

**PHYTOREMEDIATION INDICES OF WATER HYACINTH (*EICHHORNIA CRASSIPES*)
GROWING IN PANTEKA STREAM, KADUNA.**

ABSTRACT

The aquatic environment may be polluted by the release of heavy metals which are toxic in high concentrations, persistent in the environment and bio-accumulative. This investigation was conducted to assess the phytoremediation potential of water hyacinth (*Eichhornia crassipes*) using phytoremediation indices (where?). The root and shoot samples of *E. crassipes* growing at the sampling points (A, B, C and Dare represent of....) were analyzed to determine heavy metal concentrations, including those of cadmium (Cd), chromium (Cr), copper (Cu), nickel (Ni), lead (Pb), and zinc (Zn) through Atomic Absorption Spectrophotometry (AAS). The phytoremediation indices were calculated via bioaccumulation coefficient and translocation factor. The results showed that the root samples had higher accumulation of heavy metals than the shoot samples. Zn accumulated the highest in roots and shoots (335.32 ± 23.60 and 256.52 ± 30.82 please measuring unit) at sampling point B respectively. Heavy metals were translocated efficiently and had bioaccumulation coefficient and translocation factor greater than one. Nickel had the highest bioaccumulation coefficient and translocation factor, and Zn had a translocation factor that was less than one across all samples. This implies that *E. crassipes* is a potential hyperaccumulator plant for phytoremediation.

keywords: *Eichhornia crassipes*, Heavy metals, Phytoremediation, and Phytoremediation Indices

I. INTRODUCTION

One of the key priorities of this present age is the preservation of the environment's quality. The biosphere is being degraded as a result of the emission of natural and man-made substances that can harm living organisms. Heavy metals, among all pollutants, are easily transported and accumulated in the

environment [1]. Water body contamination increases at an alarming rate by human activities such as the release of pesticides from agricultural sectors, radionuclides and hydrocarbons from petrochemical industry, and hazardous metals from untreated wastewater are all instances of pollutants. Metals, unlike organic trash, are not biodegradable. Arsenic, Cadmium, Lead, Manganese, Mercury, Selenium and Zinc are examples of trace and heavy metals that must be eradicated from the environment [2]. Once heavy metals have contaminated the environment, they will pose a long-term threat to humans and animals [3]. Biological decontamination approaches, such as phytoremediation, are thought to be safe for eliminating these toxins from the environment, especially from water and soil.

A green technology known as phytoremediation uses plants to clean up contaminated areas or to dislodge pollutants from the environment. It is an environmentally benign and affordable technology [4]. On contaminated terrain, many plant species may thrive, and some may even accumulate significant amounts of heavy metals in their tissues. Hyper-accumulator plants with the capacity to develop promptly and accumulate large metal levels are required to facilitate phytoremediation [5]. Currently known metal hyper-accumulator plant species number over four hundred [6].

Phytoextraction, phytostabilization, rhizofiltration, and phytovolatilization are some of the phytoremediation approaches for treating metal-contaminated environments [7][8]. Phytoextraction refers to the absorption of metals, coupled with subsequent translocation and accumulation in vegetative organs, whereas phytostabilization refers to the hazardous ions that remain immobilized in the contaminated medium. As a result, heavy metals become stable in the rhizosphere (horizontal underground stem region in the plant). The roots of phytoremediating plants absorb, concentrate or discharge contaminants out of polluted effluents during the rhizofiltration process [9]. Harvesting plant tissue that has accumulated hazardous metals is required for phytoextraction and rhizofiltration. The plant material is normally burned, and the ash generated can be utilized to recycle metal [10][7]. The creation of "eco-catalysts" for the chemical sectors is a recent method of utilizing biomass that has been gathered from plants that highly accumulate metal [11][12]. Volatilization is another method that plants use to remove inorganic

contaminants. Metals undergo biological conversion to gaseous states, which are then discharged into the air during this process [13].

Recent strategy for phytoextraction of heavy metal contaminated soil includes the use microbial-assisted phytoremediation which is a potential approach, where a broad range of plant species coexist closely with arbuscular mycorrhizal fungi (AMF), promoting healthy growth and resilience to contaminants. AMF inhabit plant roots and expand their hyphae within the rhizosphere (is this microorganism present in *E. crassipes*?) helping the plants take up nutrients and minerals, control the buildup of heavy metals and stress tolerance [14]. [15], carried out a study using arbuscular mycorrhizal fungi to aid phytoextraction. Arbuscular mycorrhizal fungi (AMF) were used in this work to enhance the growth of *Zea mays* L. in heavy metal-rich tannery slush [16].

Eichhornia crassipes is an aquatic macrophyte belonging to the class Liliopsida and the family Pontederiaceae. It originated in tropical South America, but it can now be found in Africa, Australia, India, and a variety of other places. This plant may be used as animal feed and has a fast growth rate as well as the ability to withstand various forms of pollution [17]. An excellent plant for phytoremediation has a high biomass, high tolerance to heavy metals stress, and a high capacity for metal accumulation [18]. The said plant has these qualities; therefore, it is well-known for their ability to absorb toxic metals from water. Water hyacinth, as a good and capable hyper-accumulator plant, can accumulate a very high level of heavy metals [19].

The sole aim of this investigation was to determine the heavy metals content of the native aquatic macrophyte (*E. crassipes*) in Panteka stream and the pond (fish pond connected to Panteka stream?) and its ability to accumulate heavy metals and potency for phytoremediation. As well as, to ascertain more information on the heavy metal status of water bodies and *E. crassipes* which was seen as noxious species.

II. MATERIALS AND METHODS

A. Study Area

The study area is the stream that has its entry point from RafinGuza, and is flowing through the **Panteka mechanic village**, located between latitudes $10^{\circ} 32'37''$ and $10^{\circ} 33'56''$ N and longitude $7^{\circ} 24'29''$ and $7^{\circ} 25'7''$ E, at the Northern part of Kaduna, Nigeria. The Panteka stream flows westward, it usually decreases in volume during dry season and *Eichhornia crassipes* grows in the stream. Similarly(?), **Panteka is a mechanic village** where all kinds of cars and motorcycle spare parts are sold, and their maintenance is carried out. Alongside this, agricultural activities take place in this area and runoff from the farms and debris washed off from these anthropogenic activities mentioned above enters the stream. The control samples were collected from an enclosed pond, where farming activities are carried out, behind Mal. Abdulrakeem Fish Farm, Farin-Gida Mando Kaduna. The stream was divided into three sampling points, A, B, and C. Sampling point A is 4,218m away from B, and B is 4000m away from C. At sampling point A, the predominant activity done in this area is farming, dumping of car worn-out parts and repairs, while at sampling points B and C, there is more of mechanic repair activities(why are each sampling point so important to explain? Are they has special affected to heavy metals?).

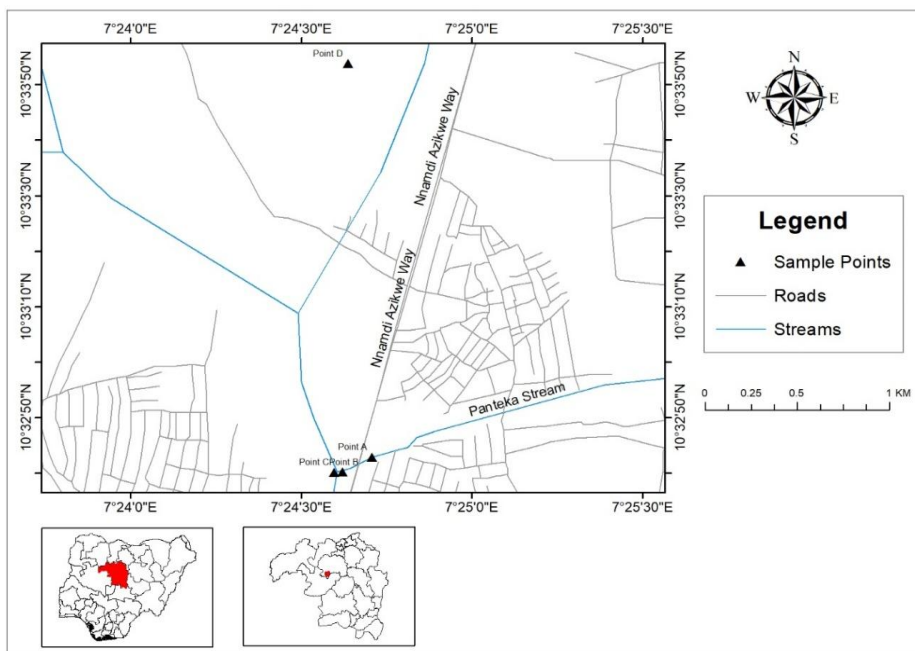


Figure.1: Panteka stream study area map with sampling points, (Source: GIS, KADSU). (Please make the picture more clearly: zone of pollution should be present, water movement, and the picture should be simplified understand)

B. Plant Sample Collection

Plant samples were collected from Panteka's contaminated water as well as a control. Plant samples from the four sampling points were wrapped in labeled polybags for each of the plants and brought to the Biological Sciences Department Laboratory Nigerian Defence Academy, Kaduna.

C. Plant Sample Processing and analysis

The plant samples were washed to remove the unwanted debris from the plant with deionized water. Followed by herbarium techniques (which include pressing and drying the plant sample using the plant presser, labeling, and storage) and full authentication of the plant. The plant samples were identified and authenticated in the herbarium, Botany laboratory NDA. The specimen with voucher number NDABIOH202030 was kept in the herbarium for reference purposes.

D. Water Sample Collection

In both dry and wet seasons, water was sampled twice at early hours of the morning in sterile plastic containers from the four sampling points. Using the method of collecting water grab samples as described by [20], and was done by holding the uncapped sterile plastic container upside down and submersed it. And, the tip of the container upright and then allowed water to fill the container and was removed from the water, screw-on cap and carefully labeled respectively. These water samples were subjected to laboratory investigation for heavy metals.

E. Heavy Metal Analysis of Water

To determine the heavy metals in water samples, [21] approach was employed. 50ml of the water samples from each sampling point foreach of the seasons were digested with 10ml of HNO₃ on a hot plate. The resulting clear solutions were filtered using Whatman filter papers and reconstituted to 50ml in volumetric flasks with deionized water. 50ml of the filtrates were analyzed for the heavy metals using Atomic Absorption Spectrophotometer (Model: PG-990). The reference solutions of metals to be assayed were

prepared and atomic absorption spectrophotometer was allowed to run for 10 minutes before immersing the sipper into the reference metal solutions to calibrate the AAS and acetylene was used as the carrier gas. Flame absorption method at a wavelength of 283.3nm (for lead only), slit 0.4nm, high voltage of 368V and lamp current of 2.0mA were utilized in the process. The absorbance was read before placing the sipper into the solution of the digested samples and the concentrations were taken from the AAS.

F. Heavy Metals Analysis of Plant

The method described by [22] was adopted to analyze heavy metals from the plant samples. Exactly 0.25g of each of the grounded plant samples (roots and shoots) were weighed after which it was placed in a 100ml beaker and digested with 30ml of mixed acid containing the following; 650ml, 80ml, and 20ml of concentrated, Trioxonitrate (v) acid (HNO₃), Perchloric acid (HClO₄) and Tetraoxosulphate (vi) acid (H₂SO₄) respectively on a hot plate. Following completion of the digestion, samples were cooled, made up to 50ml with distilled water, filtered using Whatman filter paper in volumetric flasks, and transferred into a sterile labeled plastic container. Using an Atomic Absorption Spectrophotometer (Model: PG-990), 50ml of the filtrates were utilized to determine heavy metal contents [23].

G. Characterization of Phytoremediation Potential

Bioaccumulation Coefficient (BAC): Is the plant/water concentration quotient [24]. It is calculated as follow;

$$BAC = C_{\text{root}} / C_{\text{water}} \quad BAC = C_{\text{shoot}} / C_{\text{water}}$$

Where C_{water} = Concentration of heavy metal in *E. crassipes* root (mg/kg), C_{shoot} = Concentration of heavy metal in *E. crassipes* shoot (mg/kg) and C_{water} = Concentration of heavy metals in water (mg/kg).

Translocation Factor (TF): The translocation from shoot to root was determined by translocation factor, which is given by:

$$TF = \frac{\text{Metal Concentration in the shoot (mg/kg)}}{\text{Metal Concentration in the root (mg/kg)}}$$

H. Statistical Analysis

To confirm the variability and validity of the results, One-way analysis of variance (ANOVA) and the T-Test were used to analyze the study's data. Using the Statistical Package for Social Sciences (SPSS-version 23.) and Excel version 2016, all data were presented as LSD and given a significance level of $p < 0.05$.

III. RESULTS AND DISCUSSION

A. Heavy metals concentration in the study area

The levels of these heavy metals in samples of water across the wet season from sampling points A, B, C, and D increased. This might be due to the runoff from farm lands and debris washed off from other anthropogenic activities depositing more of these heavy metals, and the increase in Zinc at sampling point A and B, during the dry season might be due to the engine oils, wastes from car parts and other items coated with zinc being washed off into the stream (*this sentence contradictory*). Among the heavy metals analyzed at Panteka Stream during the wet season, Cadmium was observed to have the highest concentration (0.78mg/kg) and this was more than the maximum concentration standards of FEPA (0.01mg/L) and USEPA (0.01mg/L). The high content of these heavy metals might be as a result of the release of effluents from car paints, car batteries, and phosphate fertilizers in the stream (are literatures support this?). The heavy metals concentration conforms with the findings of [25] who reported that heavy metals were observed from paper dumping sites (please compare to same objector condition). Cadmium concentration in the water samples was the highest among the heavy metals studied. This was similar to the reports of [26], who in their study of heavy metals in sediments collected from the Gorgan coast (please

compare to water), reported that Zinc had the highest concentration. However, the other heavy metals analyzed were within the recommended limits.

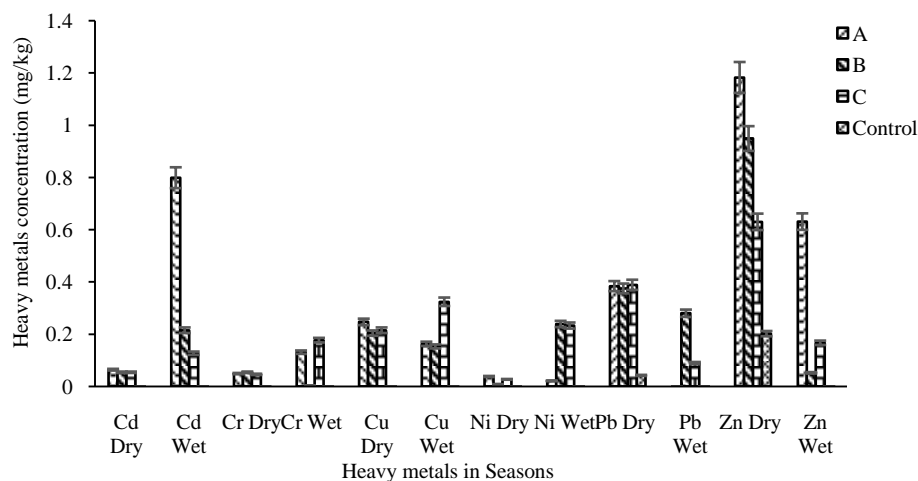


Fig. 2. Mean Concentration of H(heavy) metals at the sampling points during dry and wet seasons.

B. Heavy metal concentration in the roots and shoots of *E. crassipes*

Heavy metals concentration was entirely higher in roots than in shoots. Different plant components metals accumulation indicated heavy metals in varying concentrations, at the sampling points as shown in Table I and II. There was more accumulation of Zinc in root and shoot at sampling point B (335.32 ± 23.60 and 256.52 ± 30.82) respectively. Other heavy metals accumulated significantly in *E. crassipes* growing at all the sampling points **exceptin the control samples(Pb and Zn high in control)**. The results were in line with the findings of [27], who studied heavy metals in 12 plant species growing around the tailing dam where they found out that all metals accumulated higher in the root than in the shoot. Also, [28], observed the same higher concentration of the Cadmium in the root than in the shoot. [25] recorded a higher accumulation of heavy metals in root than in shoot of *E. crassipes* growing at **paper dumping site(?)** and further stated that, in terms of plant components, the ratio of metal concentrations does not appear to change in a way that is reliant on exposure. **Hence, there was a substantial accumulation of all heavy metals in roots and shoots of *E. crassipes* both at Panteka stream and the control ($P < 0.05$).**(this sentence seemly contrast to the upper part)

Table I: Heavy Metal Concentration in Root Samples

Metals						
Sampling points	Cd	Cr	Cu	Ni	Pb	Zn
A	27.65±30.06 ^a	47.20±21.69 ^a	36.88±22.09 ^a	50.90±16.48 ^a	55.05±61.88 ^a	224.45±30.24 ^a
B	54.76±13.49 ^a	57.96±13.11 ^a	127.12±19.45 ^a	34.52±11.88 ^{ab}	BDL	335.32±23.60 ^b
C	46.88±11.27 ^a	163.68±129.62 ^a	90.72±7.54 ^b	31.20±8.11 ^{ab}	23.32±49.69 ^a	195.56±22.10 ^a
Control	BDL	12.95±3.34 ^a	0.70±3.70 ^a	15.00±5.35 ^b	72.45±7.99 ^a	227.70±40.87 ^a

Values are expressed as mean ± standard error mean. Means with different superscripts down the column are significantly different ($P < 0.05$) at 95% confidence level using least significance difference (L.S.D), BDL-below detectible limit. The descending order of the heavy metal's accumulation in the root samples from the maximum to the minimum at the sampling points is; Zn>Cr>Cu>Pb>Cd>Ni.

Table II: Heavy metal concentration in shoot samples

Metals						
Sampling points	Cd	Cr	Cu	Ni	Pb	Zn
A	29.70±27.67 ^a	34.70±18.53 ^a	28.55±17.97 ^{ab}	51.25±18.49 ^{abc}	3.70±25.21 ^{ab}	194.00±30.39 ^a
B	58.08±9.13 ^a	39.88±14.94 ^a	50.24±17.57 ^b	24.00±4.59 ^{ab}	BDL	256.52±30.82 ^a
C	54.96±10.79 ^a	51.96±17.88 ^a	74.44±16.90 ^b	68.28±18.89 ^c	33.60±38.47 ^{ab}	195.56±22.10 ^a
Control	BDL	10.20±3.75 ^a	BDL	5.45±2.26 ^b	47.30±5.17 ^a	113.80±1.49 ^b

Values are expressed as mean ± standard error mean. Means with different superscripts down the column are significantly different ($P < 0.05$) at 95% confidence level using least significance difference (L.S.D), BDL-below detectible limit. Zn>Cu>Ni>Cd>Cr>Pb is the descending order of the heavy metals in the shoot samples from the maximum to the least accumulation at the sampling points.

C. Correlation of heavy metals, roots and shoots, and the seasons of sampling

The roots and shoots of *E. crassipes* and concentration of heavy metals in the water samples revealed a positive correlation as shown in table III. This agrees with the reports of [28], who reported a significant relationship between total Cadmium concentration in the growth media and **total Cd concentration in *Hopea odorata* (please compare to similar aquatic weed)** In their report, the overall Cadmium concentration in the growth media is significantly related to the total Cd concentration in *Hopea odorata*. Similarly, the total concentration of heavy metals in the root and shoot samples of *E. crassipes* increased in conjunction with the total concentration of these metals in the water, indicating a positive correlation. In addition, heavy metals at sampling points increased as these metals increased during the dry and wet seasons.

Table III: Pearson Correlation Coefficient between Heavy metals in the roots and shoots of *E. crassipes* and between the heavy metals in the water samples during seasons at the sampling points.

Heavy Metals	Roots and Shoots	Dry and Wet
Cadmium (mg/kg)	0.998*	0.784*
Chromium (mg/kg)	0.862*	0.833*
Copper (mg/kg)	0.836*	0.883*
Nickel (mg/kg)	0.605*	0.895*
Lead (mg/kg)	0.826*	0.457*
Zinc (mg/kg)	0.646*	0.855*

*Correlation is significant at $P < 0.05$ at 95% confidence level (seemly it is not important to write).

D. Bioaccumulation coefficient and translocation factor of *E. crassipes*

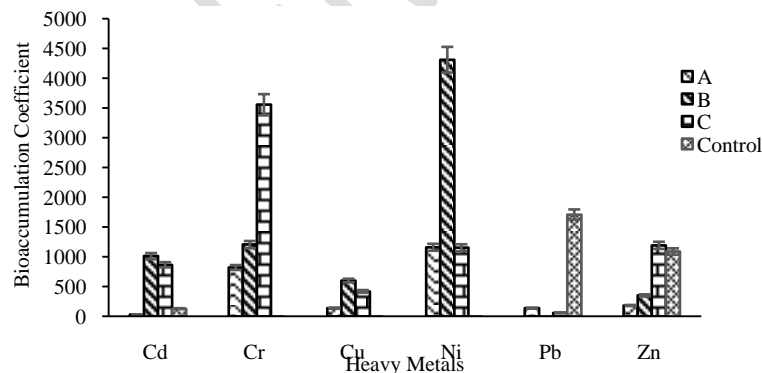


Fig. 3. Mean Bioaccumulation Coefficient of Heavy Metals in the roots of *E. crassipes*. The descending order of bioaccumulation coefficient in the roots from maximum to the minimum at the sampling points is as follows Ni>Cr> Pb>Zn>Cd>Cu.

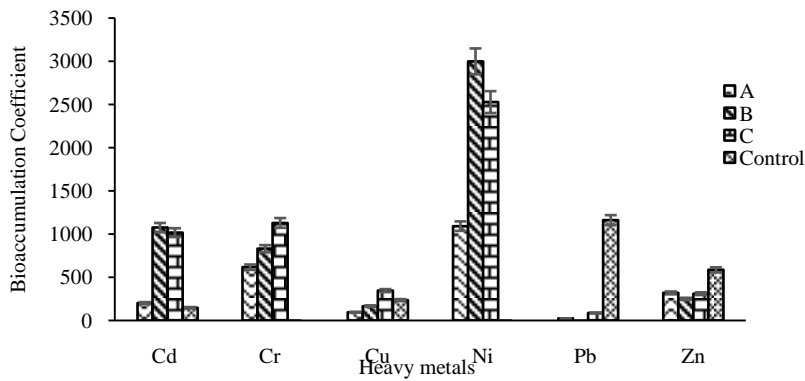


Fig. 4. Mean Bioaccumulation Coefficient of Heavy Metals in the shoots of *E. crassipes*. The descending order of bioaccumulation coefficient in the shoots from the maximum to the minimum at the sampling points is as follows; Ni>Pb>Cr>Cd>Zn>Cu.

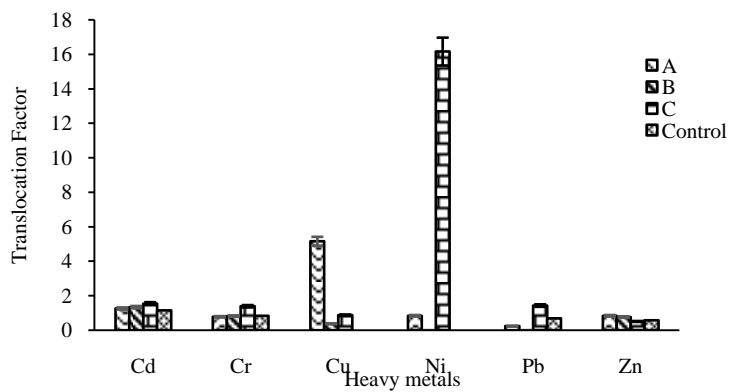


Fig. 5. Mean Translocation Factor of Heavy metals in *E. crassipes*. The descending order of Translocation factor ranged from the maximum to the minimum at the sampling points is as follows; Ni>Cu>Cd>Pb>Cr>Zn.

To assess *E. crassipes*' capability for phytoremediation of heavy metals, two phytoremediation indices were applied. These include Bioaccumulation Coefficient (BAC) and Translocation Factor (TF). Bioaccumulation Coefficient (BAC) was used to indicate the degree of enrichment of heavy metal in the plants relative to that in its habitat. Nickel showed the highest and Copper the least bioaccumulation coefficient in the root and shoot samples of *E. crassipes*. This stood in contrast with the study of [29]. Who in their study reported the highest bioaccumulation coefficient in Cadmium followed by Copper, Zinc, and Lead.

[30] reported that Fungi like *Aspergillus niger* grow on roots of water hyacinth and can grow on an alkaline media (pH greater than 7) that can adsorb metals. The adsorption of metals onto the roots of water hyacinth may have been possible due to *A. niger* as was reported by [31]. In this study, the bioaccumulation

coefficient was greater than one for all the heavy metals. This might be due to the high accumulation in root and shoot of *E. crassipes*.

If the translocation factor is less than one ($TF < 1$) the plant has the potential for phytostabilization (also known as excluder plant), whereas if the TF is greater than one ($TF > 1$) the plant is classified as a phytoextractor or accumulator [32]. This study was in accordance with the findings of [33] in the assessment of Cadmium, Chromium, Copper, and Nickel by *Tetraenaqataranse*. Where the translocation factor in nickel was the highest and was greater than one. But was in contrast with the finding of [29] who in their study, had the highest TF recorded in Zinc in the stems and leaves of *Rhazystricta*. (Dear researcher, please compare your *E. crassiper* to other similar aquatic plant, because some terrestrial plant especially desert plant should has different stem system and metabolism).

The phytoremediation potential of a plant is characterized not just by its capacity to acquire and accumulate significant concentration of metal, as well, by the plant possessing the enabling capacity to translocate the metals to aerial components while also producing huge biomass[34]. This was observed in *E. crassipes* in this study. The Translocation factor in this investigation demonstrated that there was no significant difference ($P < 0.05$) in the different heavy metals at the sampling points. The translocation factor of heavy metals from shoot to root was greater than one. Zinc was not significantly translocated. Hence, its translocation factor was less than one in all the sampling points.

IV. CONCLUSION

From this finding, the *E. crassipes* could be said to be phytostabilizer for Zinc and is a phytoextractor of Nickel, Lead, Cadmium, Chromium, and Copper. This is suggesting that *E. crassipes* is a good hyperaccumulator plant and serve as better candidate for phytoremediation.

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