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Towards a clean environment: To what extent can trees serve as phytoremediators for chemically polluted soils?

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ABSTRACT

The study investigated the potential of five common tree species in West Africa to bioaccumulate heavy metals in their various plant components over the course of different growth phases and at varying contamination levels (control, double the permissible level and triple the permissible limit). Heavy metals in plant tissues were extracted and analyzed using standard wet chemistry procedures. The results showed metal concentrations in tree tissues to positively and significantly correlate with contamination level, growth stage, and plant component. Metal mobility and availability in the tree tissues were in descending order of Mn >Zn>Cu=Pb>Cd. We found heavy metals to be principally accumulated in the stem tissues following a metal exclusion theory, independent of tree species. Tree roots demonstrated a significant accumulation of heavy metals second only to the stem. The results revealed that heavy metal mobility and bioavailability in plant tissue was tree-specific, growth stage-dependent, and metal-specific; showing that trees have a preferential affinity to accumulate a specific metal or pollutant. The study established *Shorea roxborghi* to be a mono-accumulator of manganese while *Tectona grandis* and *Terminalia ivorensis* were multi-accumulators of metals such as Mn, Cu, Zn and Pb. The interactive effects of contamination levels and tree species showed *Tectona grandis* and *Terminalia ivorensis* grown in chemically polluted soils at triple the permissible level to have the highest heavy metal uptake with the highest phytoextraction potential. We conclude that all the tree species considered in this study could potentially be effective phytoremediators of chemically polluted soils through their ability in phytoextraction of metals such as lead, zinc, copper, and manganese as well as through phytostabilization of cadmium in contaminated soils.

Introduction

Environmental pollution is a critical global concern in our world today in which the health of millions of people, plants, and animals are endangered due to industrial and atmospheric pollutants (Martinez et al., 2001; Azeez et al., 2014; Ajiboye et al., 2019; Rinklebe et al., 2019). There are of course different forms of environmental pollution such as soil, water, and air pollution; and of these soil pollution presents a major challenge owing to the dual role of soils in ecosystem sustainability and food security (Mesele et al., 2023). In recent times, attention has been drawn to issues pertaining to the degradation of the environment by a wide range of chemical pollutants some of which include heavy metals (El-Demerdash and Elagamy, 1999; Ok et al., 2020; Li et al., 2022). Heavy metal pollutants are introduced into the environment through natural and anthropogenic sources (Cui et al., 2020; Proshad et al., 2018; Azeez et al., 2014; Kabata-Pendias and Pendias, 1993). They bioaccumulate in agricultural products and thus contaminate food sources when they are present in the environment beyond the permissible levels (Lin et al., 2004).

Studies indicated that human activities could cause a large increase in the concentration and species of heavy metals in the soil (Frost and Ketchum, 2000; Mangwayana, 1995). Urban soils in the metropolitan centers of Southwestern Nigeria are probably potentially toxic due to heavy metal deposits from diverse sources (Famuyiwa *et al.*, 2018). In developed countries where agricultural activities and industrial land utilization are regulated, heavy metal contamination of agricultural soils still occurs through fertilizer application, irrigation with wastewater or sewage sludge (Devkota and Schmidt, 2000; Frost and Ketchum, 2000; Mangwayana, 1995; Mahmood and Malik, 2014). In Nigeria, the most important sources of such contamination of urban soils are derived from

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industrial processing plants or leachates from solid waste dumpsites or charcoal production (Famuyiwa *et al.*, 2018; Ajiboye et al., 2019). Such effluents and wastes hardly decompose completely but leave undegradable contaminants either in the soil, air or water (Famuyiwa *et al.*, 2018; Ogunyemi et al., 2003). It is supposed that in many cases poisoning of crops occurs when the plants take up the heavy metals present in the soil through the roots and are later accumulated in the shoot and other edible parts of the plants (Nieboer and Yassi, 1998).

There are other external factors apart from those from anthropogenic sources that affect the concentration of heavy metal in plants and within the plants (Muchuweti et al., 2006; Azeez et al., 2014) like atmospheric deposition, the degree of plant's maturity at harvesting, nature of the soil on which the plant is raised and the climatic conditions (Solymosi and Bertrand, 2012; Sahito et al., 2016). Soil is considered a key factor that determines the level of heavy metals in crop plants (Itanna, 2002; Madyiwa et al., 2002) probably because this is the binding and retention site for the pollutants. Heavy metal accumulation in agricultural soils could result in long-term metal accumulation in the food chain thereby having negative effects on human and animal health (Chumbley, 1982 Fergusson, 1990; Suave et al., 1996) and could strongly influence species diversity (Lester, 1987).

The human or animal consumption of crops grown on such polluted soils may pose serious health risks. Toxic metals like lead have mutagenic tendencies and could induce some carcinogenic effects in both humans and animals (Baudouin et al., 2002). Lead poisoning is widely known to cause neurological defects in children and adverse clinical effects on sperm morphology and counts as well as in pregnancy and female fertility (Vigeh et al., 2011). Arsenic has deleterious effects such as cardiovascular problems, skin cancer and kidney damage on the human body. Jaishankar et al. (2014) noted lead and cadmium to be the most toxic, the most abundant metals often found in the food chain.

Today, chemically polluted soils are becoming widespread and creating major environmental concerns globally. The need to have a clean and pollution-free environment therefore has never been more urgent and crucial. A number of methods have been deployed to clean metal-polluted soils. The conventional remediation approaches often involve soil incineration, in situ vitrification, landfill and excavation (excavation and disposal of contaminated soils into landfills -not eco-friendly and could become a secondary source of pollution), soil washing, soil flushing, solidification and/ or stabilization of electrokinetic systems (Sheoran et al., 2011; Wuana and Okieimen, 2011). These approaches are grouped into physical and chemical approaches and have some challenges such as high cost, intensive labour, disturbance of natural soil microflora and irreversible change in some soil properties. The chemical approaches in addition to the aforementioned challenges, can cause secondary contamination threat, making the effects therefore more devastating (Danh et al. 2009; Sarma 2011).

The use of trees in phytoremediation is emerging and gradually gaining attention. Maric et al. (2013) highlighted the criteria needed for a plant to be suitable for use in phytoremediation to include its ability to be perennial and having large amounts of biomass which are also capable of storing considerable concentrations of metal pollutants in their tissues. Many available hyperaccumulators which are mostly herbaceous (Baker et al. 2000) could not meet these criteria set by Maric et al. (2013). As an alternative therefore fast-growing vascular plants have been proposed as possible candidates for phytoextraction technology (Puschenreiter et al. 2010). Trees differ from other lifeforms in that they are perennial, long-lived, and have a larger surface area. Large canopy areas and extensive root and shoot systems in trees may place them among the top viable xenobiotic accumulators (Mench et al., 2010). There is however limited scientific information on the heavy metal accumulation potential of tree species relative to other life forms. This is often due to the cost and the long period of experimentation required. Particularly in West Africa, it is unknown how well several popular tree species can treat soils that have been contaminated by heavy metals. In light of this, the objectives of our study were developed to assess the potential for metal accumulation in five widespread tree species in West Africa and to measure the degree to which these species are selective in their accumulation of heavy metals.

Materials and methods

Study area

The study was carried out in the Forestry Research Institute of Nigeria's (FRIN) Screenhouse, Central Nursery, and Department of Sustainable Forest Management. FRIN is located between latitudes 03° 51′20′' and 07° 23′43′' east and north, respectively. The research site's climate is typical of a West African monsoon, with a bimodal rainfall distribution and wet and dry seasons. The average annual rainfall during the wet rainy season, which lasts from April through October, is 1548.9 mm, falling over roughly 90 days. The relative humidity is 71.8%, with the average maximum temperature at 31.11°C and minimum temperature at 22.76°C (FRIN, 2018).

Source of the chemical pollutants and tree seedlings

At the Bioscience Center in FRIN, chemical pollutants with varying concentrations of lead (Pb), zinc (Zn), copper (Cu), cadmium (Cd), and manganese (Mn) were obtained for the incubation studies. In aqueous solutions, salts that are water soluble were used to introduce the metals to the soil. Cadmium was added as Cd $(NO_3)_2.4H_2O$, Pb as Pb $(CH_2COO)_2.2H_2O$, Cu as $CuSO_4.5H_2O$, Zn as $Zn(NO_3)_2.6H_2O$ and Mn as MnO_4.

The Central Nursery of the Department of Sustainable Forest Management at FRIN provided three-month-old seedlings of the five tree species *Tectona grandis (TK), Gmelina arborea (GM), Shorea roxburghii (SH), Terminalia ivorensis (TV),* and *Terminalia superba (TS),* all of which are roughly the same height (30 cm).

Soil sampling and physicochemical soil analysis of the study area

Topsoil samples (0-20 cm) were used for this study and they were collected from the uncultivated site of the Arboretum at FRIN. The soil sample was air-dried at room temperature and sieved with a 2 mm sieve in preparation for soil analysis. All were done following standard procedures. The soil particle size analysis was conducted using the hydrometer method (Bouyoucous, 1951). Soil pH was measured with an integrated digital pH meter (Hanna Instruments) in 1:2 soil /water following the procedures of Thomas (1996). Soil organic carbon was determined by the wet oxidation-reduction method (Walkley and Black, 1934). The soil total nitrogen was measured based on the Macro-Kjeldahl method in accordance with Bremner (1996) procedures. Exchangeable cations such as exchangeable Ca, Mg, K and Na in the soil samples were extracted in a neutral normal ammonium acetate solution using standard protocol (Helmke and Sparks, 1996). Potassium and sodium ions were determined by a Flame Photometer. The exchangeable calcium and magnesium by Atomic Absorption Spectrophotometer (AAS).

Experimental design and treatments

Ten kilograms of soil were weighed into 360 polyvinyl pots with each bag measuring 12 × 18 cm. The soils were contaminated with lead, copper, zinc, cadmium, and manganese at levels 2 and 3 of contamination (twice and triple permitted limits), respectively. The mixture was allowed to equilibrate for 7 days. After this incubation period, 12 weeks seedlings of *Gmelina arborea, Tectona grandis, Terminalia superb, Shorea roxborghi and Terminalia ivorensis* were transplanted into the pots and irrigated at field capacity (Plate 1). The experimental design was a 3 × 5 factorial experiment arranged in a Completely Randomized Design with three replications. There were 8 pots per replicate giving a total of 360 experimental units. The experimental factors were 3 levels of contamination (control, double the permissible level, and triple the permissible limit) and 5 tree species. The levels of contamination were:

Level 1= Conta 1 = Control (clean soil with no chemical pollutants)
Level 2 = Conta 2 = Cd = 0.006 g/kg, Cu = 0.2 g/kg, Pb = 0.4 g/kg, Zn = 0.6 g/kg, Mn = 6 g/kg.
Level 3= Conta 3 = Cd = 0.009 g/kg, Cu = 0.3g/kg, Pb = 0.6 g/kg, Zn = 0.9 g/kg, Mn = 9 g/kg. This rate is prepared in accordance with Denneman and Robberse (1990) and WHO (1996). The trees were subsequently allow to grow under the prevailing environmental and climatic conditions.

Heavy metal analyses

At every 8-week interval after transplanting, forty-five (45) plants were selected for destructive sampling. The plants were separated into leaves, stems, and roots. A computerized sensitive scale was used to measure the fresh weight of each plant component. For 48 hours, the plant samples were dried in an oven at 65°C. The dried samples were mechanically ground on an electrically driven grinding machine after the oven-dried weight was measured. The ground materials were digested at a rate of 3:1 in an acid solution of nitric acid and perchloric acid. Whatman filter paper No. 40 was used to filter the solution. The filtrate was analyzed for cadmium, lead, copper, zinc, and manganese using an Atomic Absorption Spectrophotometer (AAS).

Statistical analysis

The data were analyzed using Rstudio. Analysis of Variance (ANOVA) was performed first to check and correct the data for normality and heteroscedasticity. The Scientific Graphing Functions for Factorial Designs (Sciplot) package in Rstudio was used to generate some of the interaction graphs. Clustered column charts were used to present most of the relationship between the contamination levels and tree species in the various plant parts across the different growth stages. Means were separated using LSD at 5% probability level. Total metal accumulation/uptake was computed by multiplying the overdried weights of the roots, stems, and leaves with their metal concentrations (Fig. 1).

Results

Soil physical and chemical properties at planting

As presented in Table 1, the soil is loamy sand and is slightly acidic. The soil is relatively rich in organic carbon and nitrogen. Calcium ions dominated the exchange site while magnesium and potassium are low in concentration. The concentrations of the soil micronutrients and heavy metals are within the acceptable thresholds.

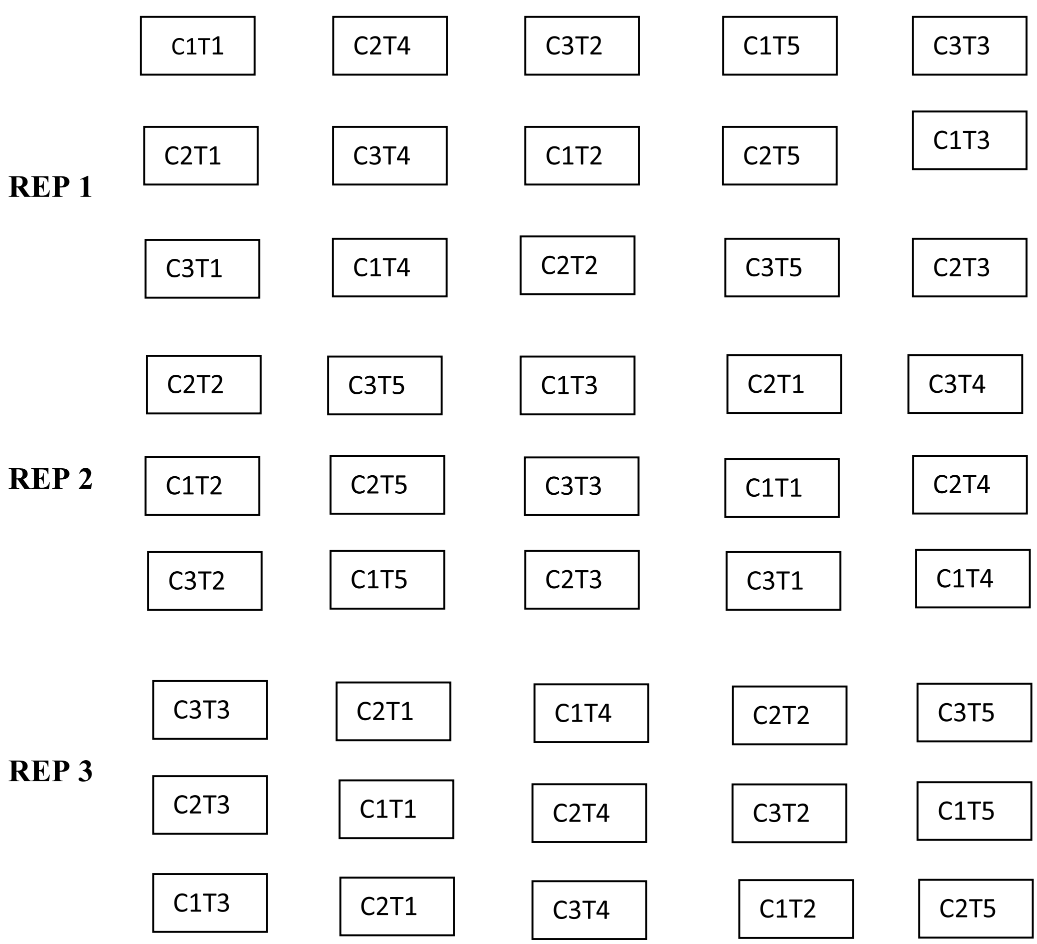

Fig. 1. Experimental layout of the screenhouse study.
C1T1 = Control T. grandis, **C2T1** = 2 X Permissible level T. grandis, **C3T1** = 3 X Permissible level T. grandis, **C1T2** = Control G. arborea, **C2T2** = 2X Permissible level G. arborea, **C3T2** = 3 X Permissible level G. arborea, **C1T3** = Control T. Ivorensis, **C2T3** = 2X Permissible level T. Ivorensis, **C3T3** = 3 X Permissible level T. Ivorensis, **C1T4** = Control S. roxborghi, **C2T4** = 2X Permissible level S. roxborghi, **C3T4** =3 X Permissible level S. roxborghi, **C5T5** = Control T. Superba, **C2T5** = 2X Permissible level T. Superba **C3T5** = 3 X Permissible level T. Superba

Table 1
The soil physical and chemical properties of the experimental site.

Properties	Values
Sand (%)	82
Silt (%)	4
Clay (%)	14
Texture	Loamy sand
pH (H_20)	6.38
Organic carbon (g kg^{-1})	19.20
Toal nitrogen (g kg^{-1})	1.70
Available phosphorus (mg kg^{-1})	2.60
Exchangeable Ca (cmol kg^{-1})	4.41
Mg (cmol kg^{-1})	0.44
K (cmol kg^{-1})	0.10
Mn (mg kg^{-1})	23.00
Zn (mg kg^{-1})	82.00
Cu (mg kg^{-1})	3.00
Fe (mg kg^{-1})	159.00
Cd (mg kg^{-1})	0.62
Pb (mg kg^{-1})	75.00

Effect of contamination levels on metals accumulation in the leaves

The effect of contamination levels on metals accumulation the leaves metal concentrations in the leaves at various growth stages were significant and varied depending on the metal species (Fig. 2). Copper concentration in the leaves was significantly high at 12 weeks after transplanting (WAT) under Conta 2 (Fig. 2A). There are no significant differences in the copper concentrations in the leaves at the various growth stages viz-a-vis the other contamination levels.

Foliar Zn concentrations varied significantly as a function of contamination level and this followed a zigzag pattern across the growth stages. Conta 2 has the highest foliar concentration of Zn at 2 and 16 WAT. At 6 and 16 WAT, Conta 3 has the highest foliar Zn accumulation.

Foliar Pb concentration was highest at 6 WAT under Conta 2 (Fig. 2C). Other differences in Pb concentration in the leaves were not significant at $p>0.05$. Foliar accumulation of Cd was significantly high at 16 WAT under Conta 3 (Fig. 2D). Cadmium concentration in the leaves slightly declined under Conta 1 at 16 WAT. There were no significant differences in Mn concentration across growth stages for the different contamination levels, hence the data is not presented here.

Effect of contamination levels on metals accumulation in the stem

Stem accumulation of heavy metals varied significantly under the different contamination levels and at various stages of growth (Fig. 3). Mn accumulation in the stem was significantly high under the control (Conta 1) while there was no significant increase in the Mn concentration in the stem at the various growth stages under the contamination levels (Fig. 3A). At week 14 and 16 WAT, accumulation of copper in the stem was in the order of Conta 1 < Conta 2 < Conta 3 following the contamination level (Fig. 3B). Stem Zn accumulation varied appreciably across the growth stages. The advance growth stage of 16 WAT, Zn accumulation in the stem was comparable in Conta 2 and 3 while the control -Conta 1 had the lowest concentration (Fig. 3C). Stem Pb concentration increased significantly as a function of contamination level from the 8 WAT. There was no significant difference in Pb accumulation under Conta 2 and 3 but these had had higher Pb concentration than those in Conta 1. The concentration of Cd in the stem was generally below the detection limit across the various stages of growth.

Effect of contamination levels on metals accumulation in the roots

Concentrations of Mn, Cu, Zn and Pb in the root followed significant trends under the various contamination levels (Fig. 4). There was a form of systemic fluctuations in the heavy metals concentration across the growth stages. Conta 1 had the highest Mn level in plant roots and the least was observed in Conta 2. Conta 3 had the highest and most significant level of both copper and zinc in the plant root. Other differences were not statistically significant. Interestingly, the variation in Pb concentration in the root across the growth stages and contamination levels followed the same pattern (Fig. 4D). Cadmium could not be detected in the plant roots at various growth stages.

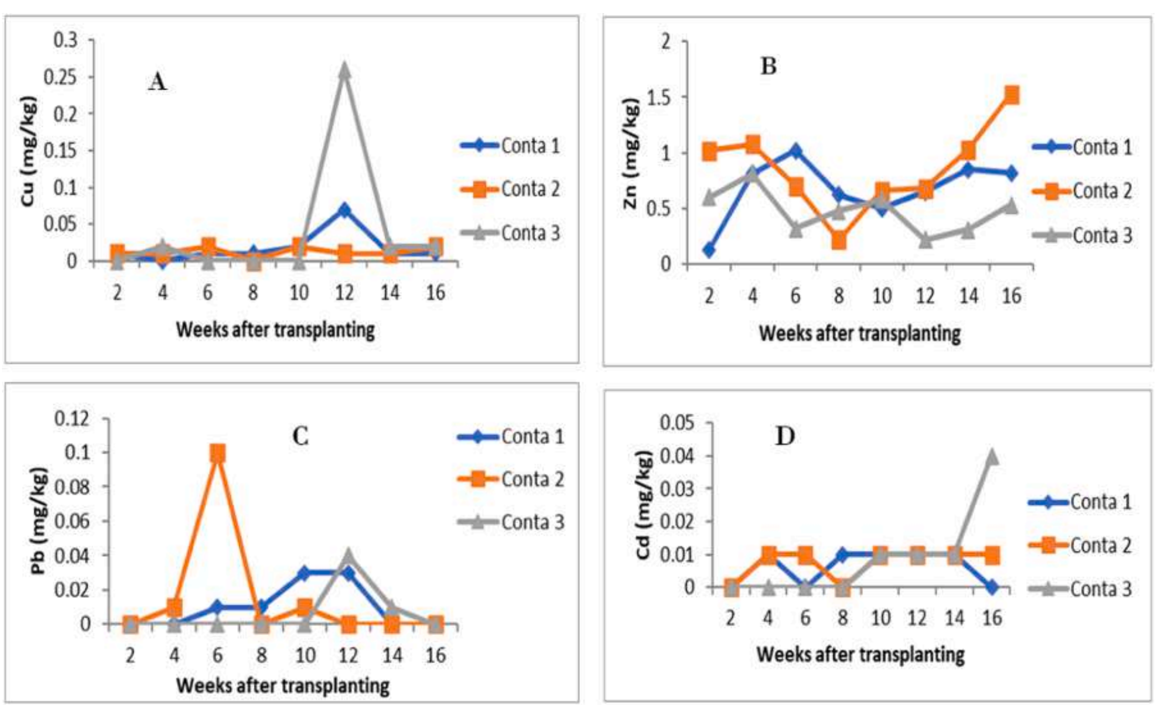

Fig. 2. Foliar concentrations of heavy metals under different contamination levels at various growth stages.
Conta 1= Control, **Conta2** = Cd = 6.0 mg/kg, Cu = 200 mg/kg, Pb = 400 mg/kg, Zn = 600 mg/kg, Mn = 6,000 mg/kg, **Conta 3** = Cd = 9.0 mg/kg, Cu = 300 mg/kg, Pb=600 mg/kg, Zn=900 mg/kg, Mn=9,000 mg/kg

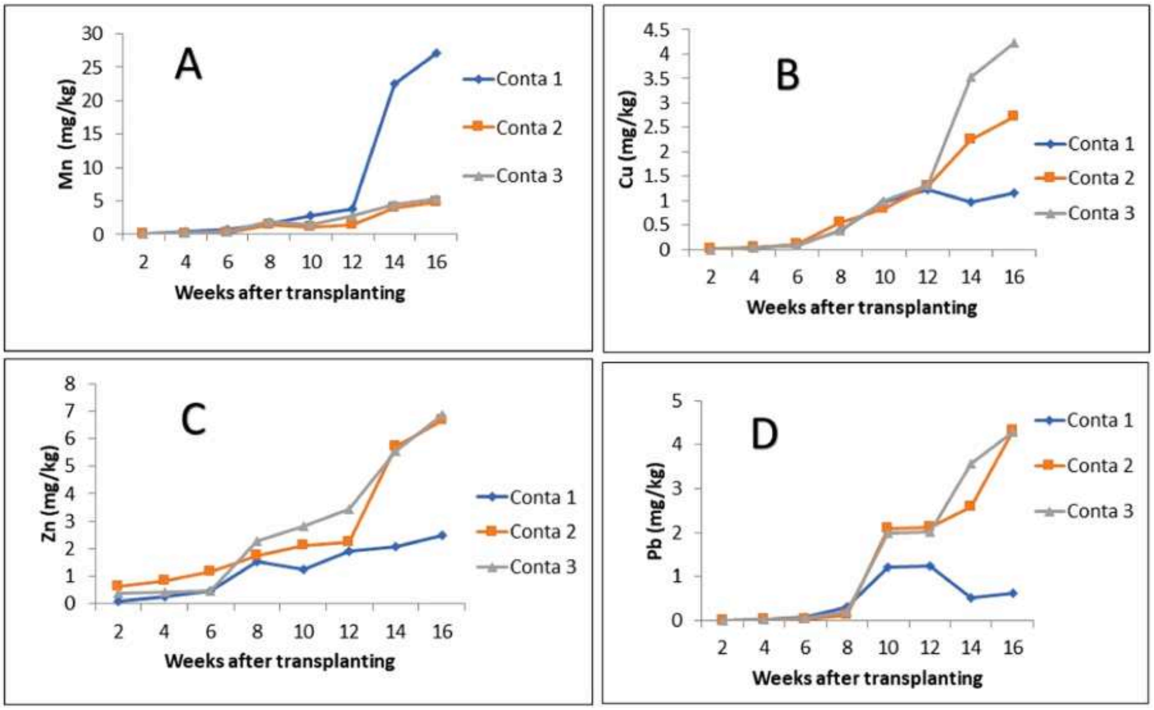

Fig. 3. Stem accumulation of heavy metals under different contamination levels at various growth stages.
Conta 1= Control, **Conta2** = Cd = 6.0 mg/kg, Cu = 200 mg/kg, Pb = 400 mg/kg, Zn = 600 mg/kg, Mn = 6,000 mg/kg, **Conta 3** = Cd = 9.0 mg/kg, Cu = 300 mg/kg, Pb=600 mg/kg, Zn=900 mg/kg, Mn=9,000 mg/kg

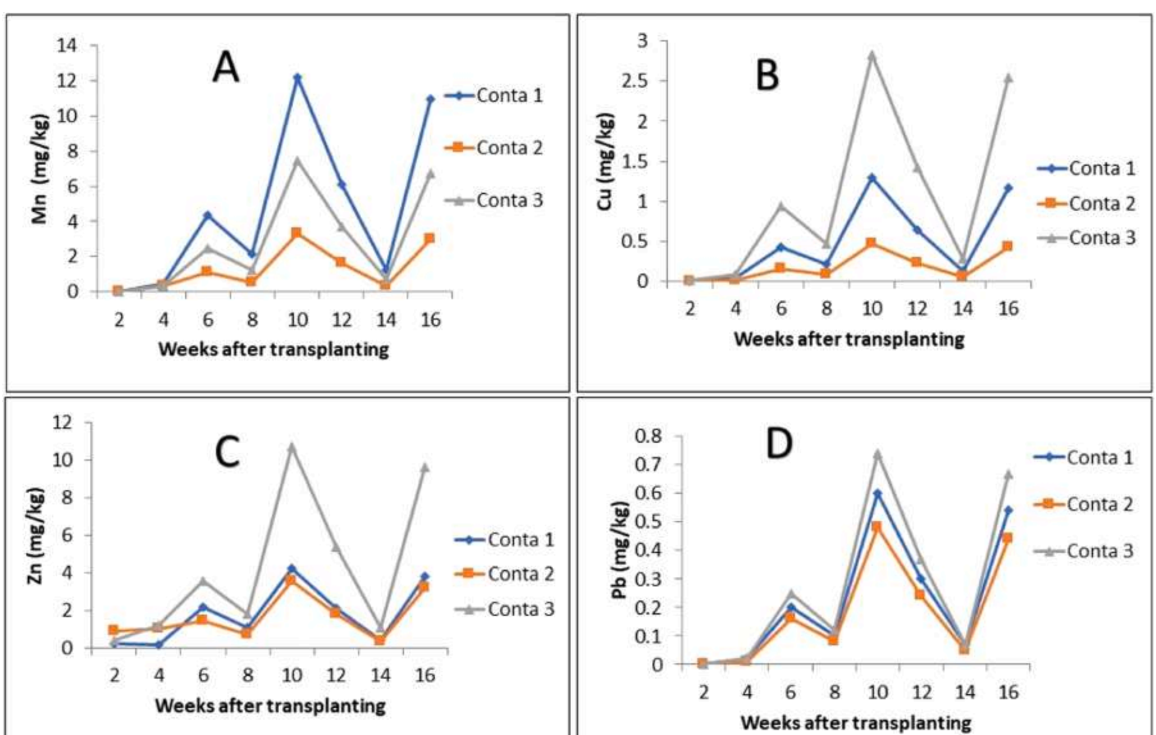

Fig. 4. Root accumulation of heavy metals under different contamination levels at various growth stages.
Conta 1= Control, **Conta2** = Cd = 6.0 mg/kg, Cu = 200 mg/kg, Pb = 400 mg/kg, Zn = 600 mg/kg, Mn = 6,000 mg/kg, **Conta 3** = Cd = 9.0 mg/kg, Cu = 300 mg/kg, Pb=600 mg/kg, Zn=900 mg/kg, Mn=9,000 mg/kg

Effect of tree species on metals accumulation in plant tissue

Foliar concentration of heavy metals

Fig. 5 presents the foliar concentrations of Cu, Zn, Pb and Cd in the various tree species. Concentrations of the metals in the foliar part at the various stages of growth were generally similar and low. TS had a significant and the highest foliar Cu accumulation at 12 WAT (Fig. 5A). SH had the highest foliar Zn concentration at 6 WAT (Fig. 5B). TV had the highest and most significant increase in lead accumulation in the leaves at 6 WAT (Fig. 5C). TS had the highest and highly significant increase in Pb concentration at 16 WAT (Fig. 5D). Other differences were not statistically significant.

Accumulation of heavy metals in the stem as a function of tree species

Metals accumulation in the stem as a function of tree species shows some dynamics across the various growth stages (Fig. 6). SH had significantly the highest Mn in its stem and this is followed by TV at 16 WAT (Fig. 6A). TK and TV gave the highest Cu concentration at 16 WAT while the lowest stem Zn concentration was observed in TS among others (Fig. 6B). There were no significant differences in the stem Zn concentrations from 2 to 12 WAT. At 14 and 16 WAT, however, TV followed by TK had the highest zinc accumulation in the stem (Fig. 6C). Pb accumulation in the stem at the various growth stages was more dramatic than for the other metals. For instance, SH had a sharp increase in Pb concentration at 10 WAT and maintained this concentration till 12 WAT before declining sharply at 14 WAT. TK and TV had a similar and progressive increase in Pb concentration over the growth stages. TS followed a similar trend as SH but at a lower concentration gradient while GM also followed the same pattern with TK and TV but also at a lower concentration gradient (Fig. 6D).

Accumulation of heavy metals in plant roots as a function of tree species

Manganese, copper, and zinc concentrations in plant roots followed a 'rise and fall' order across the various growth stages (Fig. 7). TV followed by TS had significantly the highest root accumulation of Mn, and the least was observed in SH and GM (Fig. 7A). TV followed by GM gave the highest concentration of copper in the root and the least was recorded in TK (Fig. 7B) while TK had the highest root accumulation of Zn (Fig. 7C). There was a somewhat progressive increase in root concentration of Pb with the highest occurring under TK at 16 WAT (Fig. 7D). With the exception of SH with respect to Pb accumulation, other tree species had greater accumulation of metals in their roots at 16 WAT.

Total heavy metal accumulation in plant tissues

The total heavy metal accumulation in the plant tissues varied depending on the contamination levels (Fig. 8). Conta 2 produced plants with the highest Mn concentrations followed by Conta 3 and the least was the control. There are no significant differences in Cu accumulated in plant tissues between Conta 2 and Conta 3 both had her Cu accumulation in plant tissues than Conta 1. Zn accumulation in plant tissues was in the order of Conta 3 > Conta 2 > Conta 1. Lead accumulation followed the same trend as copper. Cadmium was barely present I the plant tissues. In the order of heavy metal increase in plant tissues, Mn >Zn>CU=Pb.

Fig. 9 presents the total metal concentration in plant tissues as a function of tree species. SH had the highest accumulation of Mn followed by TV and the least was GM. TV had the highest accumulation of copper and lead in plant tissues while the highest accumulation of Zn was in TK closely followed by TV. Cadmium accumulation in the plant tissues was significantly low. Fig. 10 gives a general view of metal distribution in the different plant parts of the various tree species. In all, there is a significantly higher metal accumulation in the stem than the other plant parts of the various tree species.

SH and TK had significantly higher metal accumulation in the stem than the other tree species. TV has the lowest metal accumulation in the stem. TS has the highest metal accumulation in the root (Fig. 10).

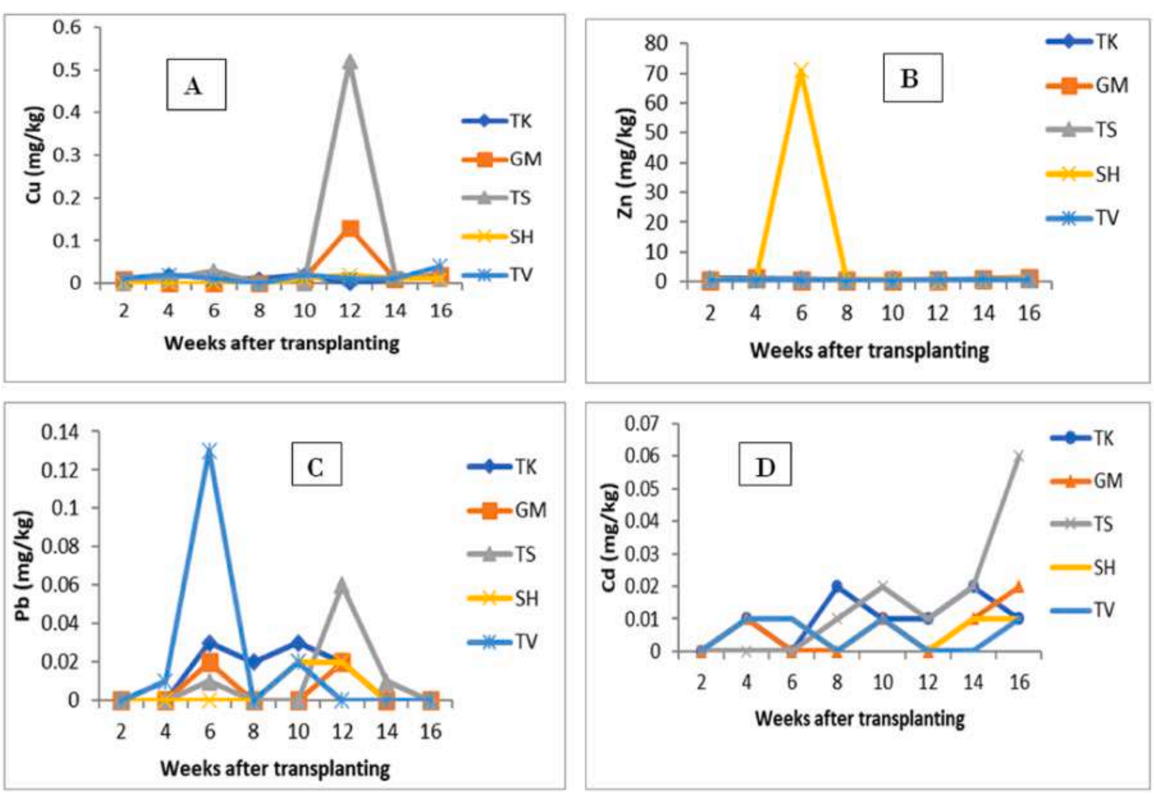

Fig. 5. Foliar accumulation of heavy metals as a function of tree species at various growth stages.
TK =Tectona grandis; GM =Gmelina arborea; TS = Terminalia superba; SH = Shorea.roxborghi; TV = Terminalia ivorensis

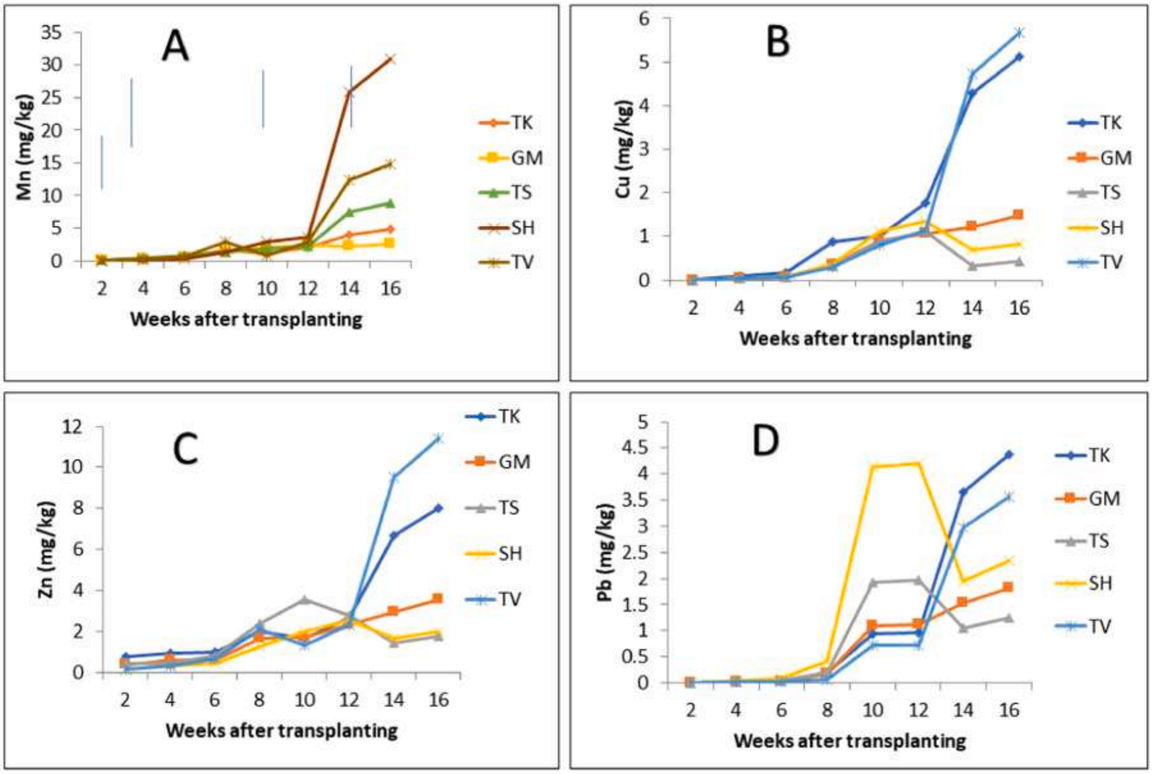

Fig. 6. Stem accumulation of heavy metals as a function of tree species at various growth stages.
TK =Tectona grandis; GM =Gmelina arborea; TS = Terminalia superba; SH = Shorea.roxborghi; TV = Terminalia ivorensis

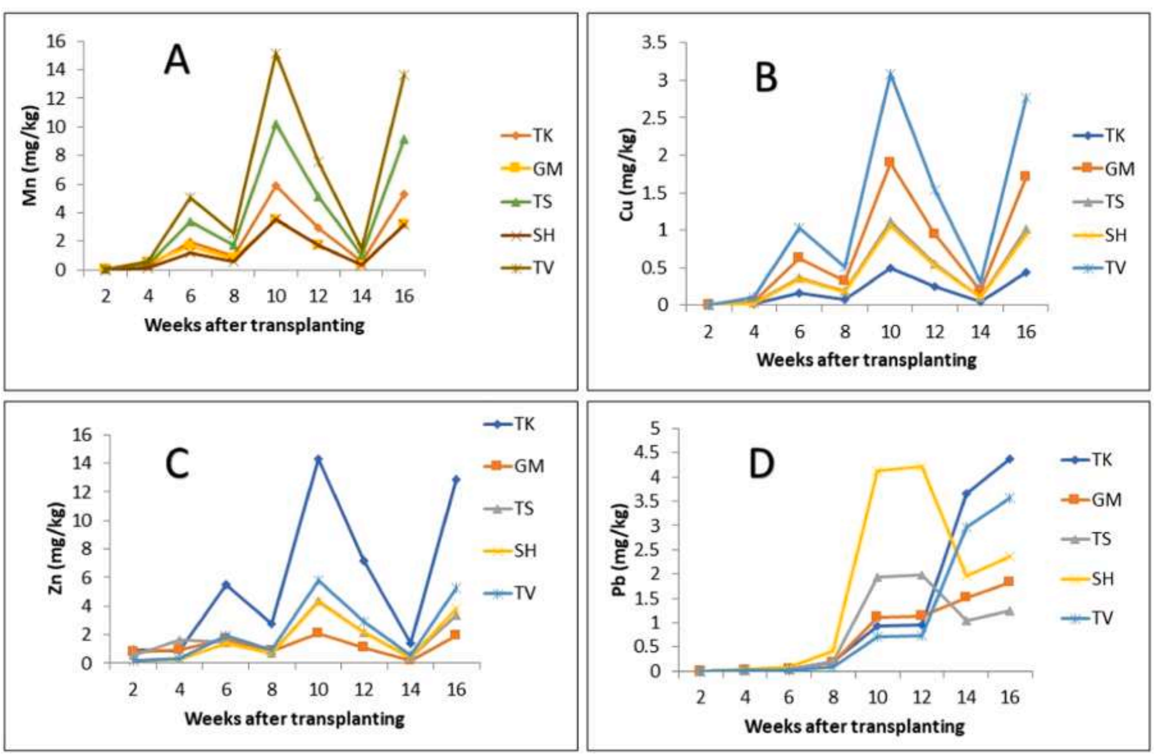

Fig. 7. Root accumulation of heavy metals as a function of tree species at various growth stages.
TK =Tectona grandis; GM =Gmelina arborea; TS = Terminalia superba; SH = Shorea.roxborghi; TV = Terminalia ivorensis

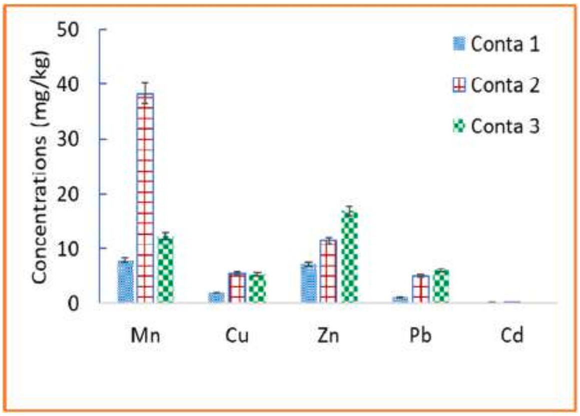

Fig. 8. Total plant tissues accumulation of heavy metals as function of contamination level.
Conta 1= Control, **Conta2** = Cd = 6.0 mg/kg, Cu = 200 mg/kg, Pb = 400 mg/kg, Zn = 600 mg/kg, Mn = 6,000 mg/kg, **Conta 3** = Cd = 9.0 mg/kg, Cu = 300 mg/kg, Pb=600 mg/kg, Zn=900 mg/kg, Mn=9,000 mg/kg

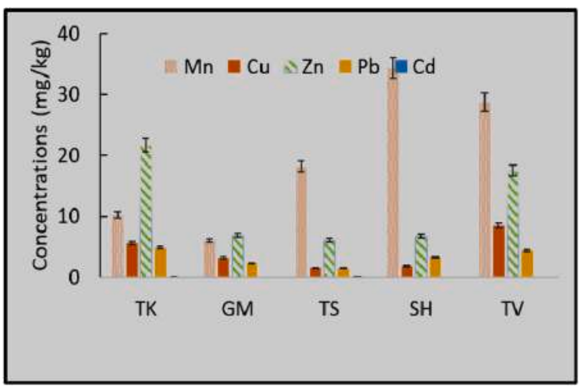

Fig. 9. Total plant tissues accumulation of heavy metals as function of tree species.
TK =Tectona grandis; GM =Gmelina arborea; TS = Terminalia superba; SH = Shorea.roxborghi; TV = Terminalia ivorensis

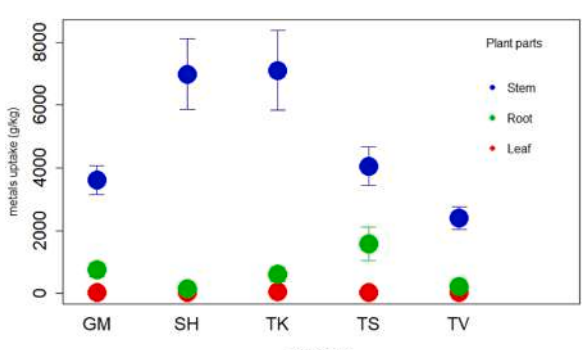

Fig. 10. General trend of metal accumulation in the various plant parts across the different tree species.
TK =Tectona grandis; GM =Gmelina arborea; TS = Terminalia superba; SH = Shorea roxborghi; TV = Terminalia ivorensis

Interactive effects of tree species and contamination levels on metals accumulation

The interactive effects of tree species and contamination levels on total metal accumulation in plant tissues are presented in Table 2. Conta 3 x TS had the highest Mn accumulation in the plant tissue and the control (Conta 1) recorded the least Mn accumulation. For Cu, Conta 2 x TK had the highest concentration followed by Conta 3 x GM. Conta 3 x TK had the highest zinc and Pb accumulation. Cd in plant tissue was highest at Conta 3 x TK.

Discussion

Heavy metal pollution of soils often arises from inappropriate land management practices associated with industrialization and poses health and ecological threats to society (Mesele et al., 2016; Proshad et al., 2018; Cui et al., 2020; Li et al., 2022). The need to ensure a clean and safe environment has never been this urgent. On a global scale, while several approaches have been proposed and implemented, phytoremediation which is an innovative biotechnique for reclaiming land polluted with metals or organic pollutants (Rosselli et al., 2003), remains an important discourse in every effort toward a clean and safe environment. Higher plants such as trees possess bioaccumulative properties and may be used as bio-indicators for the evaluation of heavy metal pollution (Kahle, 1993; Riddell-Black, 1993). In our study, we explored the potential of common and different tree species in West Africa to bioaccumulate heavy metals in their various plant components, including leaves, stems, and roots over the course of their growth phases and at varied contamination levels. By this, we examined the effect of contamination levels on metal accumulation in the different plant components, tree species' effect on metal buildup in plant tissues, and the interactive effects of contamination levels and tree species on the overall metal accumulation in plant biomass.

The findings indicated that depending on the degree of contamination and the growth stages, the metal content in tree tissues may vary greatly. This supports earlier research by Lepp and Eardley (1978), who examined how metal-contaminated sludge affected the growth of sycamore seedlings. They discovered a direct correlation between the amount of contamination and the metals that had accumulated in the plant tissues. Across the plant growth phases, there was a general trend toward larger foliar accumulation of copper, zinc, lead, and cadmium at a higher contamination level, while there were some variances that did not follow a particular order. These findings indicate that the study's trees have the potential to be metal hyperaccumulators. Riddell-Black (1993) asserted that trees often exhibit tolerance mechanisms that allow them to survive higher concentrations of heavy metals than food or herbaceous crops without showing any toxicity symptoms. In contrast, Kahle (1993) claimed that few tree species are metal-tolerant because they are unable to adapt to high soil concentrations of heavy metals.

Stems are the primary conduits by which water and nutrients are channeled from the roots to the leaves in addition to serving as a substantial reservoir of water, nutrients, and pollutants. Stem accumulation of heavy metals varied significantly under the different contamination levels and at various stages of growth. While the concentration of cadmium was generally below the detection limit in the stem tissue, there was appreciable levels of copper, zinc, and lead in the stem and this was more so under higher contamination level and at the advanced growth stage. This implies a progressive accumulation of metals in stem tissues with increasing plant age under polluted soils. This suggests that tree stems could be a good reservoir of heavy metals in chemically polluted soils and the continuous intake of metals could help clean the soil or significantly reduce the concentration of metals left in the polluted soils on which the trees are being grown. Labrecque et al. (1994) quantified the heavy metals concentration in *Salix discolor* and *S. viminalis* grown on sludge polluted with metals and observed that about 50 – 80% of the quantum of metal accumulated were in the roots and stem–branch biomass. They argued that the observed results implied metal immobilization in the root and stem relative to the leaves and that the metals accumulated in those parts are returned to the soil at the end of the growing season. The return of these metals to the soil at the end of the growing season is a disadvantage in phytoremediation though only true

Plate 1. Cross-section of experimental Set-up / Layout in the Screenhouse.

Table 2
Interactive effects of contamination level and tree species on total metal uptake in plant tissues.

		Tree species Metal accumulation (mg/kg)				
	Contamination levels	GM	SH	TK	TS	TV
Mn	*Conta 1*	37	3	55	6	36
	Conta 2	68	32	325	35	116
	Conta 3	142	312	121	677	160
	Lsd0.05	15				
Cu		GM	SH	TK	TS	TV
	Conta 1	10.3	2.3	13.3	11.3	12
	Conta 2	43	2.3	136.7	52	26
	Conta 3	64.3	30	23	18.3	49
	Lsd0.05	13				
Zn		GM	SH	TK	TS	TV
	Conta 1	96	21	98	71	58
	Conta 2	22	9	241	179	49
	Conta 3	71	55	493	101	57
	Lsd0.05	18				
Pb		GM	SH	TK	TS	TV
	Conta 1	20.7	3	3	8	3
	Conta 2	2.7	1.7	13	5	17
	Conta 3	24	47	83.7	11.7	20
Cd		GM	SH	TK	TS	TV
	Conta 1	4.67	7	9.33	6.33	4.67
	Conta 2	4.33	1.33	4.33	8.33	6.7
	Conta 3	6.67	9.67	11.33	10	8

for the herbaceous lifeform and in this trees have the added advantage of being perennial.

The ability of trees to translocate metals from the root to the shoot varies and depends on certain key edaphic and physiological conditions (Pulford and Watson, 2003). While there could be a general increase in metal concentration in the various tree parts in a chemically polluted environment as already indicated in this study, the species of the trees may however exact some influence on the total metal uptake and the distribution of the metals in the various tree parts. The results showed that the foliar concentrations of heavy metals at the various stages of growth were generally low across the tree species while there were appreciable quantities of the metals in the stem and roots of the trees. Turner and Dickinson (1993) observed that Pb ions were preferentially stored in the stem while Zn was mostly accumulated in the leaves of sycamore trees grown in contaminated soil. McGregor et al. (1996) analyzed the tissues of willow, sycamore, and birch trees which had grown naturally on sites contaminated with industrial wastes and found Pb, Cu, and Cr to be primarily accumulated in the roots while Zn was largely accumulated in the bark of the trees. Several studies documented by Pulford and Watson (2003) confirmed that metals may bio-concentrate in actively growing tissues such as shoots and young leaves of a plant grown on a contaminated site. Similar observations were made by Hasselgren (1999) on sludge-amended willow plots where they found Cu, Pb, and Cr to be mostly in the stems and Zn, Cd, and Ni were predominantly in the leaves.

Perhaps one of the most striking observations in our study was that foliar heavy metal accumulation was tree-specific, growth stage-dependent, and metal-specific as shown in Fig. 5. Thus, *Terminalia superba* had the highest foliar Cu and Pb concentrations, *Shorea roxborghi* had the highest foliar Zn concentration at 6 WAT and *T. ivorensis* had the highest foliar Pb concentration. In a similar study by Nissen and Lepp (1997) copper and zinc concentrations in Salix species were found to differ between the different tree parts and the tree species varied in their metal uptake patterns. Zinc was preferentially concentrated in the shoot tissue relative to Cu concentration or soil concentrations as also evident in our study. Our study and the one by Nissen and Lepp (1997) showed and explained what could be referred to as metal exclusion theory. This is a metal mobility mechanism where mobile metals such as zinc are translocated to the shoot system and metals such as Cu with low mobility are excluded from the translocation system and therefore remain in the root system or soil. This may have accounted for the low zinc concentrations in wood xylem tissues. Pulford and Watson (2003) suggested that metal partitioning between phloem and xylem tissues may change as the soil concentration of the metals changes; implying that at higher contamination levels, there may be a redistribution in metal partitioning among the various plant components.

Heavy metal variations in the stem tissue were also tree species dependent across the various growth stages. Notably, *S. roxborghi* stored a higher concentration of Mn in the stem while *Tectona grandis* and *Terminalia ivorensis* had appreciable Cu^{2+} in their stem tissues. Similarly, *Terminalia ivorensis* followed by *Tectona grandis* had the highest zinc accumulation in the stem. Pb accumulation in the stem at the various growth stages was more dramatic than for the other metals. For instance, *S. roxborghi* had a sharp increase in Pb concentration at 10 WAT and maintained this concentration till 12 WAT before declining sharply at 14 WAT. *Tectona grandis* and *T. ivorensis* had a similar and progressive increase in Pb concentration over the growth stages. These findings corroborated previous research by Shuping et al. (2011) who demonstrated that *the T. grandis* stem can accumulate significant heavy metals via its active transport to all plant organs. *T. superba* followed a similar trend as *S. roxborghi* but at a lower concentration gradient while *Gmelina arborea* also followed the same pattern with *Tectona grandis* and *T. ivorensis* but also at a lower concentration gradient (Fig. 6). The chemistry of these dramatic changes in Pb concentration within the stem of these tree species may require further investigation.

The concentrations of metals such as zinc, copper, and manganese in plant roots exhibited a 'rise and fall' pattern across the various growth stages (Fig. 7). *T. ivorensis* followed by *T. superba* had significantly the highest root accumulation of Mn and the least was observed in *S. roxborghi* and *Gmelina arborea. T. ivorensis* followed by *Gmelina arborea* gave the highest concentration of copper in the root while *Tectona grandis* had the highest root accumulation of Zn and Pb. These results demonstrate that root storage of heavy metals is tree-dependent. This shows that the roots of *T. ivorensis* were more effective in extracting Mn and Cu ions from the soil while the roots of *Tectona grandis* were effective in removing Zn and Pb ions from chemically polluted soils. Our results confirmed and were in partial agreement with Das and Maiti (2007) observations. They observed that metals accumulated typically in root tissues (but not exclusively) and suggested that a metal exclusion mechanism exists (obeying our proposed metal exclusion theory) in trees which is needed for metal tolerance. The accumulation of some metals greater than the toxicity threshold suggests the presence of internal metal detoxification and tolerance mechanisms in tree plants.

Quantifying the overall accumulation of heavy metals in plant tissues as a function of contamination level, tree species, and as a product function of the two parameters was another significant part of this work. Under the contamination threshold, the results showed notable differences in plant tissue metal contents, and it was noted that the proportion of metals in plant tissues rose as the contamination level increased. At higher contamination levels, there were more heavy metals in the tissues and vice versa (Fig. 8). The accumulation of Cd in plant tissue was independent of the contamination levels. In the order of heavy metal increase in plant tissues, Mn >Zn>Cu=Pb>Cd as observed in this study. In line with the suggestions of Baker et al. (1991), phytoextraction potential of any plant is determined by the plant productivity (the activity and quantum of biomass produced) and the level of contamination or contaminant concentrations.

In relation to the effectiveness of different tree species to bioaccumulate heavy metals in their biomass (leaves, stem and roots), we found *S. roxborghi* to be the highest accumulator of Mn followed by *T. ivorensis* and the least was *Gmelina arborea*. Conversely, *T. ivorensis* had the highest copper and lead accumulation while *Tectona grandis* closely followed by *T. ivorensis* had the highest accumulation of Zn; implying that trees have a preferential affinity to accumulate a specific metal or pollutant. The results further suggest two categories of trees in relation to phytoremediation potential. The first categorization is based on the metal concentration in plant tissues, where a tree could either be a low accumulator or hyperaccumulator. In this study, *T. surperba* and *Gmelina arborea* could be regarded as low accumulators while *T. ivorensis* and *Tectona grandis* could be called hyperaccumulators. The second category is based on their metal selectivity, and a tree could be either a specific/mono or non-specific/multi-accumulator. In this regard, *S. roxborghi* has proven to be a mono-accumulator of Mn while *T. ivorensis* and *Tectona grandis* represent multi-accumulators of metals such as Pb, Zn, Cu, and Mn. The effectiveness of these multi-accumulators could be enhanced by alterations in soil conditions to allow an increase in the bioavailability and uptake of the metals in plant tissues (Huang et al., 1997).

The generally low mobility and availability of cadmium in the plant tissues despite the soil being polluted with cadmium ions suggests in part that the trees may have limited potential to bioaccumulate cadmium ions in their tissues. The use of such trees, therefore, as phytoaccumulators for cadmium-polluted soils might be less effective. Igbal et al. (2019) suggested that lowering the soil pH of a polluted site could increase the efficiency of phytoremediation and thus aid the rapid removal of metals such as cadmium from the polluted soil. On the other hand, the near-neutral pH of the soils used in this study might be responsible for the low uptake of cadmium ions from the soil rather than the actual potential of the trees themselves. It has been reported that an increase in plant growth rate and cadmium uptake by plants negatively correlated with the soil pH (Ali et al., 2020; Kindtler et al., 2019). Rosselli et al. (2003) provided some criteria for a plant to be chosen for phytoremediation of a heavy metal polluted site and a major one of these criteria is that the plant should be able to grow properly on the contaminated soils. Based on this criterion, the different tree species used in our study could be considered phytoremediators as they all were established successfully on heavy metal-polluted soils and generally might have phytostabilized the cadmium ions during the growth period, hence the low concentration of cadmium in the plant tissues.

We computed the overall metal uptake in the various plant sections across the tree species to address the question of which portions of the plants the metals mostly accumulated in. We discovered that heavy metal accumulation typically occurs in the stem, regardless of the type of tree. The roots also have a considerable heavy metal accumulation second to the stem, with the leaves having the lowest accumulation capacity (Fig. 10). These findings corroborated the observations made by Olajuyigbe and Aruwajoye (2014) and Majid et al. (2011) and concluded that differences exist in the heavy metal buildup and tolerance in actively growing plant tissues. The interactive effects of contamination levels and tree species established that contamination at triple the permissible level (Conta 3) and *Tectona grandis* have the highest heavy metal accumulation and this was seconded by *T. ivorensis.* Hence, *Tectona grandis* and *T. ivorensis* significantly and consistently demonstrated a high phytoremediation potential for cleaning up chemically polluted soils based on phytoextraction mechanisms. Thus, we support and add to the conclusions made by Blaylock et al. (1999) in their investigation of a lead-polluted site in New Jersey.

Conclusion

Our study suggested that heavy metal mobility and bioavailability in plant tissue were tree-specific, growth stage-dependent, and metal-specific; showing that trees have a preferential affinity to accumulate a specific pollutant. The trees were categorized into specific/mono or non-specific/multi-accumulators based on their metal selectivity. The study suggested *Shorea roxborghi* as a mono-accumulator of manganese while *Tectona grandis* and *Terminalia ivorensis* were multi-accumulators of Mn, Cu, Zn, and Pb. The highest accumulation of heavy metals was observed in the stems followed by the roots which was independent of tree type. The interactive effects of contamination levels and tree species portend *Tectona grandis* and *Terminalia ivorensis* grown in chemically polluted soils at triple the permissible level to have the highest heavy metal uptake with the highest phytoextraction potential in our study. We conclude that all the tree species considered in our study possess some desirable traits for use as phytoremediators of chemically polluted soils through phytoextraction and phytostabilization mechanisms.

Ethical approval

Not Applicable.

Consent to participate

Not Applicable.

Consent to publish

All authors read and approve the work.

Authors contributions

All authors contributed to the study's conception and design. Material preparation, data collection and analysis were performed by Dr Victor Akanbi Olayiwola, Dr Samuel Ayodele Mesele and Dr Emmanuel Oluwakayode Ajayi. The first draft of the manuscript was written by Dr Victor Akanbi Olayiwola. The final revised version, submission, and correspondence were done by Dr Samuel Ayodele Mesele. All authors commented on previous versions of the manuscript, and read and approved the final manuscript.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

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