



Physiological Response of *Vigna unguiculata* to Heavy Metal Contamination in Mechanic Workshop Soils: A Controlled Pot Experiment

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Abstract

Soil contamination as a result of heavy metals has become a critical environmental challenge due to its far-reaching consequences for food security, human health, and ecosystem integrity. Anthropogenic activities are contributors to the release of heavy metals such as cadmium (Cd), lead (Pb), zinc (Zn), and chromium (Cr) which are of particular concern due to their high toxicity, persistence, bioaccumulation and non-biodegradability. Automobile servicing centers, commonly referred to as mechanic workshops, significantly contribute to soil contamination in urban areas because they handle various repair activities that release heavy metals into the environment. In this study physicochemical properties and heavy metals were assessed in soils from automobile workshops using pot experiments. The result showed that the soils are predominantly sandy loam, exhibiting elevated electrical conductivity (EC), signaling the presence of ionic contaminants. The study also revealed a metal concentration trend of $Zn > Cr > Pb > Cd$ in the soils, which was mirrored in the plants growing in the contaminated sites. Notably, the metal transfer factor (MTF) exceeded unity in many locations, indicating hyperaccumulation of heavy metals in the *Vigna unguiculata* and highlighting the associated environmental and health risks. The trend in MTF values was $Cd > Pb > Zn > Cr$, suggest that cadmium poses the greatest risk of bioaccumulation. These findings emphasize the urgent need for continuous monitoring of soils in automobile workshop areas to mitigate pollution risks. Implementing effective soil management practices and remediation strategies is imperative to safeguard human health, ensure food security, and protect ecosystems.

Keywords: Automobile workshops, Heavy metals, Physicochemical parameters, Pot experiment, *Vigna unguiculata*



Introduction

Soil contamination by heavy metals is a crucial issue that poses a serious challenge in recent times (Feng et al., 2023; Wang et al., 2023). Improper land use and anthropogenic activities are the major causes of heavy metal contamination (Singh et al., 2022). This global issue is a major threat to food security, human health and the ecosystems due to their high toxicity, bioaccumulation, and persistence and non-biodegradability (Wang et al., 2023). Heavy metals like cadmium (Cd), lead (Pb), copper (Cu), zinc (Zn), and chromium (Cr) are considered as the major pollutants present in soils that degrade urban and agricultural environments, posing severe risks to human health and food systems (Cheng et al., 2021).

Excess copper in the soil is potentially toxic to plants causing phytotoxicity by the formation of reactive oxygen radicals that damage cells or by the interaction with proteins impairing key cellular processes, inactivating enzymes and disturbing protein structure (Yruela, 2009). Lead (Pb) is a potential pollutant that readily accumulates in the ecosystem. It is not an essential element for plants, yet it gets easily absorbed and accumulated in different plant parts. Pb toxicity affect various morphological, physiological and biochemical aspects of the plant (Sharma and Dubey, 2005). Excess Pb causes a number of toxicity symptoms in plants including stunted growth, chlorosis and blackening of root system. It inhibit photosynthesis, upsets mineral nutrition and water balance, and causes changes in hormonal status and affects membrane structure and permeability (Sharma and Dubey, 2005). Excess Zn in plants is toxic as it affects crop productivity and threaten food security and human health worldwide (Meng et al., 2023). Zn is involved in physiological, cellular, and molecular processes (Broadley et al., 2007) and its excess is particularly toxic to most vascular plants and affects their growth and development (Kaur et al., 2021). Zn mainly accumulates in plant roots, affecting stem, leaves, and subcellular structure in plants (Meng et al., (2023). Excessive Zn can damage plant growth and development, including seed germination, root and stem elongation, and even lead to death Meng et al., (2023). Cadmium (Cd) is extremely toxic and carcinogenic,



as it shows devastating effects on human health, and crop growth and productivity. In crops, Cd antagonistically competes with nutrients acquisition and disturb plant physiological attributes, thus restricting plant survival in polluted soils (Aslam, Okal & Waseem, 2023). Cadmium interferes on mechanisms on seed germination affects seed osmoregulation, antioxidant status, photosynthetic efficiency, protein and metabolic profile, reduced radicle formation and suppressed lateral root formation. It has no biological role instead it can adversely affect even at very low concentration (Tran & Popova, 2013). Cd can replace calcium due to similar charge and ionic behaviors assisting in accumulation in human body and causing chronic liver damage and restricted calcium supply (Aslam, Okal & Waseem, M. 2023). Among the Cr species, the most mobile and toxic form is the Cr^{6+} . It can be absorbed by plant components and cause negative impact on plant growth and development, disrupting germination, stem, root, and leaf growth. Chromium also hampers vital physiological processes such as photosynthesis, mineral nutrition, and water-soil interactions (Dey, Sharma & Kumar, 2023).

In Nigeria, automobile servicing centres, popularly known as mechanic workshops significantly contribute to automobile waste in urban areas (Adewole and Uchegbu, 2010). These auto mechanic workshops are found in clusters of open plots of land in the vicinity of urban towns and cities (Nwachukwu et al., 2010). Within these clusters, are people who specialize in electrical aspects of auto repairs, while others engage in repairs of brakes and steering, automatic or standard transmission engine, and spray painting, recharging of auto batteries, welding and soldering etc (Pam, Sha'Ato & Offem, 2013). Similarly, these workshops consume various fossil fuel products, resulting in the accumulation of heavy metals in the surrounding soils. Wastes, including solvents, paints, hydraulic fluids, lubricants, and oil sludge from the auto workshops, increase soil concentrations of heavy metals. These metals, when deposited on the ground, are capable of leaching into surface and groundwater. They can degrade nearby vegetation, infiltrate plant tissues, and contaminate surface runoff causing widespread pollution. Studies have documented decreased levels of essential soil nutrients (e.g., Ca, K, Mg, and cation exchange capacity)



and elevated levels of toxic metals like Zn, Pb, Cd, and Cu in soils impacted by automobile waste (Mbah and Ezeaku, 2010). Often, indiscriminate dumping of the waste alter the soil chemistry, reduces pH, and compromise plant growth (Mbah and Anikwe, 2010). The high concentrations of heavy metals in these wastes also adversely affect diversity, soil microbial population essential for bioremediation and other soil chemistry (Ogbonna and Okeke, 2011).

Generally, pollution effects due to automobile site activities in Nigeria have received limited attention despite the fact that these activities have been shown to produce harmful wastes. In particular, the prevalence of mechanic workshops in urban settings like Kazaure are increasing and yet there is limited research on their specific impacts on soil heavy metal concentrations. This study seeks to address this gap by examining the effects of heavy metal contamination on urban soils affected by auto mechanic workshops using pot experiment.

Material and Methods

Study Area

The study was conducted at the Science Laboratory Technology (SLT) department, School of Science and Technology, Hussaini Adamu Federal Polytechnic, Kazaure, Jigawa State, Nigeria.

Description of sampling sites

Kazaure town is in Kazaure Local Government Area which is in one of the largest Local Government Areas of Jigawa State, Nigeria. The town is located in the Sudanian savannah zone of Nigeria with the geographical coordinates 12°02'N and 9°12'E. The topography is generally flat, with a few scattered hills. The climate is hot and dry, with an average annual temperature of 28 degrees Celsius. The rainy season lasts from June to September, while the dry season lasts from October to May. The region is known for its dry climate and fertile soil, which is well-suited for agriculture. The sampling sites purposively selected for this study were automobile workshops located in different parts of the town.



Sample collection

Soil samples were collected from the designated sampling sites (**Table 1**) at a depth of 30cm. The sample was screened to remove over-sized fractions. Waste cloth, stone, metal, plastic and glass were also removed. A total of eight sampling points including one control (Ctrl) were determined.

Table 1: Sampling locations, their codes and geographical locations

Sampling Site	Code	Geographical locations
Oga Aminu Mechanic Workshop	OAMW	12.110, 8.001
Oga Abba Mechanic Workshop	OABMW	12.04, 8.02
Oga Gambo Mechanic Workshop	OGBMW	12.01, 8.21
Oga Sadik Mechanic Workshop	OSDMW	12.31, 8.06
Oga Sunday Mechanic Workshop	OSMW	12.09, 8.04
Oga Halilu Mechanic Workshop	OHMW	12.30, 8.02
Oga Goje Mechanic Workshop	OGJMW	12.32, 8.09

The allocations of sampling sites were chosen on the basis of the authors' decision and on the grounds of high anthropogenic activities. Soil samples collected were placed in sterilized plastic containers and conveyed to the greenhouse ready for the pot experiment.

Pot experiments

The earthen pot experiment was performed under laboratory conditions, using greenhouse (16 h of light, 8 h of dark, 25°C during the day, 16°C at night). The earthen pots used in this experiment were 35 cm in diameter and 30 cm in height. Each pot was filled with 5 kg of the automobile workshop soil. Bean (*Vigna unguiculata*) seeds were sown in each of the pot including the Ctrl. All pots were irrigated with distilled water daily to prevent wilting during the experimental period (Pachura and Ociepa-Kubicka, 2016). The plants were harvested 10 weeks after sowing and the whole plant was removed from the pots. The whole plant was thoroughly washed with tap water, then rinsed with distilled water. The



plant samples were oven-dried to constant weight at 70 °C. The weight of the dried biomass was recorded, and then the samples were crushed to a fine powder in a porcelain mortar and pestle and stored in plastic bags and placed in a desiccator until analysis.

Sample preparation and laboratory analysis

After harvesting the plants, the soil from the pots were remove and air-dried, homogenized and screened through a stainless steel sieve (2 mm) to unify the sample size and remove residue. The sampled materials from the 10 mm and 5 mm sieves were subjected to analysis.

Soil characterization:

Soil collected from the different automobile workshops were subjected to physicochemical screening according to standard procedure (Rahul Anand, 2015; Elbehiry et al., 2020). Soil pH was determined in a 1: 2.5 (soil/water) suspension by a pH-meter meter. Soil salinity was measured as electrical conductivity (EC) in a 1:5 (soil/water) solution with an EC-meter. Soluble (Cl^-) anions were measured volumetrically (Black 1965). Soil organic matter (SOM) was analyzed using a muffle furnace at 400 °C for 4 h by the loss on ignition method.

Sample digestion

The soil samples were digested using a mixture of HNO_3 - HCl (1/3, v/v). The total heavy concentrations of Pb, Cd, Cr, Cu, and Zn, were determined according to standard method (Rong et al., 2015). To determine the metals in plant samples, 1 g of plant material was digested using a mixture of HNO_3 and $HClO_4$ for 60 min according standard procedure. The metals were thereafter evaluated using atomic absorption spectrometry AAS.

Metal transfer factor (TF)

The TF or transfer coefficient was calculated by dividing the concentration of heavy metals in plants by the concentration of heavy metal in. The transfer coefficient was calculated by dividing the concentration of heavy metals in vegetables by the total heavy metals' concentration in the soil.



$$TF = \frac{C_{Plant}}{C_{soil}}$$

where, C_{Plant} = metal concentration in plant tissue, mg/kg fresh weight and
 C_{soil} = metal concentration in soil, mg/kg dry weight.

Quality Assurance

All instruments were properly calibrated while all apparatus used was washed thoroughly with tap water and rinsed with distilled water. To ensure quality assurance of the results, all analysis was conducted in duplicate, the results were then subjected to statistical analysis where the mean and standard deviations were recorded.

Result and Discussion

Physicochemical parameters of soil from Automobile Workshops

The results of physicochemical parameters presented in **Table 1**. The range of the mean values of soil bulk density (gcm^3) is between 21.90 ± 0.04 (GBS) to 124.20 ± 0.028 (UBK). The mean value obtained from the control site was reported as $152.9 \pm 0.00 \text{ gcm}^3$ was above all the samples. High bulk density may lead to retardation in water infiltration and plant root penetration which can consequently result poor plant growth. The mean percentage soil moisture contents recorded varied from the highest $2.99 \pm 0.02 \%$ in UBK and the lowest value of $0.43 \pm 0.04\%$ in GBS. The control site had $0.95 \pm 0.035\%$ MC. The higher concentration for moisture content in UBK could be attributed to climatic conditions and the nature of the soils. It could be that the soils have higher water retention capacity owing to the particle sizes. These results agree with the findings of Devatha et al., (2019) who stated that soil moisture content depends on climatic conditions. The soil particle size also coupled with mixed oil petroleum contaminants like crankcase oils reduces soil moisture content proportionally with concentration of contaminants. Mean values of pH obtained from the Table 1 varied between 6.06 ± 0.6 in BKA to 8.27 ± 0.06 in TSH while the value obtained from the Ctrl area was 7.975 ± 0.02 . A study conducted by Azorji et al., (2021) in Imo state, also reported a similar range of pH values recorded at automobile mechanic



villages. The results of pH values showed that the soil samples were slightly acidic in BKA and basic in TSH while soil samples from other sample sites reported as neutral including control site. Previous study has shown that contamination with petroleum and its products decrease soil pH and with increasing concentration of contaminants, soils become more acidic (Ohanmu et al., 2018). Studies have revealed that pH values between 6.5 and 7.5 is considered optimum for the growth of many plants. Total chloride contents in the soils showed variations from site to site. In TSH the Cl^- content (mg/kg) recorded was the highest 12.10 ± 0.78 whereas KCB has the lowest value of 2.65 ± 0.21 mg/kg while control site with 8.40 ± 0.04 . An overabundance of Cl^- in the environment can impair agricultural land utilization by decreasing soil fertility and by causing crop toxicities. Thus, contamination of soils with Cl^- must be avoided. The mean value obtained from the electrical conductivity (EC) results indicated the highest $541.00 \pm 1.41 \mu\text{S}/\text{cm}$ at TRF and the lowest value of $199.50 \pm 0.07 \mu\text{S}/\text{cm}$ recorded at GBS which was all above the control site with $189.5 \pm 0.70 \mu\text{S}/\text{cm}$. EC is a measure of soil contamination and pollution. It is an indication of the presence of some ions in the soil as reported by (Edwin-Nwosu and Nkang 2019). Typically, the electrical conductivity values for the sampled automobile workshops indicated increased EC with corresponding increase in auto wastes. These findings are in consonance with the findings of Devatha et al. (2019) who asserted that petroleum hydrocarbons contain high amounts of ions which can increase the EC of the soils they contaminate. The implication of the EC content in this study is an indication of the presence of trace metals ions or ionizable materials in the soil (Kardol et al., 2016). Soil samples with high EC values can affect the growth of many crops (Nyarko et al., 2019). The result further showed that the textural class of the soil sample is sandy loam in which sand has very high percentage compared to clay which was very low.



Result and Discussion

TABLE 1 a. Physicochemical parameters of soil from Mechanic Workshops

SITE	BKA	KCB	UBK	OAP	GBS	TRF	TSH	Ctrl
SBD (g/cm ³)	37.4±0.14	29.85± 0.07	124.2± 0.028	35.1± 0.42	21.9± 0.04	32.45±0.21	24.2± 0.57	152.9± 0.0
MC (%)	0.45± 0.07	0.75± 0.07	2.99± 0.02	0.67± 0.04	0.43± 0.04	0.95± 0.07	1.85± 0.07	0.95± 0.35
pH	6.06 ± 0.6	7.35± 0.04	7.07± 0.05	7.44± 0.05	7.19± 0.04	7.56± 0.03	8.27±0.06	7.975± 0.02
Chloride Content (mg/kg)	4.75± 0.07	2.65± 0.21	2.90± 0.14	4.25± 0.07	5.15± 0.21	3.5± 0.14	12.10± 0.78	8.40± 0.14
EC (µS/cm)	459.00±1.41	359± 1.41	331± 1.41	449± 1.41	199.5±0.07	541± 1.41	319.5± 0.71	189.5± 0.70
SOM (%)	5.87± 0.00	4.28± 1.41	6.65± 0.71	5.23± 0.00	4.28± 1.41	4.28± 1.41	5.23± 0.00	5.87± 0.00
%SAND	61.20	62.50	58.80	80.00	60.80	43.50	42.80	62.70
%SILT	20.40	25.00	14.30	5.70	4.30	21.70	26.40	16.20
%CLAY	18.30	12.50	26.40	14.20	32.50	34.70	29.40	3.40
Soil Textural Classes	Loamy Sand	Loamy Sand	Loamy Sand	Loamy Sand	Loamy Sand	Loamy Sand	Loamy Sand	Loamy Sand

SBD = Soil Bulk Density, EC = Electrical conductivity, SOM = Soil organic matter, MC = Moisture content



This corroborates the works of (Loska, et al., 2004). The mean values for soil organic matter (SOM) obtained from UBK were relatively high with $6.65 \pm 0.71\%$ and TRF, GBS, KCB with lowest mean value of 4.28 ± 1.41 . According to the rating of Odoh, (2024) the soil organic carbon can be rated as extremely low.

Heavy Metals from the automobile mechanic workshop

Figure 2 presents the results of the heavy metal concentration (mg/kg) in the soil samples. It is seen that Cd mean concentration ranged from 0.75 mg/kg recorded at UBK, OAP and GBS to 3.00 mg/kg recorded at TSH. Compared to the other studied metals, Cd concentration was the least in the study area. However, Cd is extremely toxic and carcinogenic, as it can manifest its devastating effects on human health, and crop growth and productivity even in small quantity since it can accumulate over time (Aslam, Okal & Waseem, 2023, 2023). The level of Cd in the present study had exceeded the stipulated permissible Dutch Standards in all the samples except UBK, GBS and OAP. Lead (Pb) concentration in the sampling area ranged from 0.75 mg/kg recorded at TRF to 6.0 mg/kg observed at UBK. Lead is one of the potential heavy metals that is neither essential element nor has any role in the process of cell metabolism but it is easily absorbed and accumulated in different parts of a plant (Hadi & Aziz, (2015)). In this study, the Pb level in the soil samples did not exceed the permissible limit set by the Dutch Standards which is 85 mg/kg (WHO, 1996). Chromium (Cr) concentration (mg/kg) ranged from 15.0 mg/kg recorded at UBK to 50.25 mg/kg recorded at TSH. Oliveira (2012) reported that Cr mainly accumulates in the plant roots, followed by stems and leaves with only a small amount of the metal translocated to leaves. The permissible limit for Cr according the Dutch standards is 100 mg/kg (WHO, 1996). The Cr content in all the sample sites did not exceed the permissible limit. Among the metals, Zn showed the highest concentration with a range of 8.50 mg/kg recorded at BKA to 61.50 mg/kg recorded at TSH. Plants require Zn for normal growth and optimum yield and its deficiency can affect plant productivity and yield and decrease the amount of cereal grain and diminishes its nutritional quality (Sadeghzadeh, (2013)). The general trend of metal concentration in the automobile workshops followed the pattern $Cd < Pb < Cr < Zn$. A similar trend was reported by Mbong et al., (2013). Generally,

the mean concentrations of the heavy metals are higher in soil samples from the automobile mechanic workshops except Cr in which the Ctrl area had concentration levels higher than BKA,KCB, OAP GBS TRF and TSH.

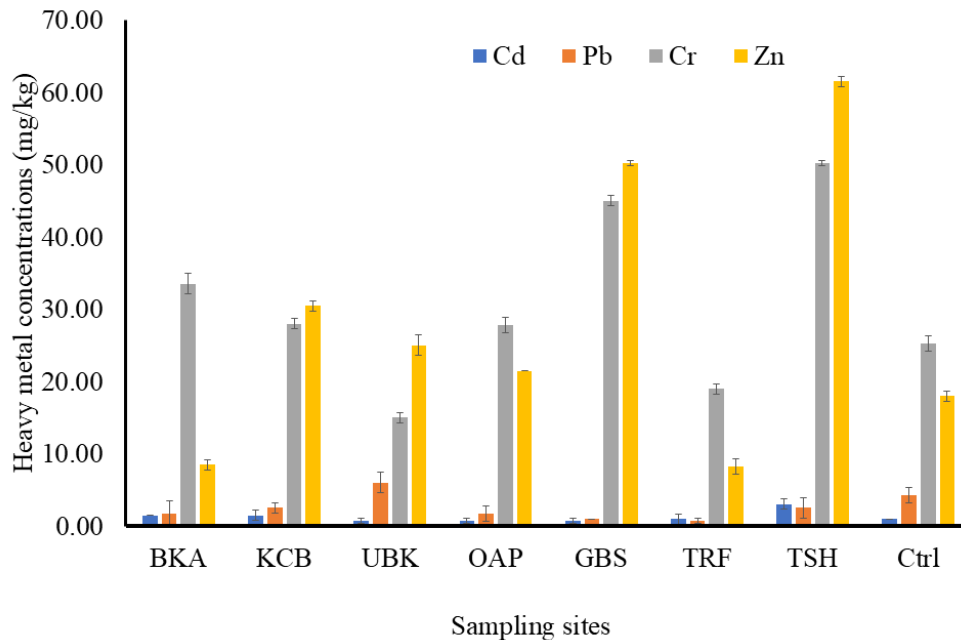


Figure 1. Heavy metal concentration (mg/kg) in soil samples collected from the mechanic workshops

The observed high concentration of the metals is attributable to the level of anthropogenic activities going on in the areas where the soil samples were obtained.

Heavy metals in plants

The results of heavy metal concentration in the studied plant (*V. unguiculata*) is illustrate in the **Figure 2**. Cd depicted the lowest concentrations (0.75 mg/kg) in three samples including BKA, GBS and TSH. The highest concentration (4.0 mg/kg) was recorded in KCB. The permissible level of Cd in plants according to the Dutch standard is 0.02 mg/kg/ which indicates that the levels in the present study have all exceeded the permissible limit. Lead on the other hand had concentration range between 0.75 (mg/kg) to 4.25 mg/kg in KCB and OAP. The plant samples in the Ctrl depicted higher Pb concentration 8.25 mg/kg. Except for KCB, OAP TSH and the Ctrl area, all the other samples had Pb concentration levels within the stipulated limit of the Dutch standard (Osmani, Bani & Hoxha, 2015) .

The Cr content in the plants depicted high concentrations which ranged between 2.25 mg/kg (UBK) to 30.0 mg/kg recorded at TRF. The Zn concentration (mg/kg) was in the range of 0.50 mg/kg recorded at TSH to 30.50 mg/kg observed at KCB and OAP, respectively. The Ctrl area also showed high concentration (29.50 mg/kg) of Zn which may be attributed to natural or geogenic process. Again, the trend in metal concentration in the *V. unguiculata* followed the trend Cd>Pb>Cr>Zn. The observed increased in the heavy metal availability index of the automobile workshop plants could be linked to the indiscriminate oil spillage, anthropogenic activities into these sampling area.

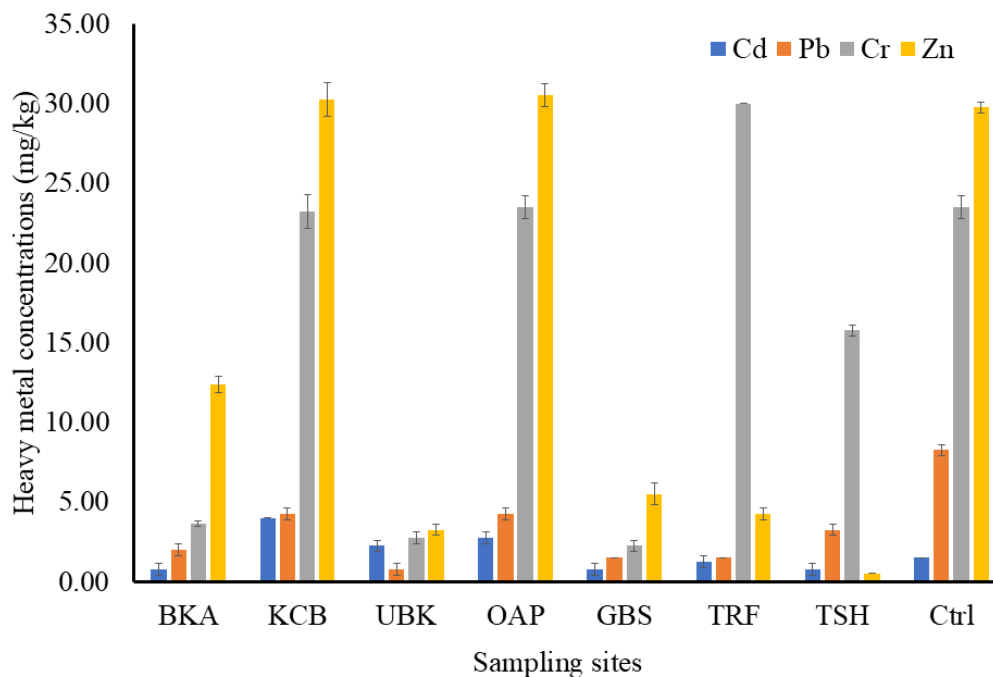


Figure 2b chart shown the heavy metals in plant from automobile workshops

Studies have shown that plants from soils of the mechanic workshops revealed higher levels of the metals (Mbong, et. al. 2013) which implies significantly increased potential of uptake of the metals. In another similar studies conducted by Achi et al., (2011), levels of heavy metals were detected in plants growing in roadside soils in Kaduna metropolis (Achi et al., 2011). This revelation confirms a direct relationship existing between anthropogenic activities like driving in the urban area and phytoavailability of heavy metals in this species due to their proximity to mechanic workshop areas. Similarly, Awofolu,

(2005), demonstrated positive relationships between plants and soil content of metals which he noted as potentials to metal toxicities to both plants and animals through their entry into food chain.

Metal transfer factor (MTF)

Metal transfer factor is considered one of the vital parameter index for assessing the bioaccumulation of the metals in the plant tissues. In other words, it represents the bioavailability of metals in a particular soil-substrate to plant species (Laptiev et al., 2024). Table 2.0 summarized the values of MTF from soil to *V. unguiculata* in the study area. The mean values of MTF for Cd, was greater than 1.0 in all the sampling areas except for BKA and TSH. The highest value (3.67) was recorded at OAP. The observed high values of MTF in Cd suggests its high bioavailability and potentials to accumulate in the plant and get transferred through food chain to humans and animals. Ranking next to Cd in MTF value is Pb. Except for UBK, the MTF values were all above unity in all the sampling sites. The highest MTF index was observed at OAP. These high values also suggest

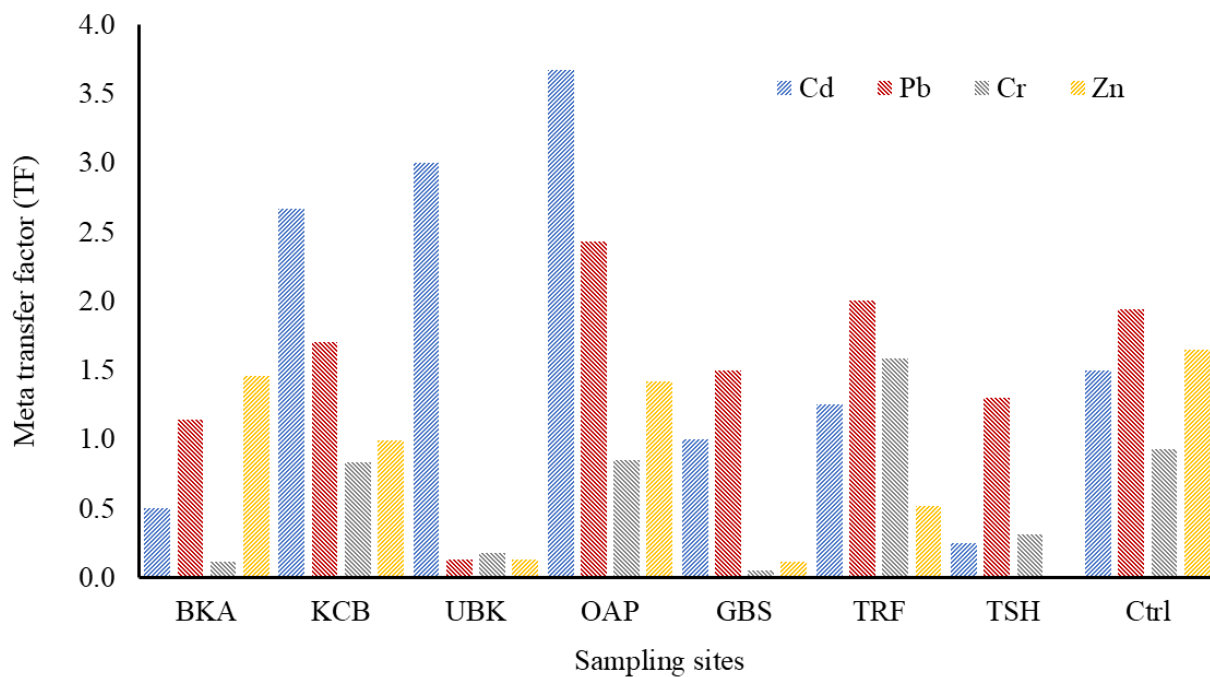


Figure 3. Metal transfer factors in the sampling sites



that the metal can bioaccumulate and get transferred through food chain to consumers. Cr showed wide variations in terms of MTF values. However, the highest recorded value was 1.58 (at TRF) which was above unity. The results shows that Cr did not reveal high potential to bioaccumulate in the *V. unguiculata* tissues. Zinc similarly depicts low MTF values in this study. The levels ranged from 0.01 (TSH) to 1.46 at BKA. Only BKA, OAP and the Ctrl area had values above unity. This implies that the metal depicted low potential to bioaccumulate in the *V. unguiculata* tissue. The general sequence of MTF values of the studied metals in the beans sample was in the order of $Cd > Pb > Zn > Cr$. Plants with MTF values greater than 1 are often referred to as hyper accumulators. The observed differences in MTF values in the study are thought to be due to the differences in soil properties, plant physiology, and environmental conditions of various study sites in the study area (Voutsas et al., 1996).

Conclusion

Concentration of different elements in plants depends upon the relative level of exposure of plants to the contaminated soil as well as the deposition of toxic elements in the polluted air by sedimentation. In this study, the soil-to plant Transfer Factor (TF) for various metals and for most common plant showed that the TF values differed significantly between locations and between plant species. TF decreased when the plants were grown in the higher soil heavy metal contamination. Heavy metal-contamination of agricultural land is one of the abiotic stresses that limit crop productivity. Genetic engineering and the recent genome editing approaches have been used to confer heavy metal resistance in plants. One of the strategies of tackling heavy metal toxicity in plants is to target the initial step of uptake of heavy metals by plants. A crucial, yet often neglected, aspect of phytoremediation involves recovery or disposal of heavy metals (accumulated in plants) in such a manner that the plant biomass can be properly handled and the associated environmental risks could be reduced. Further, investigations should be focused on rhizosphere and soil microbial diversity, which affect heavy metal solubility. Work could also be directed toward endophytes (nonpathogenic microbes inside the plant organs) that provide resistance against heavy metals.



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