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Bioremoval of Zinc in Polluted Soil using *Acalypha inferno*

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ABSTRACT

This study was to assess the Phytoextraction of *Acalypha inferno* for Zinc (Zn) contaminated soil. Stems were planted in five kilograms (5 kg) of the soil placed in each plastic pot having 0 ppm (control), 5, 10, 15, 20 and 25 ppm of Zn. The experiment was on for a period of 12 weeks. The results revealed that pH, phosphorous and moisture contents increased while nitrogen and organic carbon contents decreased in polluted soil remediated with *Acalypha inferno*. The plant compartments were analyzed for Zn uptake. Appreciable concentrations of Zn in different compartment of the plant was recorded, 7.12, 7.10 and 9.06 ppm for stem, root and leave, respectively. Bioconcentration Factor (BCF) and Translocation Factor (TF) was assessed. It was observed that more concentration of Zn was translocated from the roots to the leaves. The results obtained suggest that *Acalypha inferno* have phytoextraction ability and could be used in restoring soil polluted with zinc (Zn).

Key words: Heavy metal, zinc, phytoremediation, *Acalypha inferno*, phytoextraction, physicochemical, plant

INTRODUCTION

Heavy metals contaminate the agriculture use of soils by atmospheric deposition or by dumping of sewage sludge. This resulted as a risk of either leaching of metals into the groundwater or too much accumulation in the top layer of soil (Adriano, 1992). The amount of metal concentrations in soil ranges from less than 1 ppm to high as 100,000 ppm, even if it occurs due to the geological origin of the soil or may be as a result of human activity (Blaylock and Huang, 2000). Over indulgence concentrations of some heavy metals in soils, such as Cd, Cr, Cu, Ni and Zn have caused the interruption of natural aquatic and terrestrial ecosystems (Meagher, 2000). High amount of Cu and Zn in plants can caused some trouble. Phytoremediation is an emerging technology, which should be considered for remediation of contaminated sites because of its cost effectiveness, aesthetic advantages and long term applicability (Boonyapookana *et al*., 2005).This technology can be defined as the efficient use of plants to remove, detoxify or immobilize environmental contaminants in a growth matrix (Soil, Water or Sediments) through the natural, biological, chemical or physical activities and processes of the plants (Ciura *et al*., 2005). An appropriate plant for phytoremediation should ideally have high and fast biomass production and ability of translocation of contaminants into the plant shoot (Cunningham and Ow, 1996).

Unlike organic compounds, metals cannot be degraded and their cleanup requires their immobilization and toxicity reduction or removal. In recent years, scientists and engineers have

started to generate cost effective technologies which includes use of microorganisms/biomass or live plants for cleaning of polluted areas (Qiu *et al*., 2006; Kuzovkina *et al*., 2004). Phytoextraction (uptake) is the use of living green plants in order to remove inorganic contaminants, primarily metals, from polluted soils and concentrate them into roots and easily harvestable shoots (Lasat, 2002; Tang *et al*., 2003). Phytoremediation can be used to remove not only metals (e.g. Ag, Cd, Co, Cr, Cu, Hg, Mn, Mo, Ni, Zn) but also radionuclides (e.g., 90 Sr, 137 Cs, 239 Pu, 234 U, 238 U) and certain organic compounds (Andrrade and Mahler, 2002). Plants have shown the capacity to withstand, relatively high concentration of contaminants without toxic effects. Plant uptake of contaminants can be increased by the use of transgenic plants (Bhargava and Srivastava, 2014) or by the inoculation of engineered endophytic bacteria (Bell *et al*., 2014). Some studies aim at enhancement of phytoremediation by improving the contaminant mobility and bioavailability in soils by adding suitable chelating agents or surfactants. But in such cases, there is a possibility of contamination of the soil and ground water by the chemicals used for mobilizing the contaminants. Also, the mobilized contaminants can migrate to the ground water, thus by contaminating the ground water and also spreading the contamination. In the present study we examine the potential of *Acalypha inferno* plant for phytoremediation (uptake) of heavy metal zinc.

MATERIALS AND METHODS

Samples collection and processing: The soil sample used for this study was collected from a depth of 0-20 cm within the Federal University of Technology, Minna and transported in plastic pots to the experimental garden. The soil sample was air-dried and pre-sieved with 2 mm diameter mesh. The physicochemical properties of soil used for the study is presented elsewhere. The taxonomic classification of the experimental soil was loamy sand with pH of 6.57. Mature stem of *Acalypha inferno* was collected at Centre for Preliminary and Extramural Studies (CPES) Garden of the Federal University of Technology, Minna, Nigeria.

Preparation heavy metal contaminant: The contaminants were added as zinc sulphate ($Zn(SO_4)$ and 2.4771g of $Zn(SO_4)$ was dissolved in 1,000 mL of distilled water to make stock solutions of 5, 10, 15, 20 and 25 mL. These different concentrations were then measured from the stock solutions into a 100 mL capacity measuring cylinder and made up to the mark to give 5, 10, 15, 20, 25 and 0 ppm (control) metal concentrations. The soil was spiked with different concentration of zinc and it was thoroughly mixed (Kabata-Pendias and Pendias, 2001).

Experimental setup and treatment: Pot experiment study was conducted at Centre for Preliminary and Extra-mural Studies (CPES) Garden of the Federal University of Technology, Minna, Nigeria. The set up was a complete randomized design and the treatment was replicated three times. The experimental pots were filled with 5 kg soil pre-sieved with 2 mm sieve size. Then the stems (one stem per pot) were planted in each pot. Sampling of the plants to monitor metal uptake and soil for residual metal contents was done 12 weeks after planting. The plants were irrigated with 200 mL (per pot) of tap water daily and sampling of the plants to monitor metal uptake and soil for residual metal contents was done 12 weeks after plant.

Analysis for zinc contamination: After 12 weeks of planting, all the plants were harvested separately, according to soil treatment, separated into three compartments, viz. roots, stem and leaves. The 3 replicates of each treatment were pooled together to give composite sample of each

treatment. The plants were washed in water to eliminate dust, dirt, possible parasites or their eggs and finally with deionized water (Yusuf *et al.*, 2003). Each sub-sample was oven-dried at 70°C for 24 h. Acid digestion method of Yusuf *et al.* (2003) was used for the digestion of grounded plant samples. One gram each of this was weighed into 50 mL capacity beaker, followed by addition of 10 mL mixture of analytical grade acids: HNO_3; H_2SO_4; $HClO_4$ in the ratio 1:1:1. The beakers containing the samples were covered with watch glasses and left overnight. The digestion was carried out at a temperature of 70°C until about 4 mL was left in the beaker. Then, a further 10 mL of the mixture of acids was added. This mixture was allowed to evaporate to a volume of about 4 mL. After cooling, the solution was filtered to remove small quantities of waxy solids and made up to a final volume of 50 mL with distilled water. Zinc concentrations were determined using Atomic Absorption Spectrophotometer (AAS), Accusys 211.

Determination of bioconcentration and translocation factor: Bioconcentration Factor (BCF) and Translocation factor (TF) were calculated using the formula of Yadav *et al.* (2009):

$$\text{Bioconcentration factor (BCF)} = \frac{\text{Average metal concentration in the whole plant (ppm)}}{\text{Metal concentration in soil (ppm)}}$$

$$\text{Translocation Factor (TF)} = C_{aerial} \times 1/C_{root}$$

C_{aerial} = Metal concentration in the aerial part of plant (stem and leaf)
C_{root} = Metal concentration in root of plant

Statistical analysis of data: Statistical analysis was performed using the SPSS (version 16). Differences in heavy metal concentrations were detected using One-way Analysis of Variance (ANOVA).

RESULTS AND DISCUSSION

Concentration of zinc in soil remediated with *Acalypha inferno*: Figure 1 shows the concentration of zinc in the zinc free soil and zinc polluted soil remediated with *Acalypha inferno*. The residual Zn concentration in soil obtained after harvesting *Acalypha Inferno* were 1.12, 0.86, 1.44, 1.25 and 0.88 ppm for 5, 10, 15, 20, 25 ppm, respectively.

The residual concentration of zinc in soil 3 days after pollution indicated that the zinc was present in the soil proportionate to the amount added (Fig. 1). After 12 weeks, Zn concentrations decreased when compared with the initial addition of zinc in the soil remediated with *Acalypha inferno*. The reason for the reduction in the concentration of Zn, because the plant had phytoextraction potential to remove the heavy metal from the soil.

Zinc in harvested parts of *Acalypha inferno:* Figure 2 shows the concentration of zinc in *Acalypha inferno* plant parts. Generally, Zn concentrations in all the plant compartments increased with the developmental stages of the plants.

The concentrations of Zn after 12 weeks for leaf compartment were, 1.41, 1.90, 2.64, 1.34 and 1.77 ppm, roots; 0.71, 1.38, 1.93, 1.63 and 1.45 ppm, while 0.69, 1.38, 1.97, 1.64 and 1.44 ppm were observed in the stems at 5, 10, 15, 20 and 25 ppm, respectively. The results indicated that *A. inferno* mopped up substantial concentrations of Zn in the above-ground biomass compared to

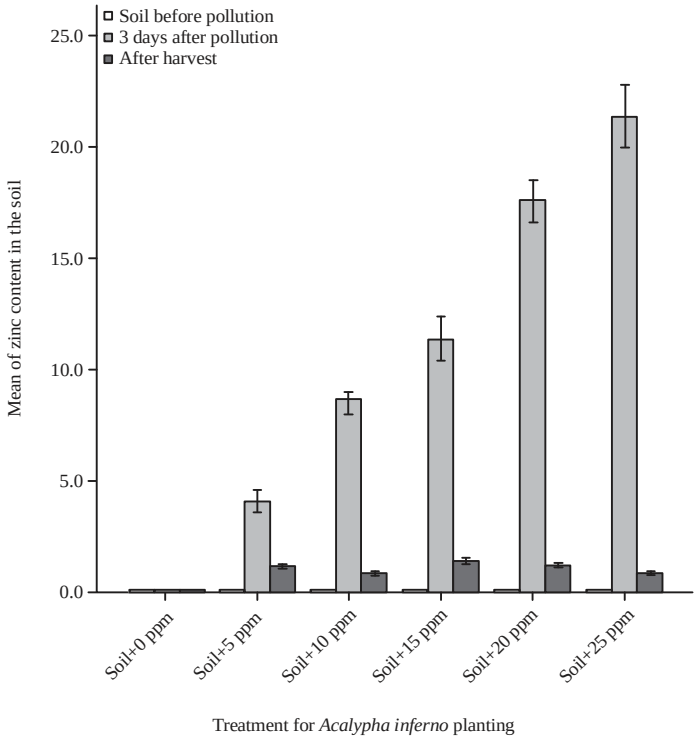

Fig. 1: Zinc content in the experimental soil of *Acalypha Inferno,* Error bars: 95% Cl

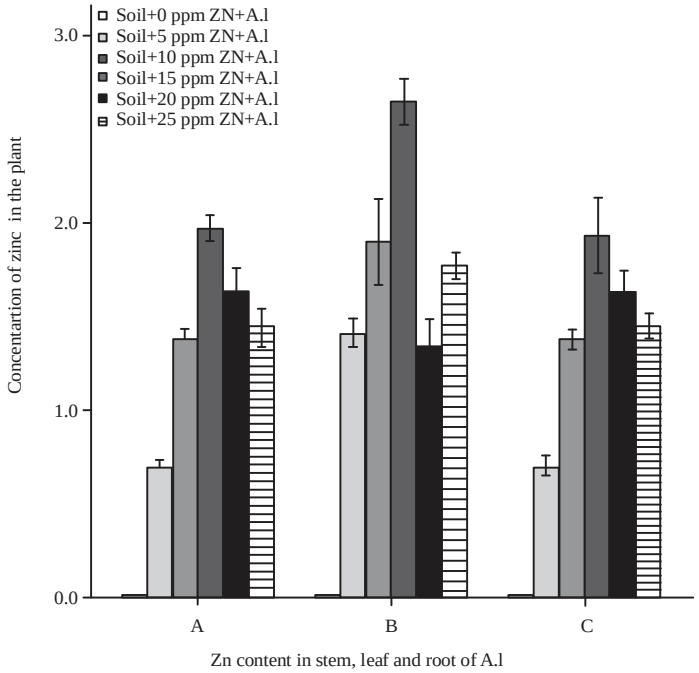

Fig. 2: Zinc concentrations in root, stem and leaves of *A. inferno* harvested from Zn contaminated soil after 12 weeks, Error bars: 95% Cl, Keys: A: Stem, B: Leaf and C: Root, Ai: *Acalypha inferno*

Table 1: Bioconcentration Factor (BCF) and Translocation (TF) of Zinc in *Acalypha inferno* remediated soil

Treatment	BCF	TF (In leaves)	TF (In stem)
Soil +0 ppm Zn	ND	ND	ND
Soil +5 ppm Zn	0.84	1.96	0.96
Soil +10 ppm Zn	2.09	1.38	1.00
Soil +15 ppm Zn	1.51	1.62	1.02
Soil +20 ppm Zn	1.23	0.82	1.01
Soil +25 ppm Zn	1.77	1.22	0.99

ND: Not Detected

concentrations in the roots. Results also showed that, at the end of twelve weeks period, the leaves accumulated the highest concentration of Zn followed by the stems and the roots. The high Zn contents in the leaves might be attributed to the high level of zinc in the soils; this is possible, because plants absorb metals based on their availabilities in the soil except the highest concentrations, where slight changes were recorded (Benzarti *et al*., 2008). Since, the leaves of *A. inferno* mopped up the highest concentrations of Zn after 12 weeks, it implies that the efficiency of the plant in cleaning Zn contaminated soil was effective at all the stages of the plant growth.

Based on the results reported in Fig. 2, it is clear that *Acalypha inferno* can take up considerable amount of metal to the plant tissue as reported in the literature (Kalisova-Spirochova *et al*., 2003). Besides, the results show that zinc is present more in leaves and in that more in the stem than the roots. This result is in agreement with an early previous study who reported that zinc is accumulating more in shoots and in that more in the leaves than the stem.

Bioconcentration factor and translocation factor of zinc in *Acalypha inferno* remediated soil: Table 1 shows the Bioconcentration Factor (BCF) and Translocation Factor (TF) of Zn in *Acalypha inferno* plant. It has been reported that the level and impact of heavy metals on the environment is greatly dependent on their speciation in soil solution and solid phase, which determine their environmental availability, geochemical transfer and mobility pathways (Pinto *et al.,* 2004).

The highest BCF was recorded in soil polluted with 10 ppm Zn with 2.09 and 0.84 was recorded as lowest BCF value in soil polluted with 5 ppm. The highest TF in leaves and stems were recorded in soil polluted with 5 and 15 ppm, respectively (1.96 and 1.02) There was no significant difference between the TF of Zn in the stem and leaves of *Acalypha inferno* at $p<0.05$ significant level (Table 1).

The results in this study showed that *Acalypha inferno* at all concentrations except at (5 ppm) had BCF values >1, indicating that the plant had the potential to be used as accumulators of Zn. On the other hand, TF values of *Acalypha inferno* in all treatments (except 20 ppm in leaves and 5, 25 ppm in stem) showed values >1, indicating that it could be an high efficiency plant for Zn translocation from root to the shoot. In general, plants that have BCF and TF values of >1 are sought for heavy metal extraction (Alkorta *et al*., 2004). Results from this experiment however, showed high BCF and TF values. This means that *Acalypha inferno* has the ability to store Zn in the leaves and the high transpiration rate indicates that this species has high potential as a phytoremediator. Therefore, could be used for phytoremediation of Zn.

CONCLUSION

This study shows the ability of *Acalypha inferno* to remediate Zn in contaminated soil. The plants generally had the highest concentrations of Zn in their leaves at 12 weeks of remediation.

This implies that the efficiency of this plant in cleaning the contaminated soils was at the development stage of the plant growth. The Zn could be potentially remediated from soil using *Acalypha inferno* and could even do more if possibly enhanced.

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