

Phytoextraction Potential Of The Plant, Triticum Aestivum for The Metals; Zinc, Cobalt, Copper, Lead And Nickel

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ABSTRACT: In this paper, laboratory pots experiment was conducted to assess the phytoremediation potential of the native crop plant species, *Triticum aestivum*. Viable seed of the grass were seeded into soil (2.0kg) amended with different concentrations of the metals, Zn, Pb, Co, Cu and Ni. Experimental soil was amended with, 1000, 2000, and 4000 ppm for Zn as $Zn(SO_4)_3 \cdot 6H_2O$; 150, 500, and 1000ppm for Ni and Pb as $Pb(NO_3)_2$ and $Ni(NO_3)_2 \cdot 6H_2O$; 150, 250, and 400ppm for Co and Cu as $CuSO_4 \cdot 5H_2O$ and $CoCl_2 \cdot 6H_2O$ respectively. Plants were allowed to grow under careful supervision with adequate watering for a period of eight weeks along with the control experiment. Plants were harvested by pulling carefully to avoid damages to the roots. Separated into roots and shoots and washed with tap water. Soil, roots and shoots of the grass were analyzed using atomic absorption spectroscopy (AAS) following digestion with aqua-regia for the soil and 6M HCl for the plant parts. The bioconcentration (BCF), enrichment (EF) and translocation factors (TF) of the metals were determined from their concentration in the soil, root and shoot. The results showed that, Zn had BCF values greater than one (1) at all the three different levels of the element in the pots as well as the control; 1.22, 4.11, 1.60, and 2.38; EF of 0.95, 2.90, 0.26, and 4.10; TF of 0.77, 0.70, 0.24 and 1.72 for the control. Lead has BCF values of 0.16, 0.08, 0.10, and 0.26; EF of 1.35, 1.30, 0.48, and 0.74; TF of 8.50, 14.60, 4.60, and 2.80 for the control. Cu has BCF values of 0.78, 1.57, 0.90, and 1.13 for the control; EF of 0.62, 1.07, 0.67, and 0.90 for the control. Nickel had the BCF values of, 1.76, 0.42, 0.20, and 0.48 for the control; EF of 0.19, 0.12, 0.05, and 0.09 for the control, TF of 0.11, 0.28, 0.22, and 0.19 for the control. High values of one (1) and above for the BCF and EF indicates absorption, high retention and concentrating of the metals in the roots zone with less translocation to the shoots. The plant, *T. aestivum* may therefore be best described as a metal excluder or stabilizer for Zn, Co and Cu having greater values of BCF and EF than TF. A phenomenon known as phytostabilization. It could also absorb and translocate the metal, Pb to the shoots having TF values greater than one (1), far greater than the EF values, since the BCF value are less than one (1), the degree of absorption is not strong, therefore the plant may best be described as Pb indicator. A phenomenon whereby plants may absorb and accumulate metals in their roots and translocating simultaneously to the shoots; thus, the metals levels in the shoots reflect the metal levels in the soil.

KEYWORDS: Plants, Heavy metal, Accumulation, Hyperaccumulation, Phytoextraction, Phytostabilization, Soil

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I. INTRODUCTION

The global problem concerning contamination of the environment as a consequence of human activities is on the increase. Contamination has resulted from industrial activities, such as mining and smelting of metalliferous ores, electroplating, gas exhaust, energy and fuel production, fertilizer and pesticide application, and generation of municipal wastes [1]. These have resulted in environmental buildup of waste products of which heavy metals are of particular concern [2]. Excessive metal concentration in the soil pose significant hazard to human, animal and plant health and to the ecosystem in general [3].

Heavy metals are natural constituents of the earth's crust [4, 5]. Their principal characteristics are an atomic density greater than 5 g cm^{-3} [6, 7] and an atomic number >20 [4]. The most common heavy metal contaminants are Cd, Cr, Cu, Hg, Pb, and Zn. From the geochemical point of view, trace elements are metal whose percentage in rock composition does not exceed 0.1% [8]. The occurrence of heavy metals in soils can result of two main sources: Natural source: Heavy metals occur naturally in the soil environment from the

pedogenetic processes of weathering of parent materials at levels that are regarded as trace ($<1000 \text{ mg}\cdot\text{kg}^{-1}$) and rarely toxic [9, 10]. Anthropogenic sources: Human activities, such as mining, smelting, electroplating, energy and fuel production, power transmission, intensive agriculture, sludge dumping, and melting operations, are the main contributor to heavy metal contamination [11, 12]. Heavy metals in the soil from anthropogenic sources tend to be more mobile, hence bioavailable than pedogenic, or lithogenic ones [13, 14]. Heavy metals are therefore the major environmental contaminants and pose a severe threat to human and animal health by their long-term persistence in the environment [15].

The remediation of soils contaminated by heavy metals is a cost-intensive and technically complex procedure [16, 17]. Conventional remediation technologies are based on biological, physical, and chemical methods, which may be used in conjunction with one another to reduce the contamination to a safe and acceptable level. In spite of being efficient, these methods are expensive, time consuming and environmentally destructive. At the same time, they are usually harmful to the natural soil environment, and generate large amounts of waste [18]. The global emphasis at present is to use natural methods to curb pollution and reclaim polluted soils. Bioremediation is based on the potentials of living organisms, mainly micro-organisms and plants, to detoxify the environment [19]. High contents of both essential and non-essential heavy metals in the soil may inhibit plant growth and can lead to toxicity symptoms in most plants [20, 21, 22]. However, some plant species have the ability to grow and develop in metalliferous soils such as near to mining sites [23]. Such plants can be used to clean up heavy metal contaminated sites. Willow (*Salix viminalis* L.), maize (*Zea mays* L.), Indian mustard (*Brassica juncea* L.), and sunflower (*Helianthus annuus* L.) has been found to be highly tolerant to heavy metals [24]. Vetiver grass (*Vetiveria zizanioides*), for instance, showed tolerance to Pb and Zn and it can be used for revegetating Pb/Zn mine tailings [25]. Plant based bioremediation technologies have been collectively termed as phytoremediation; this technology can be applied to both organic and inorganic pollutants present in soil (solid substrate), water (liquid substrate) or the air [18, 26].

Phytoremediation can be defined as the process, which uses green plants for the relief, transfer, stabilization or degradation of pollutants from soil, sediments, surface waters, and groundwater [18, 27]. It is a general term including several processes, in function of the plant-soil-atmosphere interactions. Some plant roots can absorb and immobilize metal pollutants, while other plant species have the ability of metabolizing or accumulating organic and nutrient contaminants. For heavy metal contaminated soil, four processes of phytoremediation are recognized. Phytoextraction, phytostabilisation, phytovolatilization and rhizofiltration. The two first mechanisms are the most reliable. The different forms of phytoremediation require different general plant characteristics for optimum effectiveness [28]. Plant species differ widely in their ability to accumulate heavy metals. Many authors concluded that concentrations of metals in plants growing in the same soil vary between species and even between genotypes of a species [29, 30]. Accordingly, the response of plants to bioavailable heavy metals in the soil has been classified as follows: 1) Metal excluders; are those plants that prevent metal uptake into their roots and/or avoid translocation and accumulation into shoots over a wide range of metal concentrations in the soil. They have a very low potential for metal extraction, but they can be used to stabilize the soil, and avoid further contamination spread due to erosion. 2) Metal accumulators; this group of plants can accumulate metals in their above ground tissues in concentrations far exceeding than those present in the soil, such plant species are termed as hyperaccumulators. 3) Metal indicators; these categories of plants show poor control over metal uptake and transport processes, and accumulate metals in their above ground tissues. The extent of metal accumulation in the tissues of these plants reflects metal concentration in the rhizosphere. Indicator species have been used for mine prospecting to find new ore bodies [18, 26, 31, 32]. Figure 1 summarizes definition and principle characteristics of some of the process.

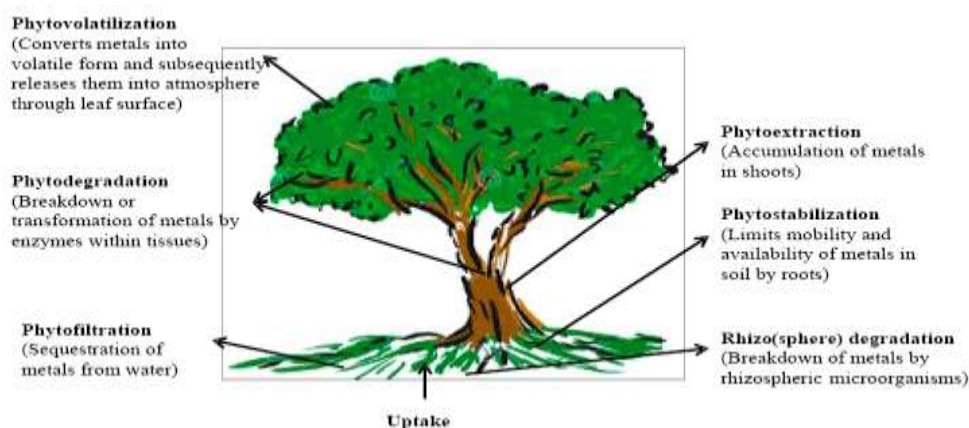


Figure 1: The different techniques of Phytoremediation

Plant species for phytoremediation are selected based on their root depth, the nature of the contaminants and the soil, and regional climate. The root depth directly impacts the depth of soil that can be remediated. It varies greatly among different types of plants, and can also vary significantly for one species depending on local conditions such as soil structure, depth of a hard pan, soil fertility, cropping pressure, contaminant concentration, or other conditions [33]. It has been reported that for phytoremediation, grasses are the most commonly evaluated plants. They have been more preferable in use for phytoremediation because compared to trees and shrubs, herbaceous plants, especially grasses, have characteristics of rapid growth, large amount of biomass, strong resistance, effective stabilization to soils and ability to remediate different types of soils. They are pioneers and usually are adapted to adverse conditions such as low soil nutrient content, stress environment and shallow soils [34, 35, 36, 37]. The large surface area of their fibrous roots and their intensive penetration of soil reduces leaching, runoff, and erosion via stabilization of soil and offers advantages for phytoremediation [38]. Wild plants such as grasses can produce closures above ground quickly and reduce dust dispersion of tailings [39].

The primary objective of the present work was to establish the accumulation capacity and phytoremediation potential of the grass plant *Triticum aestivum* in cleaning or stabilizing heavy metal laden soil, especially Zn, Co, Cu, Pb and Ni. During the experiments, some observations were also made concerning the morphological characteristics of the plants as it absorbed the metal.

II. METHODOLOGY

Sampling and experimental area

The seeds of the grass (*Triticum aestivum* L) along with the soil that support its growth were obtained from Lake Chad Research Institute situated at KM 5 Gaboru Ngala Road, Maiduguri, Nigeria.

Pot experiment design

Pot culture experiment containing 2 kg soil was conducted according to the method described by Ahalya et al. (2005) [40], the soil was spiked with the following heavy metals; Ni as $\text{Ni}(\text{NO}_3)_2 \cdot 6\text{H}_2\text{O}$, Pb as $\text{Pb}(\text{NO}_3)_2$, Zn as $\text{Zn}(\text{SO}_4) \cdot 6\text{H}_2\text{O}$, Cu as $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$, and Co as $\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$ at a concentration of 150, 250, and 400 ppm for Co and Cu; 150, 500, and 1000 ppm for Pb and Ni whereas 1000, 2000 and 4000 ppm for Zn. Viable seeds of the grass were planted into the pots. A separate pot containing non-treated soil was used to serve as a control. Experiments were exposed to natural day and night temperatures. Since humidity is one of the factors ensuring the growth of plants and the necessary physiological processes, irrigation of the pots was done with 500 ml of water after every five days in the evening hours [41] (Garba et al., 2011). Experiments were monitored for any morphological changes for a period of eight weeks. Four replicates of each pot of the grass were planted for statistical handling. At the end of the experiment, the grass and soil were collected, washed with tap water and carefully separated into; roots and shoots. Air dried at room temperature to a constant weight, ground and sieved using a 2 mm nylon sieve. The experimental soil was equally collected, homogenized, dried at 105°C to a constant weight, ground and then sieved. The soil, root and shoot of the grass were analysed for the heavy metals; Zn, Ni, Cd, Cu and Pb following digestion with concentrated HNO_3 , H_2SO_4 , and HClO_4 acid in the ratio, 5:1:1, [42] (Allen et al., 1986). The soil was characterized for its physicochemical properties.

III. RESULTS AND DISCUSSION

Physicochemical properties of the soil

The Result of the physicochemical properties of the experimental soil is shown in Table 1 below. The soil texture shows that it is more of sandy and predominantly sandy loamy in texture. Soil texture reflects the particle size distribution of the soil and thus the content of fine particles like oxides and clay [43] (Sherene, 2010). Particle size distribution can influence the level of metal contamination in a soil. Fine particles (<100 μm) are more reactive and have a higher surface area than coarser material. As a result, the fine fraction of a soil often contains the majority of contamination [43, 44] (Sherene, 2010; Evanko and Dzombak, 1997).

The pH value ranged between 7.58 to 7.61 which is in a neutral range. Soil pH plays an important role in the sorption of heavy metals, it controls the solubility and hydrolysis of metal hydroxide, carbonate, and phosphate [41] (Garba et al., 2011). Metal cations are the most mobile under acidic conditions while anions tend to sorb to oxide minerals in this pH range [45] (Dzombak and Morel, 1987). Thus, at low pH, metal bioavailability increases as more metals are released into the soil solution due to competition with H^+ ions [46]. At high pH, cations precipitate or adsorb to mineral surfaces and metal anions are mobilized [47, 48]. At neutral or alkaline pH, most of the metals in soil are not available to plants [49]. A very low EC was observed, 0.84. Low organic matter content of 0.18 was also observed as well as very low CEC 3.79 mol/100g of soil. Soil organic matter is frequently reported to have a dominant role in controlling the behavior of trace metals in the soil [50]. Increasing the amount of organic matter in the soil helps to minimize the absorption of heavy metals by plants. Land rich in organic matter actively retains metallic elements [51]. Soils with relatively low organic matter concentration are more susceptible to contamination by trace elements [52]. CEC measures the ability of

soil to allow for easy exchange of cation between its surface and soil. The low level of clay and CEC indicate the permeability and leach ability of metals in the soil [53].

Table 1: Physicochemical Properties of the soil

Parameters	Experimental Soil
pH	7.61 ±1.0
EC (dsm ⁻¹)	0.84±0.01
CEC ((cmol/100kg soil)	3.79±0.02
Organic carbon (%)	0.18±1.00
Organic Matter (%)	0.31±0.01
Silt (%)	20.70±0.10
Clay (%)	17.20±0.01
Sand (%)	62.10±0.10
Textural Class	Sandy Loamy

Data are presented as Mean ±SD. Where SD = Standard Deviation with n = 3

IV. HEAVY METAL ACCUMULATION IN THE ROOT AND SHOOT OF WHEAT (*Triticum aestivum* L.)

ZINC (Zn)

In this research work, uptake and accumulation of Zn by *Triticum aestivum* were found at higher levels in the roots at the different spiked concentration of 500, 2000 and 4000ppm of the element in the soil. Report has it that, at toxic level of Zn supply (150 µmol/L) in the soil, a higher proportion of the element may accumulate in the roots [54]. This could be due to the high level of the element in the soil which suggested that, at high level of Zn in soil, *T. aestivum* may retain most of Zn absorbed in the root. Although high level of the metal was observed in the shoot of the grass in the control experiment (Table 2). This agreed to the report by Wang et al. [55] who observed that, high level of Zn was found in the shoot of bread wheat. Ashfaq et al. [56] also reported that, higher level of Zn was found in the shoots of *X. strumarium*, and *B. repensthan* the root. Following absorption by the root, Zn is rapidly transported via the xylem to the shoot in rice [57]. The efficiency of root-to-shoot translocation is theoretically dependent on four processes [58, 59]: (1) Zn sequestration in the root; (2) efficiency of the radial symplastic passage; (3) xylem loading capacity; and, (4) Zn movement efficiency in the xylem vessels. It has been suggested that decreased root cell sequestration may facilitate enhancing Zn root-to-shoot translocation in the hyperaccumulators [60]. Report has it that, in a non-accumulator plants much more of zinc absorbed are sequestered in the root, possibly via storage in the vacuoles and rendered unavailable for translocation to the shoot [61]. Despite the high concentration of the metal in the experimental pots no visible sign of toxicity was observed on the plants, (Figure 2) as compared with the control (Figure 9). Studies have shown that, the first symptom to present itself in most species exhibiting Zn toxicity is a general chlorosis of the younger leaves [62 63] Depending on the degree of toxicity this chlorosis can progress to reddening due to anthocyanin production in younger leaves [64, 65].

Table 2: Level (ppm) of Zn in the Soil, Shoot and Root of *T. aestivum*

Amount Spiked	Soil	Root	Shoot	BCF	TF	EF
500	500.5±0.02	615 ± 0.08	476 ± 0.01	1.22	0.77	0.95
2000	170.5±0.04	701 ± 0.06	492 ± 0.04	4.11	0.70	2.90
4000	1532 ±0.07	1609.5±0.09	391 ± 0.04	1.60	0.24	0.26
Control	186 ± 0.05	442.5± 0.10	763 ± 0.06	2.38	1.72	4.10

Data are presented as Mean ±SD. No significant difference was observed at P<0.05 using one-way anova analysis and multiple comparison according to Tukey Test, SD=Standard Deviation, BCF = Bioconcentration Factor, TF = Translocation Factor, and Enrichment Factor.



Figure 2: The Grass of *T.aestivum* Spiked with different levels Zn in Experimental Pots

COBALT (Co)

Cobalt is a naturally occurring element found in rocks, soil, water, plants, and animals. Cobalt is used to produce alloys used in the manufacture of aircraft engines, magnets, grinding and cutting tools, artificial hip and knee joints [66]. Cobalt compounds are also used to colour glass, ceramics and paints, and used as a drier for porcelain enamel and paints [67]. Radioactive cobalt is used for commercial and medical purposes. ^{60}Co is used for sterilizing medical equipment and consumer products, radiation therapy for treating cancer patients, manufacturing plastics, and irradiating food [68]. ^{57}Co is used in medical and scientific research [69]. Although the average level of cobalt in soils is 8.0ppm, there are soils with as little as 0.1ppm and others with as much as 70 ppm [70]. High concentration of Co accumulated by *T. aestivum* at 150, 250 and 400ppm Co were found in the root (Table 3).

Compare to other heavy metals, Co is not regarded as very toxic to plants and other organism. Toxicity to plants rarely occurs in natural soils [71] unless they are contaminated by pollution [72]. Toxicity, when it occurs is mainly due to human practices. For example, the application of excess Co to crops and animals can cause toxicity, not only to the crops, animals themselves, but also to their consumers through the food chain. The growth of the plant, *T. aestivum* at the different concentrations, 150, 250 and 400ppm Co in this study shows no sign of toxicity (Figure 3), compared to the control (Figure 9). Toxicity of excess Co is linked to oxidative stress, inhibition of photosynthesis and iron deficiency [73, 74]. Cobalt has been reported to disrupt iron homeostasis and compete with iron for access to transporters in many organisms including plants [75]. Yellowing of the tips of older leaves progressing down the leaf margins in wheat plants, has been reported [76], this effect did not however, manifest on the plant even at higher level of the element (250 and 400ppm Co) in experimental pots.

Table 3: Level (ppm) of Co in the Soil, Shoot and Root of *T.aestivum*

Amount Spike	Soil	Root	Shoot	BCF	TF	EF
150	45.5 ± 0.01	109 ± 0.10	89 ± 0.14	2.40	0.82	1.96
250	188 ± 0.05	93.5 ± 0.01	23.5 ± 0.05	0.50	0.25	0.13
400	159 ± 0.05	58 ± 0.03	39 ± 0.03	0.36	0.67	0.25
Control	43 ± 0.02	15 ± 0.08	10 ± 0.05	0.35	0.66	0.23

Data are presented as Mean ±SD. No significantly different was observed at $P < 0.05$ using Anova Analysis and Multiple Comparison according to Tukey Test, SD=Standard Deviation. TF = Translocation Factor, EF = Enrichment Factor and BCF = Bioconcentration Factor.



Figure 3: Triticum aestivum in the Experimental Pots Spiked with Different levels of Co

NICKEL (Ni)

Nickel is a heavy metal, present in soil, water and air, usually in trace amounts. However, rapid industrialization and urbanization during the recent past have caused accumulation of Ni and many other trace elements in varied habitats. At low concentration, Ni in nutrient medium has been found to stimulate growth in higher plants [77, 78]. In this study, absorption of Ni at different concentrations in the experimental soil (150, 500 and 1000 ppm Ni) showed no sign of toxicity on the plant, *T. aestivum* (Figure 4) compare with the control (Figure 9). Reports has it that, the impact of Ni toxicity on the physiology of plants depends on the type of plant species, growth stage, cultivation conditions, Ni concentration and exposure time [29, 79, 80, 81, 82] in the soil. The toxic effects of higher concentration of Ni are observed at multiple levels, these include inhibition of mitotic activities [83], reduction in plant growth [84], plant water relation and photosynthesis [85], inhibition of enzymatic activities as well as nitrogen metabolism [86], interference with the uptake of other essential metal ions, induction of oxidative stress [85].

The uptake of Ni in plants is mainly carried out through the root system via passive diffusion and active transport [87]. Uptake of Ni by plants depends on the concentration of Ni^{2+} , plant metabolism, the acidity of soil or solution, the presence of other metals and organic matter composition [85]. However, uptake of Ni usually declines at higher pH values of the soil solution due to the formation of less soluble complexes [88]. For example, the uptake of Ni^{2+} by *Lathyrus sativus* reportedly increased with increasing pH up to 5.0 and decreased as the pH is increased further up to 8.0 [89]. Moreover, Ni^{2+} ion may also compete with other essential metal ions when it is absorbed by roots. In this study, the results showed that, high level of the element was absorbed and retained in the root, with very small fraction translocated to the shoot (Table 4). Similar observation has been reported by Chandra et al. [90], in the accumulation and distribution pattern of different metals in different parts of wheat irrigated with effluents, high level of Ni was found in the root (16.80 ± 2.08) than the shoot (4.18 ± 0.92). These two observations however, contradict the report by Wang et al. [55], who observed high level of Ni in the shoot of wheat plant. In contradiction to this findings, Ni accumulation has been reported to be more pronounced in roots rather than the shoot [91]. The path of Ni transport in plants is from root to shoot [92] and makes an exit through transpiration stream [93] via xylem.

Table 4: Level (ppm) of Ni in the Soil, Shoot and Root of *T. aestivum*

Amount Spiked	Soil	Root	Shoot	BCF	TF	EF
150	498.5 ± 0.04	877.5 ± 0.10	96 ± 0.06	1.76	0.11	0.19
500	637 ± 0.05	269. ± 0.04	78.5 ± 0.04	0.42	0.28	0.12
1000	1311 ± 0.03	268.5 ± 0.09	60 ± 0.06	0.20	0.22	0.05
Control	484 ± 0.06	230 ± 0.06	43 ± 0.01	0.48	0.19	0.09

Data are presented as Mean ± SD. No significantly different was observed at $P < 0.05$ using Anova Analysis and Multiple Comparison according to Tukey Test. SD = Standard Deviation, BCF = Bioconcentration Factor, TF = Translocation Factor, and Enrichment Factor.



Figure 4: Triticum. aestivum in the Experimental Pots Spiked with Different levels of Ni

LEAD (Pb)

The uptake, translocation and accumulation of Pb by the grass in the experimental pots spiked with the levels 150, 500 and 1000ppm Pb is as shown in table 5. The result indicates that, *T. aestivum* absorbed and translocate higher level of Pb in the shoot. This agrees with the report by Wang et al. [55] who observed high level of the metal Pb in the shoot of bread wheat, the result also agrees with Garba et al. [38] who reported that high level of Pb was observed in the shoot of *E. indica* ($326.00 \pm 4.26 \mu\text{g/g}$) by enhancing the absorption ability of the plant with EDTA (chelation), this suggest that chelation of metal may change the uptake accumulation trends in plants though this may also vary from plant to plant and with metal type. Contrary to these reports, high concentration of Pb was observed in the root compare the shoot in *T. aestivum* [90]. In this study, uptake and high-level accumulation of Pb was found without chelation. It was also observed that as the level Pb spiked in the experimental pots increases, the concentration of the metal in the shoot equally increases (Table 5). It has been reported that, a few plant species accumulate lead to a high concentration in the above-ground parts and are called hyperaccumulators [94, 95].

Uptake and accumulation of Pb in this study was with less or no physically visible stress (Figure 5). Reports has it that, Pb exerts adverse effects on morphology, growth and photosynthetic processes of plants and causes inhibition of enzyme activities, water imbalance, alterations in membrane permeability and disturbs mineral nutrition [96]. Lead is transported from the medium to the root cells by plasma membrane cation channels, especially the Ca^{2+} -channels [97]. It has been shown that high concentrations of lead caused a decrease of germination in rice seedlings and reduces their growth [98]. A considerable decrease in biomass in response to lead stress appears similar was reported in the responses of wheat to increase of lead in soil [99, 100]. It is established that lead toxicity results in enhanced ROS generation [101]. The increase of TBARS formation is a direct consequence of increased ROS formation and thus unsaturated fatty acid peroxidation. The levels of ROS in wheat seedlings are controlled as usual by a complex antioxidant system that consists of enzymes – catalase (CAT), peroxidase (POD), superoxide dismutase (SOD), ascorbate peroxidase (APX) together with scavenger enzymes such as glutathione S-transferase (GST) – and non-enzymatic low molecular mass antioxidants such as glutathione (GSH), ascorbate, tocopherols carotenoids and proline. Proline belongs to this non-specific defense system against lead toxicity, as an inhibitor of lipid peroxidation [102], a free radical scavenger [103], as well as a metal chelator [104], and the increase of its concentration can be considered as a defense reaction against lead administration. These could attribute to the absence of physical morphological toxicity sign on the plant in this study (Figure 5).

Table 5: Concentration (ppm) of Pb in the Soil, Shoot and Root of *T. aestivum*

Amount Spiked	Soil	Root	Shoot	BCF	TF	EF
150	687 ± 0.04	109 ± 0.02	929.5 ± 0.01	0.16	8.5	1.35
500	737 ± 0.02	66 ± 0.03	962.5 ± 0.05	0.08	14.6	1.30
1000	1787 ± 0.04	186 ± 0.03	860 ± 0.04	0.10	4.6	0.48
Control	130 ± 0.03	177.5 ± 0.02	511 ± 0.05	0.26	2.8	0.74

Data are presented as Mean \pm SD. No significant difference was observed at $P < 0.05$ using Anova Analysis and Multiple Comparison according to Tukey Test, SD=Standard Deviation, BCF = Bioconcentration Factor, TF = Translocation Factor, and Enrichment Factor.



Figure 5: *T. aestivum* in the Experimental Pots Spiked with Different Levels of Pb

COPPER (Cu)

In this study, the grass, *T. aestivum* grown in soil spiked with different concentration of Cu showed no visible sign of toxicity at three different concentrations (150, 250 and 400ppm). It also showed uniform growth (Figure 6) compared with the control (Figure 7). Accumulation of the element in the parts of the plant increases as the concentration of the spiked Cu increases with high level observed in the roots at 150, 250 and 400ppm Cu than in the shoot (Table 6). This result agrees with the report by Garba et al. [38] who observed high level of Cu in the root of *E. indica* ($236.00 \pm 3.72 \mu\text{g/g}$), but in contrast, Wang et al. [55], observed high level of the metal, Cu, translocated to the shoot with the leaves having the highest percentage. Copper (Cu) is an essential redox-active transition metal that is involved in many physiological processes in plant because it can exist in multiple oxidation state in vivo. Under physiological condition Cu exist as Cu^{2+} and Cu^+ , due to its ability to cycle between the oxidize Cu(II) and reduced Cu(I) states, its involved in many biological processes such as photosynthesis, respiration, oxygen super oxide scavenging, ethylene sensing, cell wall metabolism and lignification [105, 106]. Copper act as a structural element in regulatory proteins and participate in photosynthesis, electron transport, mitochondrial respiration, oxidation stress responses, cell wall metabolism and hormone signaling Revan et al. [106].

For the very reason that it is essential, Cu can also be highly toxic [107]. Free Cu catalyzes Fenton reactions that generate hydroxyl radicals causing damage to lipids, proteins and DNA [108]. Copper has been reported to interfere with iron homeostasis [109]. An overall reduction of plant biomass, inhibition of root growth, chlorosis, bronzing and necrosis are the usual reported symptoms of excess Cu due to increased production of reactive oxygen species and harmful interactions at the cellular level. Toxic levels of Cu occur naturally in some soil where as other may contain high level of Cu as a result of the anthropogenic release of heavy metals in to the environment through mining, smelting, manufacturing, Agriculture and waste disposal technology. At concentration above those required for optimal growth Cu was shown to inhibit growth and to interfere with important cellular processes such as photosynthesis and respiration. [81, 110]. However, in this study, no sign of toxicity was observed on the experimental plant as a result of high level of Cu in the soil (Figure 8). Most Cu-tolerant plants respond physiologically as Excluders sensu Baker with very limited Cu translocation from the roots to the shoots [111, 112, 113, 114, 115]. *Crassulhelmsii* (Crassulaceae) was found to accumulate $> 9000 \text{ Igg}^{-1}$ in its shoots at low Cu concentration in the nutrient solution and so is an exception [116].

Table 6: Level (ppm) of Cu in the Soil, Shoot and Root of *T. aestivum*

Amount Spiked	Soil	Root	Shoot	BCF	TF	EF
150	100 ± 0.01	77.5 ± 0.08	62 ± 0.01	0.78	0.80	0.62
250	76.5 ± 0.01	120 ± 0.01	81 ± 0.02	1.57	0.68	1.07
400	154.5 ± 0.02	139.5 ± 0.02	103.5 ± 0.01	0.90	0.74	0.67
Control	77.5 ± 0.00	87.5 ± 0.01	69.5 ± 0.06	1.13	0.79	0.90

Data are presented as Mean \pm SD. No significantly different was observed at $P < 0.05$ using Anova Analysis and Multiple Comparison according to Tukey Test, SD=Standard Deviation, BCF = Bioconcentration Factor, TF = Translocation Factor, and Enrichment Factor.



Figure 6: *T. astivum* in the Experimental Pots Spiked with Different Levels of Cu



Figure 7: *T. astivum* in the Control Pot of the Experiment

V. PHYTOREMEDIATION POTENTIAL OF THE PLANT; *T. astivum*

In most of the established criteria of identifying the metals accumulation plants, it is imperative to consider the metal concentrations in the aboveground biomass and the metal concentrations in the sediments or soil [117]. According to Usman and Mohammed [118], the success of phytoextraction process depends on heavy metal removal by the shoots' tissues. Therefore, we could propose that the investigated plant species could be considered as an accumulator or hyperaccumulators for phytoremediation, since they had generally the higher metal concentrations in their shoots' tissues rather than in their roots' tissues. A plant's ability to accumulate metals from soils can be estimated using the bioconcentration factor (BCF), which is defined as the ratio of metal concentration in the roots to that in soil.

$$BCF = \frac{\text{metal concentration in the root}}{\text{metal concentration in the soil}}$$

A plant's ability to translocate metals from the roots to the shoots is measured using the translocation factor (TF), which is defined as the ratio of metal concentration in the shoots to the roots [119].

$$TF = \frac{\text{metal concentration in the shoot}}{\text{metal concentration in the root}}$$

The enrichment factor (EF) is calculated as the ratio between the plant shoot concentrations and sediment concentrations (metal concentration in shoot/metal concentration in sediments or soil) by Branquinho et al. [120].

$$EF = \frac{\text{metal concentration in the shoot}}{\text{metal concentration in the soil}}$$

Some plants accumulate heavy metals; others exclude them while other plant species are sensitive. In this study, the BCF, EF and TF values for the metals; Zn, Co, Ni, Pb and Cu are presented in tables, 2, 3, 4, and 5 respectively at different level of the metals in the experimental pots.

For Zn, the BCF, TF, and EF values at 500, 2000, and 4000ppm are; BCF = 1.22; 4.11; 1.60; whereas the control has 1.38 respectively; TF = 0.77, 0.70, 0.24, and 1.72 for the control respectively; EF = 0.95, 2.90, 0.26, and 4.10 for the control respectively (Table 2). BCF is used in the determination of the degree of intake and component storage of toxic compounds in plants and animals [121]. For having the BCF and EF values greater than one (1), and TF values less than one (1) with exception of the control, the plant, *Triticum aestivum* may be suggested for phytostabilization of Zn in the root. Besides, plants with Bioaccumulation factor greater than one and translocation factor less than one ($BCF > 1$, $EF > 1$ and $TF < 1$) have the potential for phytostabilization [83]. The control has the ability to stabilize the element Zn in the root, and when the level was increased to 2000ppm in the pots, a property that qualifies the plant, *Triticum aestivum* to serve as a phytostabilization, though it will take time for success to be achieved. This may be suggested that, at high concentration of Zn, *T. aestivum*, can stabilize the level of Zn in the soil through the use of its root. Metal excluders are plants which effectively limit the levels of heavy metal translocation within them and maintain relatively low levels in their shoot over a wide range of soil levels; however, they can still contain large amounts of metals in their roots [122].

The BCF, TF, and EF values for Co at 150, 250, and 400ppm as shown in table 3, are; 2.40, 0.50, 0.36; and 0.35 for the control; TF = 0.82, 0.25, 0.67 and 0.66 for the control; whereas the EF = 1.96, 0.13, 0.25; and 0.23 for the control (Table 3). For Co, high degree of uptake and stabilization at 150ppm Co in the experimental pots, because it has BCF of 2.40, EF of 1.96 whereas the TF was 0.82. Such plants that has BCF, and EF, equal to one or above may be described as potential metal excluders. These are plants which have the ability to accumulate the metals in their roots with less or poor translocation to the aboveground tissues. This process of stabilizing contaminants in the roots just as in the zinc above, is called phytostabilization. Gupta et al. [123] reported that EF values of 1 and above indicate higher availability and distribution of metals in the contaminated environment, subsequently increasing the metal accumulation in the roots of plants species, thus, *T. aestivum* may best be described as Co stabilizer in the soil (a process known as Phytostabilization). This process uses the ability of plant roots to change environmental conditions via root exudates. Plants can immobilize heavy metals through absorption and accumulation by roots, adsorption onto roots, or precipitation within rhizosphere [124]. By using metal-tolerant plant species for stabilizing contaminants in soil, particularly metals, it could also provide improved conditions for natural attenuation or stabilization of contaminants in the soil. Metals accumulated in the roots are considered relatively stable as far as release to environment is concerned.

Nickel has BCF, EF, and TF values at the concentrations of; 150, 500, 1000ppm Ni and the control as; BCF = 1.76, 0.42, 0.20; and 0.48 for the control, respectively; the EF values are; 0.19, 0.12, 3.005; and 0.09 for the control, at 150, 500, and 1000ppm Ni respectively, whereas the TF values are, 0.11, 0.28, 0.22 and 0.19 for the control, respectively (Table 4). Plants with both bioaccumulation, enrichment and translocation factors greater than one (BCF and $TF > 1$) have the potential to be used as phytostabilizer or extractors. In the contrary, nickel has high BCF greater than one but has EF and TF values are less than one (1). This means, the plant can neither serve as a metal excluder or hyper-accumulator nor as a metal indicator.

Lead had the BCF, TF, and EF values at 150, 500, and 1000ppm are; 0.16, 0.08, 0.10; and 0.26 for the control; TF = 8.50, 14.60, 4.60; and 2.80 for the control; EF = 1.35, 1.30, 0.48; and 0.74 for the control (Table 5). The researchers; [125, 126], have pointed out that the ability of phytoremediation has commonly been characterized by a translocation factors (TF). Translocation factor (TF) greater than one (> 1) suggest that, the metals are effectively absorbed through the root and translocated to the shoot for storage [31, 127 and 128]. For having the TF values greater than one (1) at three different concentration of the metal in the experimental pots, *T. aestivum* may be regarded as a potential phytoextractor to reclaim Pb contaminated soil by extraction and translocating high concentration of the absorbed element to the shoot for harvesting. A process known as phytoextraction. Plants exhibiting TF and particularly BCF values less than one are unsuitable for phytoextraction [129].

Copper had the BCF, TF and EF values at the three different concentrations (150, 250, 400ppm Cu) in the experimental pots and the control as follows: it has the BCF of, 0.78, 1.57, 0.90 at 150, 250 and 400ppm Cu respectively whereas the control had 1.13. for the TF, it has, 0.80, 0.68, and 0.74 respectively; the control has 0.79. The EF values for Cu are; 0.62, 1.02, and 0.67 respectively whereas the control has the EF value of 0.90 (Table 6). A BCF value greater than one (1), signifies high degree of intake of the element. In this study, Cu has the BCF and EF values greater than one (1) at 250ppm Cu and TF value less than one (1), this envisaged that, *T. aestivum* may have the potentials to stabilize excess Cu in the soil by absorbing and retaining high concentration of the element in the root zone than translocating to the shoot. A process known as phytostabilization.

VI. CONCLUSION

The phytoremediation potential of the grass; *Triticum aestivum*, assessed in this study showed that, the grass may successfully be used as a phytostabilizer for the metals; Zn, Co, and Cu. This process reduces metal mobility and leaching into ground water, and also reduces metal bioavailability for entry into the food chain. The grass may also serve as a metal indicator for the metal Pb. These are plants which have the ability to accumulate the metals in their roots and translocating simultaneously to the shoots; thus, the metals levels in the shoots reflect the metal levels in the soil.

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