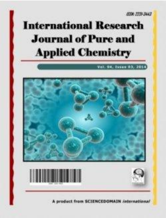

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Accumulation Ability of the Native Grass Species, *Cyperus rotundus* for the Heavy Metals; Zinc (Zn), Cadmium (Cd), Nickel (Ni) and Lead (Pb)

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Authors' contributions

This work was carried out in collaboration between all authors. Author STG designed the study, performed the statistical analysis, wrote the protocol and first draft of the manuscript as well as proof read the final report. Author MG supervised the irrigation, monitored the growth for any sign of toxicity up to harvesting period and managed the analyses involved. Authors MG and LBI both managed the literature searches. All authors read and approved the final manuscript.

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ABSTRACT

In this paper, laboratory pots experiment was conducted to assessing the absorption and accumulation potential of the native grass species, Cyperus rotundus. Viable seed of the grass were seeded into soil (2.0 kg) amended with different concentrations of the metals, Zn, Pb, Cd and Ni. Experimental soil was amended with, 150, 1000, and 3000 ppm Zn as $Zn(SO_4)_3.6H_2O$; 150, 500, and 1000 ppm Pb as $Pb(NO_3)_2$; 150, 250, and 400 ppm Cd as $Cd(NO_3)_2$; 150, 500, and 1000ppm Ni as $Ni(NO_3)_2.6H_2O$. 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 analysed 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

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concentration in the soil, root and shoot. The results showed that, Zn had BCF values of 1.38, 6.13, 5.67, and 1.93; EF of 1.46, 1.65, 3.31, and 1.55; TF of 0.27, 0.59, and 0.80 at the control, 150, 1000, and 3000 ppm Zn in the soil respectively. Lead has BCF values of 1.31, 1.87, 0.92, and 1.66; EF of 0.62, 0.88, 0.23, and 1.18; TF of 0.47, 0.47, 0.25, and 0.71 at the control, 150, 500, and 1000 ppm Pb in the soil respectively. Cadmium has BCF values of 1.01, 1.00, 1.00, and 0.87; EF of 0.89, 0.87, 1.17, and 1.19 at the control, 150, 250, and 400 ppm Cd in the soil respectively. Nickel had the BCF values of, 14.0, 2.8, 2.3, and 1.4; EF of 18.0, 3.9, 5.4, and 3.6, TF of 1.29, 1.40, 2.30, and 2.57 at the control, 150, 500, and 1000 ppm Ni in the soil respectively. High values of one (1) and above for the BCF and EF indicates absorption, high retention and concentrating of the metals in the roots with less translocation to the shoots. The grass, Cyperus rotundus may, therefore, be best described as a metal excluder or stabiliser for Zn and Ni having greater values of BCF and EF than TF. A phenomenon is known as phytostabilisation. It could also absorb and retain the metal, Pb in the roots having BCF of 1.66, EF= 1.18 at 400 ppm Pb.

Keywords: Plants; accumulation; hyperaccumulation; phytoextraction; phytostabilisation; soil.

1. INTRODUCTION

The quality of life on Earth is linked inextricably to the overall quality of the environment. It is very difficult to define soil quality, as soil composition can vary from place to place. Soil quality is concerned with more than the soil's constituents and composition, but how it functions in a specific environment [1]. There has been an increasing concern with regard to the accumulation of toxic heavy metals in the environment and their impact on both public health and the natural environment [2]. The accumulation of heavy metals in soil is becoming a serious problem as a result of industrial and agricultural practices to name but a few of the causes of pollution today. Fertilisers from sewage sludge, mining waste and paper mills all contribute to the continuous deposition of heavy metals into soils. Another point of concern is the effect of leaching on these contaminated sites which in turn contaminate water tables [3].

Phytoremediation is described as a natural process carried out by plants and trees in the cleaning up and stabilisation of contaminated soils and ground water. It is actually a generic term for several ways in which plants can be used for these purposes. It is characterised by the use of vegetative species for *in situ* treatment of land areas polluted by a variety of hazardous substances [4]. Garbisu [5] defined phytoremediation as an emerging cost effective, non-intrusive, aesthetically pleasing, and low-cost technology using the remarkable ability of plants to metabolise various elements and compounds from the environment in their tissues. Phytoremediation technology is applicable to a broad range of contaminants, including metals and radionuclides, as well as organic compounds like chlorinated solvents, polychlorobiphenyls, polycyclic aromatic hydrocarbons, pesticides/insecticides, explosives and surfactants. According to Macek et al. [6], phytoremediation is the direct use of green plants to degrade, contain, or render harmless various environmental contaminants, including recalcitrant organic compounds or heavy metals. Plants are especially useful in the process of bioremediation because they prevent erosion and leaching that can spread the toxic substances to surrounding areas [7]. Phytoremediation is an integrated multidisciplinary approach to the cleanup of contaminated soils, which combines the disciplines of plant physiology, soil chemistry, and soil microbiology [8]. The different techniques involved can be summaries as in Fig. 1 [9].

Plants are capable of using metals through different ways such as complexing them in their sedentary nature, binding them into cell wall, and/or combining them to produce certain organic acid or proteins [10] Therefore, plant species are considered as good bioindicators in the early stages of heavy metal pollution. Additionally, they can be used for monitoring the state of the aquatic ecosystem and the changes or alterations in the aquatic environments [11] High heavy metal content in soil, water, sediments, and/or the air is found to be the most common stress factor which is faced by plant species. Therefore, it is imperative that plant species must adapt to different environmental conditions in order to survive. According to their adaptation strategies and heavy metals content, plant species can be classified into three main groups: metal excluders, indicators, and accumulators or hyperaccumulators [12].

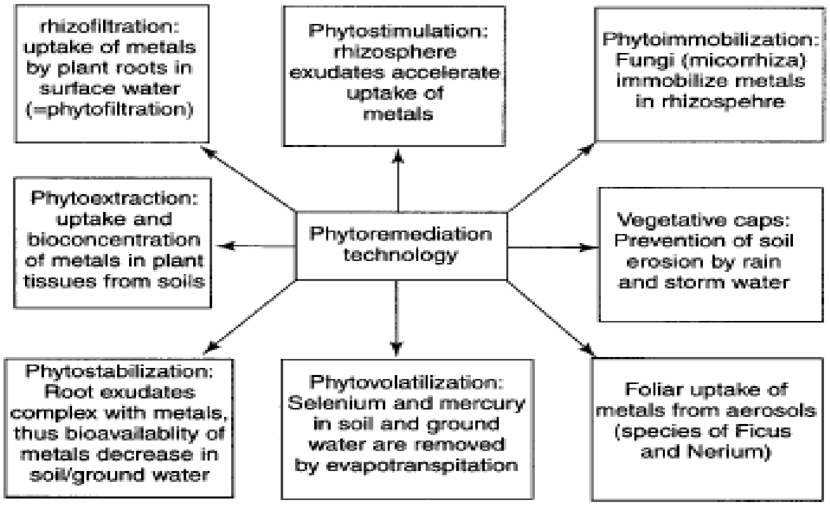

Fig. 1. The different techniques of phytoremediation. Source [9]

Hyperaccumulator plants are widely used in phytoremediation. This is due to the fact that these plants can contain Pb, Cu, Co, Cr, and Ni >1000 μg/g or 10.000 μg/g of Fe, Mn, and Zn or Cd >50 μg/g in any aboveground tissue in their natural habitat without suffering toxic effects [12, 13]. Metal excluders can be defined as plants that can restrict translocation of heavy metals from their roots into their aboveground tissues. These species can maintain relatively low levels of metal concentrations in their shoots as compared with the elevated metals concentrations in their roots [10]. Indicator plants are plants which have the ability to accumulate the metals in their aboveground tissues; thus, the metals levels in the tissues reflect the metal levels in the soil [14,15]. However, this type of plants dies off under continued uptake of heavy metals. Determination of the hyperaccumulator, indicator, and excluder plant species is dependent on several criteria. A plant species can be considered as a hyperaccumulator for heavy metals if it meets one of the following four strict criteria: (1) the ratio of heavy metal concentrations of shoot to root must be greater than 1 (metal concentration in shoot/metal concentration in Root) ≥1 [14]; (2) (metal concentration in root/metal concentration in sediments or soil) > 1 [15,16]; (3) the hyperaccumulator plant must be 10–500 times greater than the same species growing in noncontaminated sites [14, 17] and (4) plants with Pb, Cu, Co, Cr, and Ni>1000 μg/g or 10.000 μg/g of Fe, Mn, and Zn or Cd >50 μg/g in any aboveground tissue in their natural habitat without suffering toxic effects can be classified as hyperaccumulator plants [14,17]. According to Mganga et al. [14], “a plant which has high levels of heavy metals in the roots but with shoot/root quotients less than one (1) is classified as a heavy metal excluder.” This research work is aimed at assessing the phytoremediation ability of the native grass species, Purple nutsedge grass otherwise called Cocograss, (*Cyperus rotundus*) for the heavy metals; Zinc (Zn), cadmium (Cd), nickel (Ni) and lead (Pb) through their bioaccumulation, enrichment and translocation factors.

2. MATERIALS AND METHODS

2.1 Sample Collection

Viable seeds of the grass, *Cyperus rotundus* were collected from the plants dried husks. The soil that supported the growth of the grass was equally collected from the surface to subsurface portions, just beneath the roots of the grass (0-30cm depth). Samples were collected from Lake Chad Research Institute situated at Km 5 Gamboru Ngala Road Maiduguri, Borno State, Nigeria.

2.1.1 Laboratory pots experiment

Pot culture experiment was conducted using 2 kg soil spiked with the soluble salt of the metals Zn, Cd, Ni, and Pb. Experimental soil was spiked with the salt of Zn as $Zn(SO_4)_3.6H_2O$, Ni as $Ni(NO_3)_2.6H_2O$, Pb as $Pb(NO_3)_2$ and Cd as $Cd(NO_3)$ at a concentration of 150 ppm, 250 pmm, 400 ppm for Cd; 250, 1000 and 3000 ppm for Zn whereas 150, 500, and 1000 ppm was for Pb and Ni respectively. Viable seeds of the grass, Coco grass, were sawn into the pots. Separate pots containing the same amount of untreated soil (2 kg) was equally seeded to serve as a control [18]. 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. Plastics trays were place under each pot and the leached was collected and put back in their respective pots in other to prevent loss of nutrients and trace element from the samples [18]. The grasses were allowed to grow for a period of eight weeks. Four replicates of experimental pots for each element was seeded for statistical handlings.

2.1.1.1 Sample preparation and analysis

Set samples of the grass were harvested at the end of the pot experiment. Grass samples were carefully washed and separated into roots and shoots, dried at room temperature, ground separately and digested using 6M HCl according to Radojevic and baskin, [19]. The soil samples were equally collected from the different experimental pots including that of the control, ground, sieved and digested using concentrated HNO_3, H_2SO_4, and $HClO_4$ acid in a ratio of 5:1:1, [20]. Analysis of the digested samples for the metals: Zn, Cd, Pb and Ni was carried out using Atomic Absorption Spectroscopy, model PG 990.

3. STATISTICAL DATA HANDLING

All statistical data handling was performed using SPSS 12 package. The difference in mean of heavy metal concentration among the different samples was detected using one-way ANOVA, followed by multiple comparisons using Tukey test. A significant level of ($P = 0.05$) was considered throughout the analysis.

4. RESULTS AND DISCUSSION

4.1 Physicochemical Properties of the Experimental Soil

The physicochemical properties of the experimental soil are as shown in Table 1. The taxonomy classification of the soil was found to be sandy loam with pH of (6.27). The less acidic nature of the soil is generally within the range for soil in the region; soil pH plays an important role in the sorption of heavy metals, it controls the solubility and hydrolysis of metal hydroxide, carbonate and phosphates [21]. A very low organic carbon was observed in the soil sample (0.53) which led to the low organic matter content observed (0.90) as well as low cation exchange capacity (CEC) (4.09 mol/100kg soil). CEC measure the ability of soil to allow for easy exchange of cations between it surface and soil. The low level of clay and CEC indicate the permeability and leachability of metals in the soil. Appreciable amount of silt was observed in the soil sample (20.70), silt improves the soil, resulting in better plant growth.

Presently, phytoremediation has become an effective and affordable technological solution used to extract or remove inactive metals and metal pollutants from contaminated soil. Phytoremediation is the use of plants to clean up a contamination from soils, sediments, and water. This technology is environmentally friendly and potentially cost effective. Plants with exceptional metal-accumulating capacity are known as hyperaccumulator plants [22]. Phytoremediation takes the advantage of the unique and selective uptake capabilities of plant root systems, together with the translocation, bioaccumulation, and contaminant degradation abilities of the entire plant body [23]. Many species of plants have been successful in absorbing contaminants such as lead, cadmium, chromium, arsenic, and various radionuclides from soils.

4.2 Uptake and Translocation of the Metals; Zn, Cd, Ni and Pb by *C. rotundus*

4.2.1 Zinc (Zn)

Table four showed the variation in the level of Zinc in the parts of the plant, C. rotundus, grown in the experimental pot spiked with the levels; 150, 1000 and 3000 ppm Zn. The uptake and translocation of the element was found to increase as the level in the experiment pots increases. For instance, the level of Zn in the root of the control experiment was observed to be 315±0.006 ppm whereas 459±0.002 ppm was observed in the shoot. When the soil was spiked with 150 ppm Zn, the level observed in the root and shoot was found to increased. At 3000 ppm Zn in the experimental pot, the concentration in the root was 3060 ppm Zn, and the amount translocated to the shoots was observed to 2459 ppm Zn. At these level (1000 and 3000 ppm) of the element in the pot, the concentrations observed in the roots were found to be higher than what was translocated to the shoot (Table 2).

The accumulation and translocation of Zinc by C. rotundus, showed that, uptake of zinc at concentration of 150 ppm, 1000 ppm and 3000 ppm in the experimental pots were found in the root but with maximum amount translocated to the shoot at higher level than the root in the plant C. rotundus (Table 2). Report has it that, Zn transport in plants takes place through both the xylem and the phloem. Following absorption by the root, Zn is rapidly transported via the xylem to the shoot [24]. In rice plant, adequate Zn supply leads to a high proportion of Zn located in the shoots, while with toxic level of Zn supply (150 µmol/L), a higher proportion of total Zn may accumulate in the roots [25]. The efficiency of root-to-shoot translocation is theoretically dependent on four processes [26,27]: (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 [28]. It has been reported 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 [29].

4.2.1.1 Zinc toxicity on c. rotundus in the experimental pots

Despite the hiked in the concentration of Zn in the experimental pots, absorption by the plants in the pots, showed no phenotypical changes or sign of toxicity (Fig. 2) compared with the control experiment (Fig. 6). It has been envisaged that, the first symptom to present itself in most species exhibiting Zn toxicity is a general chlorosis of the younger leaves [30,31]. Depending on the degree of toxicity this chlorosis can progress to reddening due to anthocyanin production in younger leaves [32,33]. In this study, however, the control and the experimental plants were found to be normal throughout the experiment (Fig. 2 and 6). it has been reported that, plants exhibiting Zn toxicity have smaller leaves than control plants [30]. Glycine max plants normally have horizontally orientated unifoliate leaves. However, Zn stressed plants exhibit vertically oriented leaves [31]. Brown spots become apparent on the leaves of some species [31]. In severe cases plants may exhibit necrotic lesions on leaves and eventually entire leaf death [32]. In roots, Zn toxicity is apparent as a reduction in the growth of the main root, fewer and shorter lateral roots and a yellowing of roots [30]. Cyperus rotundus in this study, exhibit no sign of these symptoms as shown by both the experimental and the controlled test (Fig. 2 and 6).

4.3 Lead (Pb)

Table three below shows the distribution of the element Pb in the parts of Cyperus rotundus both in the control as well as the experimental pots spiked with different levels of Pb (150, 500, and 1000ppm). The results indicated that, most of the metal absorbed are retained in the roots including the control. The experimental pot spiked with 1000 ppm Pb has the highest level in the root (643 ±0.004). Lead adsorption onto roots has been documented to occur in several plant species: Vigna unguiculata [34], Festuca rubra [35], Brassica juncea [36], Lactuca sativa [37], and Funaria hygrometrica [38]. Lead may enter the roots through several pathways, and a particular pathway is through ionic channels. Although lead uptake is a non-selective phenomenon, it nonetheless depends on the functioning of an H^+/ATPase pump to maintain a strong negative membrane potential in rhizoderm cells [39, 40]. Inhibition of lead absorption by calcium is well-known [41,42] and is associated with competition between these two cations for calcium channels [43].

For most plant species, the majority of absorbed lead (approximately 95% or more) is accumulated in the roots, and only a small fraction is translocated to aerial plant parts, as has been reported in Vicia faba, Pisum sativum, and Phaseolus vulgaris [44, 45], V. unguiculate [34, Nicotiana tabacum, [46] Lathyrus sativus [47], Zea mays [48] Avicennia marina [49], non-accumulating Sedum alfredii [50], and Allium sativum [51]. This agrees with the results of this study (Table 4). However, these reasons are not sufficient to explain the low rate of lead translocation from root to shoot. Reports has it that, the endoderm, which acts as a physical barrier, plays an important role in this phenomenon. Indeed, following apoplastic transport, lead is blocked in the endodermis by the Casparian strip and must follow symplastic transport [52].

Table 1. The physicochemical properties of the experimental soil

Parameters	Soil
pH	6.27 ±0.004
EC (dsm^{-1})	0.38 ±0.006
CEC (mol/100kg soil)	4.09 ±0.007
Organic Carbon (%)	0.53 ±0.005
Organic matter content (%)	0.91 ±0.005
Silt (%)	20.70 ±0.006
Clay (%)	14.70 ±0.005
Sand (%)	64.60 ± 0.003
Textural Class	Sandy loam

Data are presented in mean and ± standard deviation (SD) with n = 3

4.3.1 Lead Toxicity on the Growth of *C. rotundus* in the Experimental Pots

High concentration of Pb accumulated by *C. rotundus* at 150 ppm, 500 ppm and 1000 ppm Pb were found in the root, with no noticeable symptoms of toxicity observed in the germination and growth of the plants, *Cyperus rotundus* (test), compared with the control experiment (Fig. 3 and 6). Although at 150 ppm Zn, a sign of poor growth was observed (Fig. 3), this effect did not however, show at higher level of the element (500 and 1000 ppm Pb). Report has it that, when plants are exposed to lead, even at micromolar levels, adverse effects on germination and growth can occur [34].

Table 2. Concentration (ppm) of Zn in the Soil, Shoot and Root of *C. rotundus* and itsTranslocation (TF), Enrichment (EF) and Bioconcentration Factor (BCF)

Amount spiked	Soil	Root	Shoot	BCF	TF	EF
150	622 ± 0.005	3811 ±0.003	1024 ±0.007	6.13	0.27	1.65
1000	348 ±0.003	1972 ±0.009	1153 ±0.006	5.67	0.59	3.31
3000	1590 ±0.013	3060 ±0.025	2459 ±0.017	1.93	0.80	1.55
Control	315 ±0.007	435 ±0.006	459 ±0.002	1.38	1.55	1.46

Data are presented in mean and ± Standard Deviation (SD), means were found not significant at P = .05 using one-way anova and multiple comparison according to Tukey test

Fig. 2. The plants, *C. Rotundus,* in the experimental pots spiked with different levels of Zn

Table 3. Concentration (ppm) of Pb in the soil, shoot and root of *C. rotundus* and its Translocation (TF), Enrichment (EF) and Bioconcentration Factor (BCF)

Amount spiked	Soil	Root	Shoot	BCF	TF	EF
150	317 ± 0.003	592 ± 0.003	278 ±0.007	1.87	0.47	0.88
500	639 ±0.004	588 ± 0.008	145 ± 0.003	0.92	0.25	0.23
1000	387 ± 0.004	643 ± 0.004	455 ± 0.004	1.66	0.71	1.18
Control	256 ± 0.007	335 ± 0.006	159 ± 0.002	1.31	0.47	0.62

Data are presented in mean and ± Standard Deviation (SD), means were found not significant at P = .05 using one- way anova and multiple comparison according to Tukey test

4.4 Cadmium (Cd)

The uptake and distribution of the element cadmium in the root and shoot of the grass, C. rotundus, is as shown in table 4. The results showed that, higher level of Cd was retained in the root than the shoot of the control and the experimental pot at 150 ppm spiked Cd. For instance, at a normal level, the control, the root had 281±0.008 ppm Cd, when the level in the soil was increased to 150 ppm, the uptake was found to equally increased (393±0.001) in the root. Generally, it has been suggested that, the important uptake route of elements in plants are the roots, and it is expected that roots will have a higher uptake as compared to the shoot [53]. A heavy metal ATPase was suggested to be involved in Cd accumulation in vacuoles of root cells causing Cd retention in roots and decreasing the transport to the shoot [54].

However, at 250 and 400 ppm Cd in the experimental pots, the uptake and translocation trend changes. At 250 ppm, the root had 386±0.004 ppm whereas the shoot had 432±0.002 ppm Cd. At 400 ppm Cd in the pot, translocation rate to the shoot increases (Table 4). It has been reported that, the accumulation of Cd in the shoots of an emergent plant is generally dependent on the roots as its primary source [55]. Translocation of Cd from root to shoot has been studied in several species, including ryegrass Secale cereal, [56] tomato (*Lycopersicon esculentum* [57], bean, Phaseolus vulgaris, [58], maize [59], and durum wheat [60]. Movement of Cd from roots to shoots is likely to occur via the xylem and to be driven to the shoot by transpiration pull from the leaves. Evidence for this was provided by Salt et al. [61], who showed that ABA-induced stomatal closure dramatically reduced Cd accumulation in shoots of Indian mustard. High level of Cd observed in the shoot of C. rotundus of this study therefore could be attributed to the transpiration pull by leaves. This observation agrees with the report of Hartel et al. [62], who observed higher shoot Cd accumulation in bread wheat cultivar. It has been reported that, Cd not only prefers to form bonds with sulphydryl ligand groups, but also binds to N and O ligand groups. Thus, cysteine and other sulphydryl-containing compounds (phytochelatins, glutathione etc.) and various organic acids (citrate) and other amino acids in xylem sap could be important in transporting Cd from roots to shoots [63].

4.4.1 Cadmium toxicity on the growth of *C. rotundus* in the experimental pots

Although no sign of toxicity of Cd on the plants (Fig. 4) was observed, reduction in growth has been associated with cadmium treatment which was reported to caused inhibition of protein synthesis [64]. A similar observation was made on the grass at 150 ppm Cd in the pot (Fig. 4). The presence of Cd decreased the content of chlorophyll and carotenoids and increased non-photochemical quenching in *Brassica napus* [65]. Similarly, the synthesis and level of chlorophyll decreased in other plant species under the influence of the cadmium [66,67,68].

4.5 Nickel (Ni)

Table five below present the result for the uptake, translocation and accumulation of the metal, Ni, in the roots and shoots of the grass C. rotundus. Most of the metal absorbed were translocated and retained in the shoot. For instance, when the experimental pot was spiked with 250 ppm Ni, the level in the root was 160.0 ppm ± and the shoot had 375.0 ppm. The same trend of accumulation in the shoot was observed when the level of the metal in the experimental pot was increased to 400 ppm, the root has 173 ppm Ni whereas the shoot had 445.5 ppm Ni. It has been extensively reported that, a higher concentration of Ni is found in the above-ground parts (the shoot) of plants rather than in the roots [69,70,71]. Report has it that, because of the presence of carrier for the transport of Ni in plants, heavy metal get absorbed from the soil easily, crosses the cell wall and plasma membrane of the root and through xylem gets accumulated in the leaf vacuole [72]. Nevertheless, different Ni distribution patterns were observed in other plant species. For example, Marques et al. [73] reported that in Rubus ulmifolius, Ni was only distributed in the root. Uptake of Ni by plants depends upon various factors, the most important of course, being the ionic, Ni concentration in the medium [74,75]. Soil pH values below 5.6 seem to favour the absorption of Ni and is largely due to the fact that the exchangeable Ni content of the soil increases with the increasing soil acidity [76].

4.5.1 Nickel toxicity on the growth of *C. rotundus* in the experimental pots.

In this study, absorption of Ni when its concentration in the soil was amended with; 150, 500 and 1000 ppm Ni showed no sign of toxicity effect on the plant (Fig. 5) compare with the control experiment (Fig. 6). However, physiological role of nickel and its toxic effects on higher plants has been reported [77], the phytotoxic effects of the metal have also been observed [78]. Growth of most plants species is adversely affected by tissue concentration above 50 µgg^{-1} dry weight. Report has it that, it is toxic at elevated concentration in plant [79]. High concentration of nickel inevitably binds organic macromolecules and denatures them. Furthermore, nickel can replace iron, zinc and magnesium due to the chemical affinity with those elements, interfering with their metabolism [80]. High Ni concentrations retard shoot and root growth, affect branching development, deform various plant parts, produce abnormal flower shape, decrease biomass production, induce leaf spotting, disturb mitotic root tips, and produce Fe deficiency that leads to chlorosis and foliar necrosis. Additionally, excess Ni also affects nutrient absorption by roots, impairs plant metabolism, inhibits photosynthesis and transpiration, and causes ultrastructural modifications [81].

Fig. 3. The Growth of C. rotundus, in the experimental pots spiked with different level of Pb

Fig. 4. The growth of *C.* r*otundus,* in the experimental pots spiked with different levels of Cd

Table 4. Concentration (ppm) of Cd in the Soil, Shoot and Root of *C.* r*otundus* and its Translocation (TF), Enrichment (EF) and Bioconcentration Factor (BCF)

Amount spiked	Soil	Root	Shoot	BCF	EF	TF
150	389 ± 0.005	393 ±0.001	339 ± 0.004	1.00	0.86	0.87
250	369 ± 0.008	386 ± 0.004	432 ± 0.002	1.00	1.17	1.12
400	375 ± 0.005	328 ±0.003	391 ± 0.002	0.87	1.04	1.19
Control	289 ±0.003	291 ± 0.008	258 ± 0.002	1.01	0.89	0.89

Data are presented in mean and ± Standard Deviation (SD), means were found not significant at P = .05 using one-way anova and multiple comparison according to Tukey test

Table 5. Concentration (ppm) of Ni in the soil, shoot and root of *C. rotundus* and its translocation (TF), enrichment (EF) and bioconcentration Factor (BCF)

Amount spiked	Soil	Root	Shoot	BCF	EF	TF
150	13.00 ±1.000	36.00 ±1.000	51.00 ±0.005	2.80	3.90	1.40
500	69.00 ± 0.001	160.00 ±0.005	375.0 ±0.028	2.30	5.40	2.30
1000	124.0 ± 0.001	173.00 ±0.003	445.5 ±0.014	1.40	3.60	2.57
Control	0.500 ± 0.001	7.000 ± 1.00	9.000 ±0.002	14.00	18.00	1.29

Data are presented in mean and ± Standard Deviation (SD), means were found not significant at P = .05 using one-way anova and multiple comparison according to Tukey test

Fig. 5. The Growth of *C. rotundus* in the experimental pots spiked with different levels of Ni

Fig. 6. The growth of the plant, *C. rotundus* in the control experimental pot

5. PHYTOREMEDIATION EFFICIENCY OF THE GRASS *C. ROTUNDUS*

In most of the established criteria of identifying the metal accumulation in plants, it is imperative to consider the metal concentrations in the aboveground biomass and the metal concentrations in the sediments or soil [13]. According to Usman and Mohammed [82], the success of phytoextraction process depends on heavy metal removal by the shoot 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 shoot tissues rather than in their root 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.

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

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 [83].

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

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. [16].

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

In this study, the BCF, EF and TF values for the metals; Zn, Cd, Ni and Pb are presented in tables, 2, 3, 4, and 5 respectively at different level of metal in the experimental pots. For Zn, the BCF, TF, and EF values at 150, 1000, and 3000ppm are; BCF = 6.13; 5.67; 1.93; and 1.38 for the control respectively; TF = 0.27, 0.59, 0.80, and 1.55 for the control respectively; EF = 1.65, 3.31, 1.55, and 1.46 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 [84]. 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 *Cyperus rotundus* may be suggested for phytostabilisation 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 phytostabilisation [83]. Only the control has the ability to translocate the element Zn from the root to the shoot, a property that qualifies *C. rotundus* for phytoextraction, though it will take time for success to be achieved. This may be suggested that, at high concentration of Zn, *C. rotundus* can stabilise the soil by retaining Zn in the root at a concentration higher than the shoot.

The BCF, TF, and EF values for Pb at 150, 500, and 1000ppm are; 1.87, 0.92, 1.66; and 1.31 for the control; TF = 0.47, 0.25, 0.71; and 0.47 for the control; EF = 0.88, 0.23, 1.18; and 0.62 for the control (Table 3). A BCF value greater than one (1), signifies a high degree of intake and storage. For having the BCF and EF values greater than one (1) at 1000ppm Pb and TF value less than one (1) *Cyperus rotundus* may have the potentials to stabilised Pb by absorbing and retaining high concentration of the element in the root than shoot. A process known as phytostabilisation. Plants exhibiting TF and particularly BCF values less than one are unsuitable for phytoextraction [85].

The BCF, TF, and EF values for Cd at 150, 250, and 400ppm are; 1.00, 1.00, 0.87; and 1.01 for the control; TF = 0.86, 1.12, 1.19 and 0.89 for the control; whereas the EF = 0.87, 1.17, 1.04; and 0.89 for the control (Table 4). For Cd, *C. rotundus*, has high degree of uptake and translocation because Cd has BCF of 1, EF of 1.17 and TF of 1.12. Such plants that has BCF, EF, and TF equal to one or above may be described as potential metal indicators. These are plants which have the ability to accumulate the metals in their roots and translocating simultaneously to the aboveground tissues; thus, the metals levels in the tissues reflect the metal levels in the soil [14, 15]. However, this type of plants dies off under continued uptake of heavy metals.

Nickel has BCF, EF, and TF values separately as; BCF = 2.80, 2.30, 1.40; and 14.00 for the control, 150, 500, and 1000 ppm Ni respectively; the EF values are; 3.90, 5.40, 3.60; and 18.00 for the control, 150, 500, and 1000 ppm Ni respectively, whereas the TF values are, 1.40, 2.30, 2.50 and 1.29 for the control, 150, 500, and 1000 ppm Ni respectively. McGrath and Zhao, [15] and Sun et al. [86] classify the bioconcentration factor as a parameter for classification as a hyperaccumulator species. The researchers; [87,88,82,83] have pointed out that the ability of phytoremediation has commonly been characterised by a translocation factors (TF). Translocation factor (TF) greater than one (> 1) that translocation of metals effectively was made to the shoot from root [12, 89, 90]. Plants with both bioaccumulation and translocation factors greater than one (BCF and TF>1) have the potential to be used in phytoextraction. In the contrary, nickel has higher BCF and EF values than TF although the TF equally has value greater than 1, the BCF and EF values are 2-3 folds greater. This means, the quantities of trace elements accumulated in the root tissues exceeded those in the shoot tissues. Gupta et al. [91] 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, *C. rotundus* may best be described as Ni stabiliser in the soil (a process known as Phytostabilisation). This process uses the ability of plant roots to change environmental conditions via root exudates. Plants can immobilise heavy metals through absorption and accumulation by roots, adsorption onto roots, or precipitation within rhizosphere [92]. By using metal-tolerant plant species for stabilising contaminants in soil, particularly metals, it could also provide improved conditions for natural attenuation or stabilisation of contaminants in the soil. Metals accumulated in the roots are considered relatively stable as far as release to environment is concerned.

6. CONCLUSION

The phytoremediation potential of the grass; *Cyperus rotundus*, assessed in this study showed that, the grass may successfully be used as a phytostabiliser for the metals; Zn, Pb, Ni. This process reduces metal mobility and leaching into ground water, and also reduces metal bioavailability for entry into the food chain. One advantage of this strategy over phytoextraction is that the disposal of the metal-laden plant material is not required. The grass may also serve as a metal indicator for the Cd. 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.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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