



PHYTOREMEDIATION STUDIES OF SELECTED PLANT SPECIES ON BIOAVAILABILITY AND HYPER ACCUMULATION OF CADMIUM FROM CONTAMINATED SOIL

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ABSTRACT

Soil and water contaminated with toxic heavy metals posed significant environmental and human health hazards that are still in need of an effective and affordable biotechnological solution. Phytoremediation analysis of soil sample contaminated with cadmium has been carried out using different species of plants with the aim of finding a solution towards a method that could effectively mitigate the degree of soil contamination due to toxic heavy metals. This process was achieved through a controlled experiment carried out in a green house. The soil sample used was treated accordingly and spiked with 1% of cadmium in form of cadmium nitrate to achieve a significant level of contamination. Ten different plant species were planted separately in a plastic vase containing the contaminated soil and allowed to grow along side with control for the period of eight weeks. After harvesting, the plants were sorted into root, stem and leaves. The cadmium content of each part was analysed using Atomic Absorption Spectrophotometric (AAS) method of analysis. The result indicated that the shoot of *Ricinus communis* accumulated the highest concentration of cadmium ($686.463 \pm 0.0028\text{mg/kg}$), followed by that of *Azadirachta indica* ($678.888 \pm 0.0031\text{mg/kg}$) and *Senna occidentalis* demonstrated the lowest level of cadmium accumulation ($11.650 \pm 0.0005\text{mg/kg}$). But with respect to the accumulation in the root and stem, *Azadirachta indica* has shown the highest cadmium levels of $533.577 \pm 0.0035\text{mg/kg}$ and $582.013 \pm 0.0018\text{mg/kg}$ respectively, while the lowest level in the root was recorded in *Amaranthus spinosa* ($30.922 \pm 0.0003\text{mg/kg}$) and the stem of *Jatropha curcas* accumulated the least mean level of $23.634 \pm 0.0007\text{mg/kg}$. The results obtained indicated that the concentrations of cadmium in the parts of the plants grown in the contaminated soil were greater than those grown in the unspiked soil as control. It is therefore suggested that the analysed plant species could be potentially useful in the immobilisation, phytoextraction and subsequent removal of cadmium from the contaminated soil.

Keywords: Phytoremediation, Cadmium, AAS, Contaminated soil, Bioavailability, Hyper accumulation

INTRODUCTION

Phytoremediation is the use of vegetation or plants to contain, sequester, remove or degrade inorganic or organic contaminants in soil, sediments, surface water and ground water (Tsao, 2003). The process is a

biotechnological technique that is environmentally friendly, safe and cost effective used to eliminate pollutants from an environment. The phytoremediation technique is basically subdivided into five different categories: phytoextraction, phytostabilization,

phytovolatilization, phytofiltration and phytodegradation (USEPA, 2000).

The enormous growth of artisanal mining, use of agro allied chemicals, industrialization, heavy traffic congestion and other anthropogenic activities have resulted into serious environmental pollution in which soil is among the most affected area. The accumulation of toxic heavy metals in soil, surface and ground water not only affect the natural sources but also poses a significant impact on ecosystem and human health. Thus, the presence and bioaccumulation of toxic heavy metals in our environment have become a major area of great concern. Contaminants such as cadmium and other heavy metals penetrate the environment through industrial waste, use of agro allied chemicals and land fill run off (Adriano, 1986; Sharma and Sachdeva, 2015).

Exposure to heavy metals in low concentration has been found to cause kidney damage, liver damage and anaemia while in high concentration, heavy metals can be carcinogenic and teratogenic (McCluggage, 1991). Despite the regulation of the industrial waste by the Environmental Protection Agency (EPA), most of the wastes exceed the regulated amount to be disposed. Thus, when these are released into the environment, they naturally concentrate in wet lands and soils. The natural process of transportation of metal ions between soil and water consolidate heavy metal contamination in concentrations that affect the natural environment (Ernst, 1996).

As a result of increasing environmental concern with regards to heavy metal contamination, it has become apparent to develop a method or to improve on the current method of removing such toxic metals from the contaminated soil environment. The extent of environmental pollution due to heavy metals has gone to the level that can cause serious effect on our health. Hence, there is an urgent need to carry out some investigations

that would bring about an improvement on the existing conventional methods of technology so as to effectively control, minimize or mitigate the effect of such accidental environmental pollution which has a serious impact on our health.

The current conventional methods of soil remediation have only been focused on the use of physical and chemical immobilization products that alter the heavy metals present in the contaminated soil to a less soluble and bioavailable forms (Robinson *et al.*, 2003). These methods are environmentally destructive, highly expensive and difficult to implement. It is therefore pertinent and the responsibility of all concerned to devise another means or method which is environmentally friendly and cost effective for the extraction and removal of toxic heavy metals from the contaminated soil as well as water. Therefore, if phytoremediation techniques are employed and applied effectively, our indigenous plants will no doubt have a great role to play in sanitizing our environment with respect to heavy metals contamination. Also since they cannot be degraded nor destroyed (Chen and Chen, 2001), heavy metals can be effectively immobilized, extracted and finally removed through a biotechnological process of phytoremediation. It is a well-known fact that our environment has been blessed with different varieties of plant species, thus phytoremediation make use of this natural opportunity to isolate, extract and remove the toxic heavy metals that are persistent in the environment. Therefore, the aim of this research was to evaluate the phytoremediation potentials of the selected indigenous plant species towards the remediation of cadmium contaminated soil.

MATERIALS AND METHODS

Soil collection and Treatment

The soil used was obtained as a composite sample top soil from a remote agricultural environment in Zaria and stored accordingly. The soil sample collected was air dried, ground, and sieved to obtain a desired particle size. The soil was divided into ten different portions of about 5kg as described by Vwioko *et al.*, 2006. Each of these portions was spiked with 500ml of 1% solution of cadmium inform of cadmium nitrate (Reddy and Chinthamreddy, 2000) and incubated for a period of 2 weeks to simulate artificial contamination with cadmium. Each of the treated soil sample was put in perforated plastic vase and placed appropriately in a screening house. A control medium was also set up for each of the plants.

Plant Sample Collection

Ten healthy young growing plant samples each with a height of 20 - 30 cm were obtained from the Kamacha botanical garden in Zaria metropolis. These plants were transferred and planted into each of the respective contaminated cadmium soil according to the method described by Tu and Ma (2002). All the conditions necessary for the continuous growth and survival of these plants were provided accordingly.

Growing and Harvesting of the Plants

After planting, the vases containing the plants were watered every other day with tap water. The plants were allowed to grow for a period of eight weeks alongside the control in a green house. The plant in each contaminated soil was carefully uprooted and gently teased to remove any of the soil particle attached to it. The root of each plant was gently washed

with water and then rinsed with distilled water (Reuter *et al.*, 1988). The three parts of the plants (root, stem and leaves) were removed separately and oven dried at 90⁰c for two days.

Sample Digestion and Analysis of the Cadmium Contents

The root, stem and leaves of each plant were ground separately into powdered form using pestle and mortar. One gram (1g) of each part was weighed into a clean ceramic crucible, transferred into a murfle furnace and then heated at 500⁰C for 5 hours (Allen *et al.*, 1986). The residual ash of each part was put into a separate clean beaker of 100 ml capacity followed by the addition of 20 ml of aqua regia. The resulting mixture was heated on a hot plate at about 80⁰C for 20 minutes. The digest was allowed to cool, 10ml of deionised water was added and then filtered using a Whatman No. 1 filter paper. The filtrate of the digest was made up to 50 ml with distilled water. Working standards were prepared by the serial dilution of 1000 ppm Cadmium stock solution. The cadmium contents of each part of the digested plant samples were analyzed using Atomic Absorption Spectrophotometer equipped with computer accessories and Cadmium hollow cathode lamp (228.8 nm).

RESULTS

Table 1: Result of Soil Characterization

Parameters analysed	Corresponding values
pH	6.70 ± 0.31
Moisture content (%)	0.604 ± 0.52
Cation Exchange Capacity (cmolkg ⁻¹)	14.90 ± 0.04
Organic Matter (%)	3.279 ± 0.08
Electrical Conductivity (dsm ⁻¹)	0.013 ± 0.72
Cadmium content (mg/kg)	0.362 ± 0.14

Table 2: Comparative Mean Concentrations of Cadmium in Different Parts of the Plants Grown on Cadmium Contaminated Soil

Plant species	Concentration (mg/kg)			TF*
	Root	Stem	Leaves	
<i>Azadirachta indica</i>	533.570±0.0035	582.010±0.0018	678.880 ±0.0031	1.3
<i>Eucalyptus citodora</i>	335.400±0.0017	102.600±0.0006	384.160±0.0007	1.1
<i>Ricinus communis</i>	276.530±0.0006	210.540±0.0011	686.460±0.0028	2.5
<i>Calotropis procera</i>	443.710±0.0032	348.140±0.0006	113.300±0.0007	0.3
<i>Eugenia jambolana</i>	168.090±0.0050	53.000±0.0024	57.000±0.0021	0.3
<i>Senna occidentalis</i>	212.550±0.0005	43.870±0.0003	11.650±0.0005	0.05
<i>Senna siamae</i>	484.770±0.0032	459.800±0.0011	333.860±0.0025	0.7
<i>Amaranthus spinosa</i>	30.920± 0.0003	62.230±0.0001	60.550±0.0009	2.0
<i>Khaya senegalensis</i>	102.400± .0007	381.160 ±0.0027	146.640±0.0008	1.4
<i>Jatropha curcas</i>	33.200± 0.0004	23.630 ±0.0007	19.400±0.0002	0.6

*TF- Translocation Factor

Table 3: ANOVA

	Concentration of Cadmium (mg/kg)				
	Sum of Squares	df	Mean Square	F	Sig.
Parts	6425.136	2	3212.568	.068	.935
Error	1283184.399	27	47525.348		
Total	1289609.535	29			

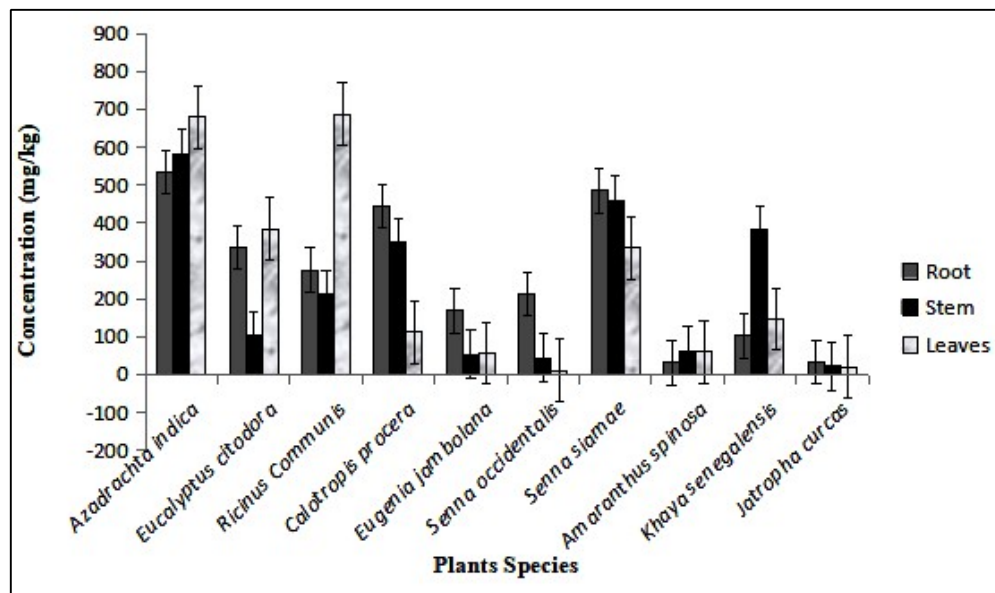


Figure 1: Plot of Mean Concentrations of Cadmium (mg/kg) in the root, stem and leaves of the plants grown in the contaminated soil.

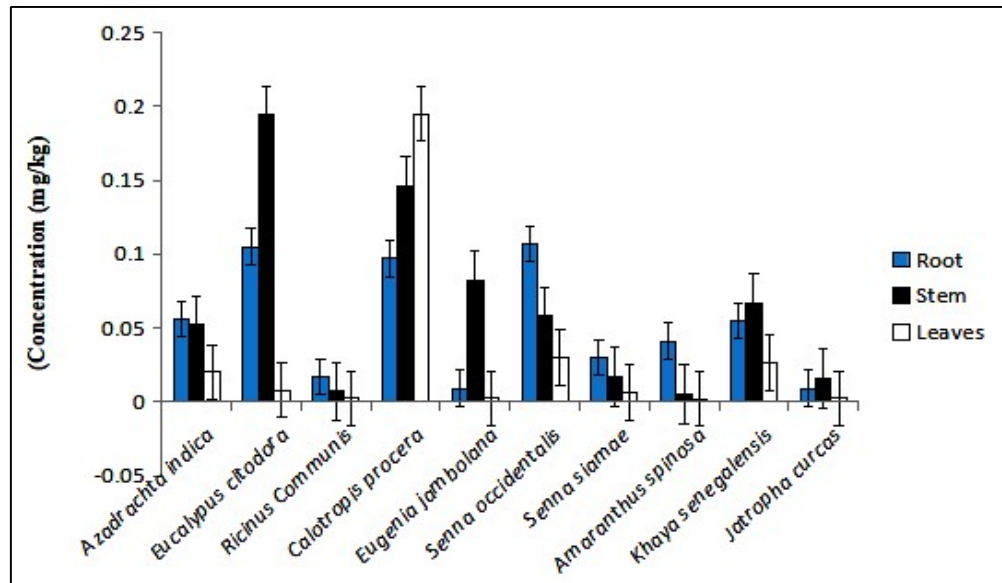


Figure 2: Plot of Mean Concentrations of Cadmium in the root, stem and leaves of plants grown in unspiked soil as control.

DISCUSSION

Phytoremediation study was carried out to investigate the potentials of some indigenous plants towards the remediation of cadmium contaminated soil. The corresponding results obtained were tabulated and represented accordingly. Table 1 indicates the results of the physicochemical properties of the soil being used in the analysis. The pH of soil influences the bioavailability of metals in the soil matrix. Bioavailability of metals in soils usually decreases when the pH is above 6.0 (Prasad, 2003). The percentage organic matter of $3.279 \pm 0.08\%$ is still higher than values found in soils of the Nigerian savannah region which ranges from 0.8 to 2.9% (Yashim *et al.*, 2016). The result for cation exchange capacity (CEC), $14.90 \pm 0.04 \text{ cmol/kg}^{-1}$, was found to be within the normal range (3 - 25 cmol/kg) for soils low in organic matter (Yashim *et al.*, 2016). But a soil with high clay content or organic matter (OM) usually develops a greater CEC at near-neutral pH than under acidic conditions (Donald, 2000). The mean concentration of cadmium in the experimental soil was found to be lower than the level of other heavy metals such as lead.

Table 2 indicates the levels of cadmium absorbed and accumulated in various parts of the plants examined. It has been generally observed that all the plants used in the experiments were able to survive and successfully grow on the cadmium contaminated soil. It has also shown that the cadmium has been absorbed from the contaminated soil and subsequently translocated to the shoots and leaves in a similar fashion as hyper-accumulator species. The level of accumulation of cadmium in the leaves ranges from 11.650 - 686.460 mg/kg indicating the highest level of accumulation in the whole parts of the analysed plant species. On the individual basis, *Ricinus communis* was found to accumulate the highest level of cadmium in the leaves (686.460 mg/kg) followed by *Azadirachta indica* (678.880 mg/kg) while *Senna occidentalis* accumulated the lowest concentration (11.650 mg/kg). This is not surprising as similar case has also been reported (Yashim *et al.*, 2016). The level of accumulation of cadmium in the stem ranges from 23.630-582.010 mg/kg showing that stem also plays a significant role in the phytoremediation

process of cadmium contaminated soil. With respect to the root the level of absorption ranges from 30.920-533.570 mg/kg, also indicating that roots are very vital in the phytoextraction process of cadmium from the contaminated soil. *Azadirachta indica* accumulated the highest concentrations of cadmium in both root and stem, even in the leaves the level of accumulation was just a little bit below the highest concentration (Table 2).

The emergence of *Azadirachta indica* as the overall accumulator of cadmium from the contaminated soil might be ascribed to the genetic ability of the plant to extract, immobilize, absorb and translocate the cadmium into its shoot and leaves. This is in line with the report that some plants have genetic potential of extracting and subsequent accumulation of significant quantities of heavy metals in their above ground shoots (Salt *et al.*, 1995 and Mitch, 2002). The values of translocation factors in table 2 indicate clearly that 50% of the selected and analysed plants have the TF values greater than 1. Hence half of these plants are hyper-accumulators of cadmium. It has been reported from a related literature that for a plant to be regarded as hyper accumulator it must have a TF value greater than 1 (Hakanson, 1980). This is a great achievement because most of the related researches conducted on this aspect have only reported few cases of plants with $TF > 1$.

Comparing the concentrations or levels of accumulation of cadmium in the various parts of the different plants, the one-way analysis of variance (ANOVA) was applied for the test (Table 3). From the result, $p = 0.935 > 0.05$, indicating that there is no significant difference between the mean concentrations of cadmium absorbed and accumulated in different parts of the plants. This has therefore indicated that most of the indigenous selected plant species have relatively accumulated

significant amount of cadmium in their various tissues with significant level of correlation.

CONCLUSION

Careful observation of the magnitude of the results obtained has indicated that most of the plants used in this study possess the ability to immobilize, absorb and subsequently bioaccumulate significant level of cadmium in their respective tissues. It is certain that when such plants are secured and allowed to grow for longer period of time they would definitely extract and bioaccumulate remarkable and excessive amount of cadmium, thereby reducing the level of such metal from the contaminated soil. Since *Azadirachta indica* has accumulated the highest level of cadmium in its root, the potential of this plant would therefore be more effectively harnessed in the process of phytostabilization of the metal and to some extent could also serve as hyperaccumulator. Similarly, *Ricinus communis* has a greater potential to be used as cadmium hyperaccumulator being the plant that accumulated highest level of cadmium in its above ground shoot. It is therefore hoped that the findings of this work could be effectively employed and extrapolated as an alternative method of mitigating the hazardous effect as well as the level of cadmium in our environment.

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