



Trends in  
**Applied Sciences  
Research**

ISSN 1819-3579



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## Phytoremediation of Iron (Fe) and Copper (Cu) by Water Hyacinth (*Eichhornia crassipes* (Mart.) Solms)

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### ABSTRACT

This study was carried out to investigate the ability of water hyacinth (*Eichhornia crassipes* (Mart.) Solms.) to absorb and translocate iron (Fe) and copper (Cu). The study was conducted with three concentration gradients of Fe and Cu at 10, 15, 20 mg L<sup>-1</sup> and control (no metal). The whole set-up was carried out in triplicate and the experiment lasted for 12 weeks. The results showed that iron (Fe) had the highest accumulation value in the root (11.22±6.69 mg kg<sup>-1</sup>), while copper (Cu) had the highest value in the leaf (3.80±0.12 mg kg<sup>-1</sup>) both occurred at treatment spiked at 20 mg metal/L of water. Statistical analysis showed that there was significant difference (p<0.05) in metal accumulation among treatments. The Translocation Factor (TF) values for Fe ranged from 0.49±0.57 to 0.68±0.27 in leaf and 0.64±0.17 to 0.77±0.18 in the stem while the TF values for Cu ranged from 0.78±0.08 to 1.12±0.12 in leaf and 0.72±0.32 to 1.09±0.19 in the stem. This reveals that Cu had better translocation capability than Fe. Highest values of Bioconcentration factor (BCF) for Fe and Cu were 2.32±0.65 at 20 mg L<sup>-1</sup> and 0.72±0.01 at 15 mg L<sup>-1</sup> obtained in the root and leaf respectively, indicating that the accumulation potential of Fe by water hyacinth is higher than Cu. So, according to the accumulation capabilities of the investigated plant (*Eichhornia crassipes*), this study showed that the plant was found to be a promising candidate for phytoremediation and adequate for bio-monitoring programmes for contaminated water.

**Key words:** Phytoremediation, translocation factor, bioconcentration factor, biomonitoring, bioaccumulation

### INTRODUCTION

The rapid pace of industrialization and urbanization has increased the problem of heavy metal pollution. There are many pathways by which pollutant (heavy metals) leave their sites of application and are distributed throughout the environment and enter the aquatic ecosystem. The major route of heavy metals to water ecosystems in urban areas is through rainfall runoff and atmospheric deposition; another source is from municipal and industrial discharges (Werimo *et al.*, 2009).

In small quantities, certain heavy metals are nutritionally essential for a healthy life. Some of these are trace elements e.g., iron, copper, manganese and zinc (Odjegba and Fasidi, 2004). Heavy

metal toxicity and the danger of their bioaccumulation in the food chain represent one of the major environmental problems of our modern society. The most common heavy metal contaminants are: Cadmium (Cd), Chromium (Cr), Copper (Cu), Mercury (Hg), Lead (Pb), Nickel (Ni) and Zinc (Zn) (USEPA, 1997). However, when heavy metals find their route into aquatic environment (through burning of fossil fuels, mining and smelting of metallic ferrous ores, municipal wastes, fertilizers, pesticides and sewage sludge etc.) they are deleterious to fish and man; therefore, there is a need to remove heavy metals from the aquatic system (Peng *et al.*, 2006; Xiong, 1998). However, clean-up technologies have been developed for the removal of heavy metals but often these are expensive or are environmentally deleterious. Phytoremediation is a developing technology that can potentially address the problems of contaminated water, agricultural land or more intensely polluted areas affected by urban or industrial activities. It is a promising new method that uses green plants to assimilate or detoxify metals and organic chemicals (Pollard *et al.*, 2002).

At present, there are nearly 400 known hyperaccumulators but most are not appropriate for phytoextraction because of their slow growth and size. Several researches have screened fast growing, high biomass accumulating plant, including agronomic crops, for their ability to tolerate and accumulate metals in their shoots (Mellem *et al.*, 2012). In this study, water hyacinth (*Eichhornia crassipes*) was chosen because of its growth rate which is among the highest for any plant and the populations can double in as little as 12 days by sending off short runner stems which develop into new plants. If the plant is not utilized or harnessed, it can constitute several menace: Block waterways making fishing and recreation very difficult; limit boat traffic, flood control and wildlife use by shading and crowding out native aquatic plants. This exotic species by its action reduce biological diversity in aquatic ecosystems. Its proliferation can adversely affect fish populations, as vegetation continually falls to the bottom, decays and consumes oxygen that otherwise would have been available to aquatic fauna like fish (Moyo and Mapira, 2012).

## **MATERIALS AND METHODS**

**Plant species:** Water hyacinth (*Eichhornia crassipes*) were collected from Ologe Lagoon, Lagos State, Nigeria and brought to the Ecotoxicology and Aquatic Restoration laboratory of Department of Fisheries, Lagos State University, Ojo, Lagos State, Nigeria. The plants were rinsed in tap water to remove any epiphyte and insect larva on it. The set-up was in triplicate and experimental units were kept outside the laboratory for exposure to sun. Each experimental unit is a circular plastic bowl containing 9 L of water.

**Experimental procedure:** The experiments were conducted with three concentration gradients of Cu and Fe at 10, 15 and 20 mg L<sup>-1</sup> with a control. They were in 2 groups; the first group was not spiked with Cu and Fe (control), the second group, was spiked with Cu and Fe at 10, 15 and 20 mg L<sup>-1</sup> concentration gradients, respectively. The experiment lasted for 12 weeks (3 months).

**Sampling:** The plants were harvested at 2 weeks interval over 3 months period for heavy metals analysis. Plant samples were separated into roots, stems and leaves and then taken to the laboratory for analysis.

**Heavy metal analysis:** The parts of the plants (root, stem and leave) were crushed with the aid of a ceramic mortar and pestle. Two grams of each sample (root, stem and leaf) was measured (weighing balance (Mettler PM 400)) into a beaker. About 10 mL of concentrated Trioxonitrate (v)

acid (HNO<sub>3</sub>) and 5 mL of hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) were added. The mixture was heated on laboratory hot plates to get digested sample; distilled water was added to the mixtures to make up the volume to 20 mL. Extracts were kept separately in labelled 60 mL sample bottles and transported for heavy metals analysis. Metals analysis was carried out on the filtrate using Atomic Absorption Spectrophotometer (Perklin Elmer A Analyst 100).

**Translocation Factor (TF):** To evaluate the potential of *Eichhornia crassipes* for phytoextraction, the Translocation Factor (TF) was calculated. This ratio is an indication of the ability of a plant to translocate metals from its root to its aerial parts (Mellem *et al.*, 2012). It is represented by the equation:

$$TF = \frac{\text{Metal concentration in aerial parts}}{\text{Metal concentration in roots}}$$

Metals that are accumulated by plants and largely stored in the roots of the plants are indicated by TF values less than 1, with values greater indicates translocation to the aerial parts of the plant (Mellem *et al.*, 2009).

**Bioconcentration factor:** Bioconcentration Factor (BCF) was used to determine the quantity of heavy metal absorbed by plant from the water. This is an index used to measure the ability of the plant to accumulate a particular metal with respect to its concentration in the surrounding water medium and is calculated using the equation of Ghosh and Singh (2005):

$$BCF = \frac{\text{Metal concentration in tissue of whole plant}}{\text{Initial concentration of metal in substrate (water)}}$$

The higher the BCF value, the more suitable is the plant for phytoaccumulation (Blaylock *et al.*, 1997). The BCF values greater than 2 were regarded as high (Mellem *et al.*, 2009).

**Statistical analysis:** The SPSS software was used for statistical analysis of data. One way ANOVA was used to assess significant differences among the various copper and iron concentration in the plant parts (root, stem and leaf). The comparisons of mean using the least significant difference test were done when p-values (p<0.05) was considered significant.

## RESULTS

**Metals concentration in plant samples:** Table 1 shows the results of comparison of metals accumulation in leaves, stems and roots. The accumulation of Fe was highest in leaves of plant spiked with metals (Fe and Cu); concentration of Fe was 6.30±2.36 mg kg<sup>-1</sup> in treatment spiked with 10 mg metal/L of water, while the lowest mean value was obtained in the control (0.87±0.57 mg kg<sup>-1</sup>). However, accumulation in leaves of plant spiked with copper was highest (3.80±0.12 mg kg<sup>-1</sup>) at 20 mg L<sup>-1</sup> treatment and the lowest accumulation was obtained in the control (0.23±0.14 mg kg<sup>-1</sup>). The accumulation of metals in stems of plants spiked with Iron (Fe) gave the highest mean value of 7.13±3.83 mg kg<sup>-1</sup> at 15 mg L<sup>-1</sup> while the lowest was obtained in the control with the mean value of 0.65±0.21 mg kg<sup>-1</sup>. However, the highest accumulation of Cu

Table 1: Comparison of metal (Fe and Cu) accumulation in the leaves, stems and roots of *Eichhornia crassipes* in metal-spiked water

Concentration (mg L <sup>-1</sup> )	Leaves ----- (mg kg <sup>-1</sup> )-----	Stems ----- (mg kg <sup>-1</sup> )-----	Roots ----- (mg kg <sup>-1</sup> )-----
<b>For Fe</b>			
Control	0.87±0.57 <sup>a</sup>	0.65±0.21 <sup>a</sup>	1.48±1.08 <sup>a</sup>
10	6.30±2.36 <sup>b</sup>	6.30±3.90 <sup>b</sup>	10.20±2.02 <sup>b</sup>
15	4.42±1.13 <sup>b</sup>	7.13±3.83 <sup>b</sup>	9.37±2.44 <sup>b</sup>
20	5.10±1.47 <sup>b</sup>	6.32±2.39 <sup>b</sup>	11.22±6.69 <sup>b</sup>
<b>For Cu</b>			
Control	0.23±0.14 <sup>a</sup>	0.28±0.12 <sup>a</sup>	0.35±0.10 <sup>a</sup>
10	0.32±0.15 <sup>a</sup>	0.35±0.05 <sup>a</sup>	0.45±0.14 <sup>a</sup>
15	0.37±0.08 <sup>ab</sup>	0.30±0.15 <sup>a</sup>	0.45±0.16 <sup>a</sup>
20	3.80±0.12 <sup>b</sup>	0.30±0.13 <sup>a</sup>	0.37±0.19 <sup>a</sup>

Values in the same column and with the same superscripts are not significantly (p>0.05) different

Table 2: Translocation factor of metals (Fe and Cu) in leaf and stem of water hyacinth (*Eichhornia crassipes*)

Concentration (mg kg <sup>-1</sup> )	Leaves ----- (mg kg <sup>-1</sup> )-----	Stem ----- (mg kg <sup>-1</sup> )-----
<b>For Fe</b>		
10	0.66±0.15 <sup>a</sup>	0.64±0.17 <sup>a</sup>
15	0.49±0.57 <sup>a</sup>	0.77±0.18 <sup>a</sup>
20	0.68±0.27 <sup>a</sup>	0.76±0.26 <sup>a</sup>
<b>For Cu</b>		
10	0.81±0.22 <sup>a</sup>	0.81±0.67 <sup>a</sup>
15	0.78±0.08 <sup>a</sup>	0.72±0.32 <sup>a</sup>
20	1.12±0.12 <sup>a</sup>	1.09±0.19 <sup>a</sup>

Values in the same row and with the same superscripts are not significantly (p>0.05) different

(2.80±0.12 mg kg<sup>-1</sup>) occurred in the control while the lowest value (0.30±0.13 mg kg<sup>-1</sup>) was obtained in the stem of treatment spiked with 20 mg L<sup>-1</sup>. The root accumulated more Fe than the leaf and stem. The highest accumulation of Fe in the root (11.22±6.69 mg kg<sup>-1</sup>) occurred in treatment spiked with 20 mg metal/L of water, while the lowest value (1.48±1.08 mg kg<sup>-1</sup>) was obtained in the control. The root of water hyacinth in the treatment spiked with Cu at 15 mg L<sup>-1</sup> had the highest accumulation (0.45±0.16 mg kg<sup>-1</sup>) while the lowest value (0.35±0.10 mg kg<sup>-1</sup>) was recorded in the control.

**Translocation of metals (TF):** Table 2 shows the results of Translocation Factor (TF). The plants that were spiked with Fe had it highest TF value at 20 mg L<sup>-1</sup> (0.68±0.27) and the lowest TF value at 10 mg L<sup>-1</sup> (0.66±0.15 mg L<sup>-1</sup>) in leaf. The highest TF value in stem was obtained in treatment spiked with 15 mg metal/L of water (0.77±0.18) and lowest was recorded at 10 mg L<sup>-1</sup> (0.64±0.17). However, TF for Cu recorded the highest value (1.12±0.12) in treatment spiked with 20 mg Cu/L of water and lowest at 15 mg L<sup>-1</sup> (0.78±0.08) in leaf. In stem, the highest TF value for Cu was at 20 mg L<sup>-1</sup> (1.09±0.19) and lowest at 15 mg L<sup>-1</sup> (0.72±0.32).

**Bioconcentration factor of metals:** In this study, the Bioconcentration Factor (BCF) was used to determine the quantity of heavy metals absorbed by plant from the water. Bioconcentration factor is the ability of plant to accumulate a particular substance with respect to its concentration in water. Table 3 shows the BCF values of metals in leaf, stem and root. Plant treated with Fe had

Table 3: Bioconcentration factor of metals (Fe and Cu) in leaf, stem and root of water hyacinth (*Eichhornia crassipes*)

Concentration (mg L <sup>-1</sup> )	Leaves -----	Stems -----	Roots -----
<b>For Fe</b>			
10	1.39±0.22 <sup>a</sup>	1.40±0.35 <sup>a</sup>	2.27±0.81 <sup>b</sup>
15	0.66±0.07 <sup>a</sup>	1.07±0.23 <sup>ab</sup>	1.40±0.15 <sup>b</sup>
20	0.57±0.07 <sup>a</sup>	1.40±0.15 <sup>b</sup>	2.32±0.65 <sup>b</sup>
<b>For Cu</b>			
10	0.09±0.02 <sup>a</sup>	0.10±0.00 <sup>a</sup>	0.11±0.01 <sup>a</sup>
15	0.72±0.01 <sup>a</sup>	0.10±0.01 <sup>a</sup>	0.10±0.02 <sup>a</sup>
20	0.07±0.02 <sup>a</sup>	0.14±0.01 <sup>a</sup>	0.11±0.01 <sup>a</sup>

Values in the same row and with the same superscripts are not significantly ( $p > 0.05$ ) different

its highest BCF value in root (2.32±0.65) in treatment spiked with 20 mg Fe/L of water and the lowest in leaf (0.57±0.07) still at 20 mg Fe/L of water. However, when water hyacinth were spiked with Cu at 10, 15 and 20 mg L<sup>-1</sup>; their BCF values were highest in roots (0.11±0.01), leaf (0.72±0.01) and stem (0.14±0.01), respectively.

## DISCUSSION

According to Barazani *et al.* (2004) and Marchiol *et al.* (2004), for a plant to be considered for phytoremediation, it should have a few of the following traits: ability to extract, degrade or stabilize the contaminant, tolerance to high levels/concentrations of the contaminant, rapid growth rate and high biomass production, cosmopolitan growth and ease of harvesting. Water hyacinth removes nutrients and metals by assimilating them from water (Rogers *et al.*, 1991). The aquatic weed utilizes these in its metabolic processes such as protein and nucleic acid synthesis. These are also subsequently used for growth and other functions. The rate of uptake is influenced by factors such as: Growth rate, temperature and density of water (Sooknah, 2000). As these parameters increases so does the rate of uptake. The ability of water hyacinth to remove metals from water has been demonstrated in different studies (Ndimele, 2003; Kamal *et al.*, 2004; Akinbile and Yusoff, 2012; Gamage and Yapa, 2001).

**Phytoremediation:** Water hyacinth (*Eichhornia crassipes*) assimilation of nutrients and its subsequent harvesting is another mechanism for pollutant removal. Low cost and easy maintenance make the aquatic plant system attractive to use (Kanabkaew and Puetaipoon, 2004). Thus, aquatic plants are increasingly applied as a viable treatment for wastewater. The accumulation of metals in various parts of aquatic plants is often accompanied by an induction of a variety of cellular changes, some of which directly contribute to metal tolerance capacity of the plants (Prasad *et al.*, 2001).

This study showed the great potential of *Eichhornia crassipes* as an efficient remedy for pollution issues when properly harnessed and utilized. In Table 1, Iron (Fe) at 0 mg L<sup>-1</sup> (control) showed relatively low bioaccumulation (ranged from 0.65±0.21 to 1.48±1.08 mg kg<sup>-1</sup>) when compared with values in the treatments which showed higher bioaccumulation values (10 mg L<sup>-1</sup> ranged from 6.30±2.36 to 10.20±2.02 mg kg<sup>-1</sup>, 15 mg L<sup>-1</sup> ranged from 4.42±1.13 to 9.37±2.44 mg kg<sup>-1</sup> and 20 mg L<sup>-1</sup> ranged from 5.10±1.47 to 11.22±6.69 mg kg<sup>-1</sup>) in the leaves, stems and roots. However, the lowest accumulation of Fe occurred in the leaves, while the highest was in the roots. This observation is similar to that of (Mellem *et al.*, 2012), where they showed that

Cr at 25 ppm had no toxic effect but at higher concentrations such as 75 and 100 ppm, the plants showed signs that *A. dubius* tolerated high Cr concentrations as indicated by the high BCF index. Statistical analysis showed that there was no significant difference ( $p > 0.05$ ) in iron absorption among treatments but was significant between control and other treatments ( $p < 0.05$ ).

The lowest uptake of Cu ( $0.23 \pm 0.14 \text{ mg kg}^{-1}$  (control)) was found in the leaf, while the highest uptake ( $3.80 \pm 0.12 \text{ mg kg}^{-1}$  ( $20 \text{ mg L}^{-1}$ )) was in the root. Kalac and Svoboda (2000) reported that the lowest uptake of Hg, (3 ppm) was found to be in the leaves, while the roots had 666 ppm. There are several other studies that showed that plant roots accumulates higher metal concentration when they were exposed to contaminated medium (soil/water) (Godbold and Huttermann, 1988; Cavallini *et al.*, 1999; Blaylock and Huang, 2000). The results showed that there was no significant difference ( $p > 0.05$ ) in Cu uptake in different parts of water hyacinth except the leaf that had a remarkable difference of  $0.38 \pm 0.08 \text{ mg kg}^{-1}$  in  $15 \text{ mg L}^{-1}$  and  $3.80 \pm 0.12 \text{ mg kg}^{-1}$  in  $20 \text{ mg L}^{-1}$ . The study shows that while Fe uptake/bioaccumulation was highest in the root, it was best in the leaf for Cu. Therefore, it revealed that the plant, *E. crassipes* had some preference or select different metals in specific aerial parts. This is in agreement with Lokeshwari and Chandrappa (2006) who reported that the metal concentration in water and water hyacinth sample were found to vary greatly (Zn>Ni>Fe>Cu>Pb>Cd>Cr).

**Translocation Factor (TF):** All higher plants are capable of accumulating heavy metals in different concentration. However, a significant difference in metal accumulation exists among plant population. Water hyacinth is a cosmopolitan plant with a rapid growth rate, high biomass, easy to harvest and metal is sequestered from the water and transported to the aerial parts (Mellem *et al.*, 2012). Translocation Factor (TF) for metals (Fe and Cu) in leaf and stem revealed that *E. crassipes* had a poor translocation capability because most of the TF values were less than 1. Badr *et al.* (2012) and Mellem *et al.* (2009) opined that metals that are accumulated by plants are largely stored in the roots. Therefore, such plants have poor translocation capabilities, which is indicating TF values less than one ( $TF \leq 1$ ); and good translocation coefficient when greater than one ( $TF \geq 1$ ). In addition, statistical analyses showed that the TF values among treatments were not significantly different ( $p > 0.05$ ). This report revealed that TF values of Fe ranged from  $0.49 \pm 0.57$  to  $0.77 \pm 0.18$ . While in Cu, it ranged from  $0.72 \pm 0.32$  to  $1.12 \pm 0.12$ . This proves that Cu is better translocated in *E. crassipes* than iron (Fe) especially at higher concentrations. The present study has shown that water hyacinth has the ability to translocate Cu than Fe and this is in agreement with the finding of Badr *et al.* (2012) who reported that the translocation factor values for Cu are  $>1$  while that of Fe are  $<1$ . However, Lu *et al.* (2004) opined that the inability of water hyacinth to translocate metal may be due to some physiological barriers against metal transport to the aerial parts.

**Bioconcentration Factor (BCF):** According to Zhu *et al.* (1999) and Abd-Elmoniem (2003), the ratio between plant metal concentration and that of the growth media expresses the Bioconcentration Factor (BCF) which reflects the affinity of aquatic macrophytes to a specific heavy element or pollutant. Lu *et al.* (2004) mentioned that metal accumulation by macrophytes can be affected by metal concentration in water and sediments. BCF values greater than 2 are regarded as high (i.e. the ability of plant to absorb metal from water is high).

There was no significant difference ( $p>0.05$ ) in BCF values of treatment spiked with Cu. However, BCF varied significantly ( $p<0.05$ ) among treatments spiked with Fe. The highest BCF value for experimental units spiked with Fe was obtained in the root ( $2.32\pm 0.65$ ) at  $20\text{ mg L}^{-1}$  while the lowest was recorded in the leaf ( $0.57\pm 0.07$ ) also at  $20\text{ mg L}^{-1}$ . Conversely, BCF values in Cu was highest in the leaf ( $0.72\pm 0.01$ ), followed by the stem ( $0.14\pm 0.01$ ) and lowest in the root ( $0.11\pm 0.01$ ). This is in agreement with the finding of Ondo *et al.* (2012) who reported that the BCF values for Fe was higher in root than leaf while for Cu, it was higher in leaf than in root for both Roselle and Amaranth.

## CONCLUSION

This study has revealed that water hyacinth (*Eichhornia crassipes*) is able to accumulate high amounts of Fe in its root and Cu in its leaf. Therefore, metal content of water hyacinth can serve as a good bio-indicator of metal pollution in aquatic environments. The accumulation and translocation potential of Cu by water hyacinth was higher than that of Fe, indicating that water hyacinth can be primarily utilized as a good phytoaccumulator of Cu and Fe too. Therefore, water hyacinth was found to be a promising candidate for phytoremediation and adequate for biomonitoring programmes for metal-contaminated water.

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