at $p < 0.05$ levels

Therefore, it can be concluded that the key source of contamination of heavy metals by macrophytes species in River Kisat is from the water. The BAF values >1 indicate higher concentration of heavy metals in macrophytes in comparison to levels in water was evidence that Zn and Pb were bioaccumulated in the macrophytes and that River Kisat water was contaminated with high enough levels of Pb and Zn to lead to bioaccumulation which indicated poor water quality.

CONCLUSION

Mean heavy metal concentrations of Zn was within recommended WHO levels for water for consumption. Mean concentrations of Pb were recorded to be higher than WHO limits for water meant for drinking except at one sampling site which recorded levels within WHO set limits (0.01mg/L). Cd levels were below detectable limit. Therefore, it was concluded that the amount of heavy metals in River Kisat water is a threat to the water ecosystem. The three macrophytes in River Kisat recorded higher levels of heavy metal concentration than in the water

from River Kisat, which indicated bioaccumulated heavy metals. The concentration of Zn and Pb in *E. crassipes*, *C. papyrus* and *V. cuspidata* showed positive significant correlation with Zn, Pb in water from River Kisat and BAF values >1 was evidence that bioaccumulation factors were indicators of heavy metal pollution. The study shows that the agricultural and industrial activities being conducted along the River Kisat contribute to pollution of the water by heavy metals and subsequently BAF values confirmed accumulation in the macrophytes indicating poor water quality of the river.

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