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EFFECT OF CHANGES IN SOIL CATION EXCHANGE CAPACITY ON THE RECLAMATION OF LEAD BY *ELEUSINE INDICA* (L.) GAERTN

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ABSTRACT

Contamination of soil by various heavy metals such as lead poses a serious environmental problem. Lead (Pb) is one of the harmful and frequently encountered heavy metals that is uptake by plants. Cation Exchange Capacity (CEC) is one of the important factors influencing metal uptake and accumulation. Rate of this uptake causes a number of toxic symptoms in plants. This study was carried out to evaluate the effect of changes in CEC on accumulation of lead by *Eleusine indica.* Seven soil CEC ratios were obtained by mixing clay (C) and humus (H) soils (C, 90C:10H, 75C:25H, 50C:50H, 25C:75H, 10C:90H, H) in two replicates. The soil was polluted with 675.2mg/kg of Pb and *Eleusine* plants were transplanted into the 5kg soil. Results showed that CEC of the soils influenced the accumulation of Pb in the root of test plant. Highest CEC (0.710 meq/L) was obtained in the H soil with highest Pb root uptake (29.5mg/kg), while the lowest CEC (0.210 meq/L) was obtained in C soil with low Pb root uptake (12.5mg/kg). The CEC was observed to increase with addition of H ratio. Significant reduction ($P<0.05$) in plant height, leaf length and leaf area were recorded in the C soils polluted with Pb. Furthermore, 5 additional tillers were observed in plants sown in Pb-polluted H soils. However, no additional tillers were obtained in the Pb-polluted C soils. This may suggest that *Eleusine* plant flourish more and accumulate higher Pb in H rich soils. This approach can be employed to improve remediation of Pb pollution.

Keywords: Soil cation exchange capacity, Reclamation, Lead, *Eleusine indica.*

INTRODUCTION

Heavy metals are naturally occurring toxic metals, having a density of more than 6 g cm^{-3} and atomic weight of more than iron (Musa et al., 2019). They are naturally occurring elements with high density and high atomic weight. Their density is at least five times greater than water (Tchounwou et al., 2014). The distribution of heavy metals in the environment is governed by the properties of the metal (Khlifi and Hamza-Chaffai, 2010). They can be found forming complexes in the soil and water. Sources of heavy metals in soil include emissions from the rapidly expanding industrial areas, disposal of high metal wastes, leaded gasoline and paints, land application of fertilizer etc.

Soils are the major sink for environmental heavy metals. Compared with organic contaminants which can be degraded, most heavy metals cannot be degraded by chemical or microbial means (Wuana and Felix, 2011). Heavy metals remain in soil for a very long time after their introduction and they have a residual time ranging from a few to several hundred years (Kabata, 1995). Urban soils are frequently polluted with heavy metals such as lead, zinc, cadmium and copper. These heavy metals can find their ways into arable lands through run-off or as synthetic fertilizers (Merrington and Alloway, 1994). On land, heavy metals easily contact plants through their roots.

Generally, heavy metals are toxic to plants. They have been documented to cause inhibition of cytoplasmic enzymes and disruption of enzyme activities that are important in plant metabolism. They can also replace or block essential metals and nutrients in plants roots (Chibuike and Obiora, 2014). Reduction in plant growth is the primary symptom of metal toxicity in plants. High heavy metal concentrations could also lead to the death of the plant. Some heavy metals however, such as zinc are required in small amounts for plant growth and development. Different plants have different ways of responding to heavy metals toxicity and toxic levels (Abolghassem et al., 2015).

Heavy metal contamination of soil has become worldwide problem because of the threat it poses to the ecosystem (Liu et al., 2018; Musa and Ikhajiagbe, 2019). For this purpose, researchers have developed remediation techniques involving physical, chemical and biological methods. Of these methods, biological methods are considered most effective and eco-friendly (Liu et al., 2018). Biological method otherwise, bioremediation is the use of living organisms such as plants and microorganisms to clean up soil (Sardrood, 2013). When plants are used, the technique is referred to as phytoremediation.

Phytoremediation use higher plants to remove heavy metals from soil or stabilize them into less harmful forms (Mahood et al., 2015). Several plants have earlier been studied on their phytoremediation capacity, by assaying for their ability to uptake and accumulate heavy metals in their roots and shoots.

Eleusine indica is one of the plants that has been widely used in phytoremediation. It has the ability to grow in harsh environmental conditions, which include metal contaminated soils. Lu et al. (2010) demonstrated the abilities of *Eleusine* to clean up petroleum contaminated soils in a greenhouse experiment. Results showed that the *Eleusine* plants reduced the contamination by 47%. This shows *Eleusine* as hyper accumulator of heavy metals such as lead. Lead (Pb) has been established as one of the harmful and frequently encountered heavy metals that is uptake by plants (Farouk and Muhammad, 2018). The uptake is mainly dependent on the soil pH, soil texture, redox potential, concentration of metal in soil, specie of plant, plant age, particle size, root exudation and cation exchange capacity (CEC). Hazleton and Murphy (2017) reported that humus soil with rich organic matter has the highest CEC level. This study aimed at investigating the effect of changes in CEC on accumulation of Pb by *Eleusine indica,* with a view to selecting best soil condition for Pb uptake and reclamation.

MATERIALS AND METHODS

Experimental site

This research was conducted in the botanical garden of the Department of Plant Biology and Biotechnology in the University of Benin, Benin, Edo state. Plants found therein include *Eleusine indica* and *Axnopus compressus.*

Land preparation

The study began by demarcation of an experimental plot of the size 2m by 3m in the botanical garden. The plot was cleared out before experimental bowls were purchased and put on the plot.

Soil collection

Two types of soil were earmarked for this experiment; namely clay soil (C) and humus soil (H). The clay soil used for this project was collected from the bank of the Ikpoba hill river, Benin city after which it was taken to the Department of Soil Science in the Faculty of Agriculture where it was confirmed to be clay soil. The humus soil used was obtained from a garden, at an area surrounding a banana tree. Care was taken to ensure that in the collection of the two soils, only top soil (0-10cm deep) was taken.

Preparation of soil samples

After collection, the humus and clay soils were air dried to remove moisture. Seven samples of the soils were then prepared by mixing the clay and garden soils in different ratios. The different ratios used were: Sample A: 100% clay soil, 0% humus soil, Sample B: 90% clay soil, 10% humus soil, Sample C: 75% clay soil, 25% humus soil, Sample D: 50% clay soil, 50% humus soil, Sample E: 25% clay soil, 75% humus soil, Sample F: 10% clay soil, 90% humus soil, Sample G: 0% clay soil, 100% humus soil. 5 kg each of these soils was filled in different bowls and made in two replicates, amounting to fourteen bowls of soil ratios.

Collection of *Eleusine indica*

Young tillers of *Eleusine indica* were collected from the area surrounding the life sciences complex in the Faculty of Life Sciences, University of Benin, Benin, Nigeria. The tillers were about 7-8cm in height having about five leaves each. The tillers were then taken to a nursery prepared earlier in the botanical garden for acclimatization.

Pollution of the soil with heavy metal (Pb)

100mg/kg of lead was required to pollute the soil. However, in order to achieve this, it was important to determine the exact weight of lead chloride that would be needed for the experiment. Given that molecular weight of lead chloride is 278.1g (i.e 207.1+35.5+35.5). It means that the weight of lead in lead chloride was 74%. Thus, 74% of the new weight of $PbCl_2$ (w) must be equal to the required 100mg/kg of lead of the study. 0.74w = 100mg/kg. Therefore w=100/0.74. w=134.05mg/kg of $PbCl_2$. Since 5kg of soil was required for each bowl. It meant that each bowl would be polluted with 5x135.05=675.25mg. Therefore 675.25mg/kg of lead was used to pollute each bowl.

MORPHOMETRIC PARAMETERS

Plant height (cm): The height of the plant was measured using a metre rule, once a week for seven weeks. **Leaf length**: The leaf length was measured using a metre rule and a tape rule. Readings were taken once a week for the duration of the experiment. **Number of leaves**: This was determined by counting. **Leaf width (cm)**: Leaf width was measured using a metre rule for the experimental duration. **Leaf area**: The leaf area was calculated using Area= Leaf length x leaf width x 0.9 following Shabani and Sepaskhah (2017). **Chlorophyll content**: Chlorophyll content analysis was estimated as an index by using a chlorophyll content meter (CCM 200 plus, Apple G. instrument). The average meter reading of 5 leaves per plant was taken as the CCI. **Foliar colour**: The foliar colour of the plants was determined using a colour code.

SOIL PARAMETERS

Exchangeable acidity was determined by titration method of Mclean as described by Udo et al. (2009). The electric conductivity was determined using conductivity meter following Udo et al. (2009). The Cation Exchange Capacity (CEC) was determined using boric acid *indica*tor mixture depending on the NH_4^+ content following Morteza et al., (2017).

Data Analysis

The data collected were subjected to statistical analysis using SPSS software and significant mean were separated using LSD at 5% probability level.

RESULTS AND DISCUSSION

Results for soil parameters before pollution with Pb and after remediation with *Eleusine indica* were presented in Table 1. It was observed that exchangeable acidity and CEC of humus soil was significantly higher than that of clayey soil ($P<0.05$). It was also observed that the exchangeable acidity and CEC increased significantly with increasing proportions of humus soil before pollution with Pb; however, electric conductivity decreases. This may be due to addition of the humus soil as it is considered to have the highest CEC (Moore et al., 2008; Singh, 2002). This is consistent with the work of Onwuka et al., (2016), who suggested that CEC of soil can be increased by addition of soil organic matter. Humus soils are rich in organic matter which explains the high CEC in them at increasing H.

Similar results were observed after remediation with *E. indica* except for electric conductivity and exchangeable acidity (Table 1). There was significant increase in the CEC of the soil after remediation at increasing ratio of H and a significant reduction in exchangeable acidity at increasing H. This may be due to remediation activities and natural attenuation at increasing H. This could be that organic matter reduced exchangeable acidity of soils by forming strong bonds known as chelates with aluminium and this resulted in a decrease in aluminium solubility and soil exchangeable acidity. George (2009) suggested that soils with high exchangeable acidity have low soil acidity, while increasing CEC is an indication of low heavy metals.

Furthermore, the result showed significant difference between the CEC of the mixed soil sample with the highest proportion of humus (10C:90H) and the mixed soil sample with the lowest proportion of humus (90C:10H) before and after remediation. This portrays CEC as important indicator in remediation of Pb by *E. indica*. There was however no considerable difference between the electric conductivity of pure clayey soil (C) and pure humus (H). Soil salinity is one of the major factors that determine electric conductivity (Corwin and Yemoto, 2017).

Table 1: Soil parameters before and after remediation

Soil mix	Exchangeable acidity (ppm)	Electric conductivity (µs/cm)	Cation exchange capacity (meq/L)	Exchangeable acidity (ppm)	Electric conductivity (µs/cm)	Cation exchange capacity (meq/L)
	Before transplanting			**7 weeks later**		
CS	301b	114.2b	0.210b	200a	145.1b	0.396b
HS	490a	107.4b	0.710a	100b	150.1b	0.898a
90C10H	402ab	173.9b	0.552a	184a	230.4a	0.572ab
75C25H	451a	147.9b	0.571a	169a	225.2a	0.568ab
50C50H	426a	144.5b	0.611a	186a	115.2b	0.579ab
25C75H	433a	127.6b	0.622a	143b	183.4ab	0.750a
10C90H	319b	233.2a	0.685a	132b	119.4ab	0.815a
	0.667	0.53	0.474	0.016	0.024	0.113

Mean on the same column with similar alphabetic superscripts do not differ from each other ($p>0.05$)
Soil mix are CS clayey soil, HS humus soil, 90C10H 90 parts of clay in 10 parts of humus, other mix are 75C25H, 50C50H, 25C75H, and 10C90H, C being clayey and H being humus.

Plant Pb accumulations and soil residual concentration were determined in Table 2. Result showed that bioaccumulation of Pb was higher in the roots than shoots of the test plant. Studies have shown that plants take up metals through the root and translocate it to other parts (Bieby et al., 2011). According to Galadima and Garba, (2012) *Eleusine* plant was observed to accumulate most of the elements of interest in the root. Gothberg et al., (2004) suggested that one of the mechanisms by which uptake of metal occurs in the roots may include binding of the positively charged toxic metal ions to negative charges in the cell wall. The result also showed significant reduction in rhizoaccumulation of Pb in the clay soil compared to humus soil ($P<0.05$). The humus soil has the highest rhizoaccumulation concentration of Pb (29.5 mg/kg), while clayey soil has the lowest (12.5 mg/kg).

Increase in rhizoaccumulation of Pb was also observed at increasing addition of H except for (25C:75H ratio). This may be due to the increased CEC of H soil compared to the C soil with addition of H. Plants from soils with higher CEC showed higher Pb accumulation. Significant differences between the Pb detected in the root and stem of *E. indica* was also obtained. Pb appeared to be more concentrated at the root than the stem for all the soils except the clayey soil. Soil residual concentration was lower in the plant with the highest Pb accumulation and reduces with reduced Pb accumulation. This indicated that increasing proportion of Pb was removed from the soil with increasing CEC, which could be traced to phytoextraction potential of *E. indica* and the soil condition. This is consistent with the work of (Wilson et al., 2014; Wang et al., 2013; Ahmad and Goni, 2010) that soil parameters such as CEC strongly influence heavy metal accumulation by plants. USEPA (2000) reported that heavy metals when mopped up by plants have reduced presence in the soil.

Table 2: Plant Pb accumulation and soil residual concentration

	Heavy metal accumulation			Soil residual concentration (mg/kg)
	Shoot (mg/kg)	Root (mg/kg)	p-value	
CS	19.5	12.5	0.142	45.1±4.6
HS	19.5	29.5	0.238	23.4±9.5
90C10H	16.6	19.5	0.331	38.2±7.3
75C25H	15.6	23.4	0.049	33.4±9.5
50C50H	11.7	15.6	0.031	30.2±8.3
25C75H	9.82	29.5	0.029	37.8±9.1
10C90H	23.4	15.2	0.214	25.4±9.53
	0.025	0.046	-	0.388

Mean on the same column with similar alphabetic superscripts do not differ from each other (p>0.05)
Soil mix are CS clayey soil, HS humus soil, 90C10H 90 parts of clay in 10 parts of humus, other mix are 75C25H, 50C50H, 25C75H, and 10C90H, C being clayey and H being humus.

Figure 1 and 2 presents the morphometric parameters such as plant height and leaf length observed in the *E. indica* after the seven weeks experiment. It was observed that the plants growing in clayey soil had the least plant height and leaf length (9 and 6 cm), while the plants growing in the humus soil had the highest plant height (39 and 33 cm). Increasing plant height and leaf length was also observed at increasing addition of H except for the 25C:75H mixture. Recall that H soils showed higher CEC and plants from such soil showed more rhizoaccumulation of Pb.

The increasing height of Plants with higher H may be that the test plant accumulate Pb for its growth or tolerant to Pb pollution. According to Bouaziz et al. (2010) Pb is plays essential role in plant growth; however, at higher concentration, it becomes toxic and affect plant growth.

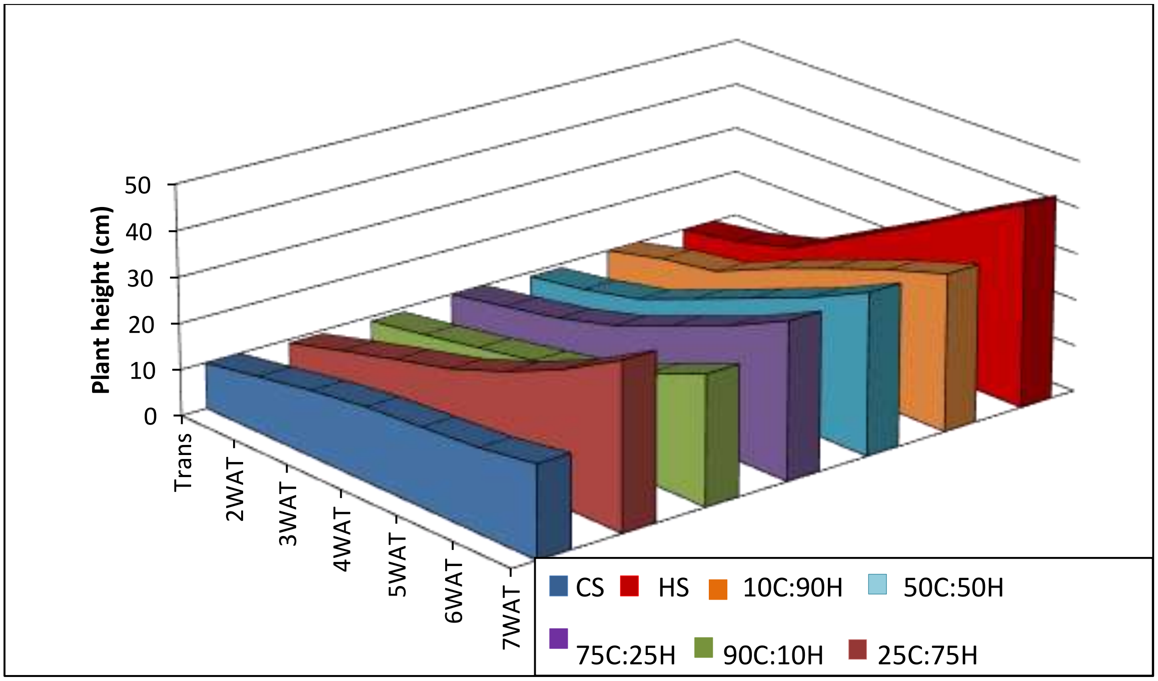

Fig. 1: Plant height of *Eleusine indica* in a humus-clay soil mix after soil pollution with Pb

HS humus soil, CS clayey soil. 90C10H means 90 parts of clay in 10 parts of humus soils respectively. Same applies for other soil mixes.

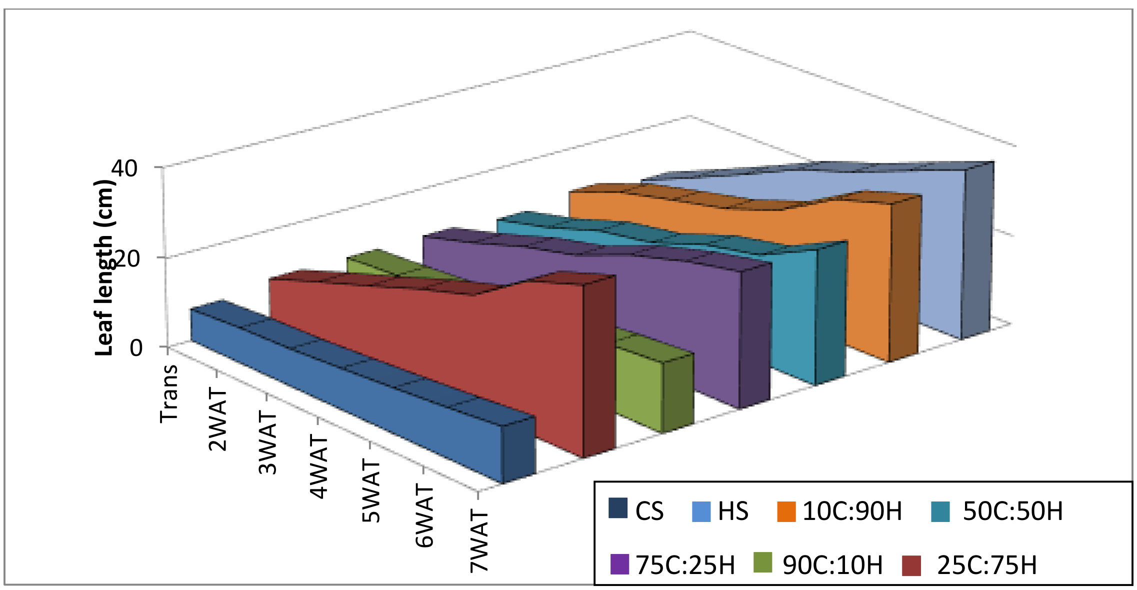

Fig. 2: Progression in leaf length of *Eleusine indica* in a humus-clay soil mix after soil pollution with Pb.

Other variations in plant morphometric parameters were recorded in table 3. Plants from humus soil showed highest number of leaves, leaf length, leaf width, leaf area, chlorophyll content index and prominent foliar colour; however, plants grown from clayey soil significantly showed decrease in these parameters. This is likely as a result of low CEC of the clayey soil and low rhizoaccumulation of Pb in the test plant.

Table 3: Morphometric parameters of *Eleusine indica* in a Pb-polluted study soils after 7 weeks of remediation.

Soil mix	*No. of leaves	Leaf length (cm)	Leaf width(cm)	Leaf area (cm2)	Chlorophyll content index (cci)	prominent foliar Colour
CS	$5^{d}\pm2$	$11.2^{b}\pm2.1$	0.3 ± 0.1	$4.9^{c}\pm1.3$	$1.2^{c}\pm0.2$	sea green
HS	$38^{a}\pm7$	$33.1^{a}\pm5.3$	0.4 ± 0.1	$11.8^{b}\pm3.6$	$2.3^{b}\pm1.4$	Yellow green
90C10H	8d±2	$14.4^{b}\pm3.2$	0.3 ± 0.2	$3.8^{c}\pm2.1$	$2.6^{b}\pm1.8$	Olive drab
75C25H	12d±3	$17.5^{b}\pm6.2$	0.4 ± 0.2	$9.9^{b}\pm2.9$	$3.5^{ab}\pm1.6$	Dark olive green
50C50H	$15^{c}\pm2$	24.9 ± 6.5	0.5 ± 0.3	$10.8^{b}\pm3.1$	$4.9^{a}\pm1.8$	Dark olive green
25C75H	$18^{c}\pm4$	$31.5^{a}\pm3.1$	0.6 ± 0.3	$18.01^{a}\pm2.7$	$5.5^{a}\pm1.6$	Dark olive green
10C90H	$25^{b}\pm5$	$33.2^{a}\pm7.2$	0.6 ± 0.2	$18.93^{a}\pm1.8$	$4.3^{a}\pm0.3$	Dark olive green
	0.003	0.039	0.481	0.009	<0.001	NA

**Mean rounded off to the nearest integer*

Mean on the same column with similar alphabetic superscripts do not differ from each other (p>0.05)

Soil mix are CS clayey soil, HS humus soil, 90C10H 90 parts of clay in 10 parts of humus, other mix are 75C25H, 50C50H, 25C75H, and 10C90H, C being clayey and H being humus

Plate 1: Test plant after harvest at 7 weeks after plant exposure.

CONCLUSION

From the present study, it can be concluded that the CEC of soil significantly influenced accumulation of lead in *Eleusine indica*. Increasing humus content of soil leads to increasing CEC values. Plants grown in soil mixed with the least CEC values showed least amounts of lead in them compared to the plants in the soils with higher CEC values, which showed higher amounts of lead in them. The observed increase in morphometric parameters at increasing Pb accumulation shows the tolerant nature of *E. indica* to Pb. The ability to alter soil properties such as CEC improve phytoremediation strategies. However the electric conductivity of the soils in this research did not follow a flowing trend. This could be due to the limited time for the study. It is recommended that in further studies, soil samples should be left for a longer period of time to track changes in other soil properties.

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