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Active biomonitoring of heavy metals uptake by some native plants, grown in Southwestern, Sinai, Egypt

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ABSTRACT

Heavy metal pollution is one of the most important environmental problems today. Phytoremediation is an effective and low-cost interesting technology. The aim of this study is to compare the accumulation of metals in the investigated native plants, with metal concentrations in roots and in soils, and to assess the feasibility to use these plants for phytoremediation purpose as well as its use as bioindicators for heavy metals in different soils. Four plants species (*Hyoscyamus muticus*, *Citrullus colonythisis*, *Artemisia judaica* and *Cleome droserifolia*.) were collected from Abu Zeneima area; in Southwestern Sinai with their associated soils. Concentrations of heavy metals were determined in both soil and plants using atomic adsorption. All plant species can uptake heavy metals and aluminum but with different concentration. *Hyoscyamus muticus* was effective in taking up Ni while *Citrullus colonythisis* was effective in taking up Cu and Mn.

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KEYWORDS

Trace elements;
Wild plants;
Hyoscyamus;
Citrullus;
Artemisia;
Cleome.

INTRODUCTION

Accumulation of heavy metals in the environment may cause chronic damage to living organisms and must be carefully controlled. They are harmful to humans, animals and tend to bio-accumulate in the food chain. The threat the heavy metals pose to human and animal health is aggravated by their long-term persistence in the environment. Several technologies are available to remediate soils heavy metals contaminated that. However, many of these technologies are costly (e.g. excavation of contaminated material and chemical/physical treatment) or do not achieve a long-term nor aesthetic solution^[5,16].

Phyto-remediation can provide a cost-effective and long-lasting solution for remediation of contaminated sites^[13]. Phytoremediation is an emerging technology that employ the use of higher plants for the cleanup of contaminated environments.

Fundamental and applied research have unequivocally demonstrated that selected, plant species possess the genetic potential to remove, degrade, metabolize, or immobilize a wide range of contaminants .i.e. through uptake and accumulation of metals into plant shoots which can then be harvested and removed from the site. phytoremediation depends on the interaction among soil, contaminants, microbes, and plants. This complex interaction, affected by a variety of factors, such as cli-

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matic conditions, soil properties, and site hydrogeology, argues against generalization, a

It is important to use native plants for phyto-remediation because these plants are often better in terms of survival, growth and reproduction under environmental stress than plants introduced from other environment. A few studies have evaluated the phyto-remediation potential of native plants under field conditions^[14, 28]. Heavy elements like, Cu, Fe, Mn, Ni and Zn, are essential elements required for normal growth and metabolism of plants. These elements can easily lead to poison and severe phyto-toxicity when their concentration rises to supra-best values.

The phyto-toxicity of these metals may result from alterations of numerous physiological processes caused at cellular/molecular level by inactivating enzymes, blocking functional groups of metabolically important molecules, displacing or substituting for essential elements and disrupting membrane integrity. Another consequence of heavy metal poisoning is the enhanced production of reactive oxygen species (ROS) due to interference with electron transport activities, especially that of chloroplast membranes^[12, 20].

Biological responses can be considered more representative than data supplied by chemical or physical detectors, in that they are spatially and temporally extensive; lower or higher plants can act as bio-indicators, bio-monitors and bio-accumulators^[6, 25]. The aims of this study were: 1) to determine the concentrations of heavy metals (Iron, Zinc, Copper, Nickel, Manganese) and Aluminum in plant biomass growing on a contaminated site; 2) to compare metal concentrations in the aboveground biomass to those in roots and in soils, and 3) to assess the feasibility to use these plants for phyto-remediation purpose. Information obtained from this study should provide insight for using native plants to remediate metal contaminated.

MATERIALS AND METHODS

Plant species and sampling locations

Plant samples, were collected, with their associated soil samples based on their coverage at the study area (Abu Zeneima area in southwestern Sinai). Soil samples from the rooting zone (0–30cm) were taken

from each location, air-dried for two weeks, and then sieved through 2 mm mesh. Samples were then analyzed for total metals (Al, Zn, Cu, Ni, Mn and Fe) using atomic adsorption. Soil pH was measured using a 1:2 soil to water ratio. Selected characteristics of the soil samples collected from this study are shown in TABLE (1).

Plant sampling and analysis

Three replicates of different plant species were collected from the studied area. These plants were *Hyoscyamus muticus*, *Citrullus colonythisis*, *Artemisia judaica* and *Cleome droserifolia*. Each replicate consisted of sub samples collected from three different points. Plant samples were thoroughly washed with running tap water and rinsed with deionised water to remove any soil/sediment particles attached to the plant surfaces. The aboveground and underground tissues were then separated and air dried to constant weight. The dried tissues were weighed and ground into powder for metal concentration analysis. Metal analysis (Al, Zn, Cu, Ni, Mn and Fe) of the plant samples was carried out by acid digestion [conc. HNO₃ + conc. HClO₄ (4:1, v/v)] followed by measurement of total concentrations of all elements of interest using atomic adsorption.

RESULTS

Soil properties and metal concentrations

Previous research has shown that these soils has relatively high organic matter (5.1%)^[15, 18]. Selected properties of the 12 collected soil samples are listed in TABLE (1). The distributions of heavy elements in sedimentary rocks (soils) were mainly controlled by ionic size, charge and bond character but with different influences. Other factors controlling the distribution of trace elements are precipitation following by oxidation or reduction beside various reactions with organic matter

The investigated soils characterized by high level of various heavy metals, all exceeded the relevant local soil background values. Total iron concentrations in the soil samples collected from the sites rang from 980 at site 12 to 21546 mg kg⁻¹ at site 1 (TABLE 1), Many trace elements like Cu, Co, Ni, and Zn can be incorpo-

rated together with Fe^{3+} , and Al^{3+} , in hydrated phosphates (Mikula and Indeka, 1997 and Xiulan et al, 1999). This case is observed in ferrasol and sandy soils of Abu zeniema area. It reflects the high concentration of iron and aluminum in the investigated soil samples^[15]. The concentration of Cu, Ni and Zn ranges between (32- 650), (22- 1250) and (34- 922) mg Kg^{-1} respectively (TABLE 1). Their high concentration is controlled by the presence of iron as well as the presence of clay components^[15,18]. The manganese concentration in the soil samples range between 151 and 7689 mg/Kg .

TABLE 1 : Selected properties of soil samples from Abu Zienema area

Site	Location	Soil pH	Total Fe (mg kg^{-1})	Total Cu (mg kg^{-1})	Total Ni (mg kg^{-1})	Total Al (mg kg^{-1})	Total Zn (mg kg^{-1})	Total Mn (mg kg^{-1})
1	W. Shalal	6.5	21546	42	25	18225	34	7689
2	W. Sahw	6.8	19888	32	29.6	19380	42	799
3	W. Nasib	6.3	22145	57	15.3	16258	48	815
4	W. Sahw	5.7	15234	320	45	8254	922	33.6
5	W. Shalal	5.9	4056	18	77	12354	340	46.8
6	W. Hamata	6.9	2546	17	83	14050	450	25.8
7	W. Ba' ba	6.9	12457	61	64	7658	71	425
8	W. Sidri	7.4	11546	84	139	8574	86	288
9	W. Shalal	7.8	14256	40	64	5687	59	322
10	W. Hamata	7.6	8254	300	48	11234	280	425
11	W. Sahw	6.3	1254	280	31	10800	340	312
12	W. Sidri	5.8	980	144	22	8954	210	238

Metal concentrations in plants

Plants can accumulate trace elements especially heavy metals in their tissue due to their great ability to adapt many variable chemical properties of the environment. Plants are intermediates reservoirs where trace elements move from soils to man and animals. Plant may be passive reporter for trace elements (fallout interaction or root) but they also exert, control uptake or rejection of some elements but appreciate physiological reaction^[29]. The transfer of heavy elements within the soil-plant chain is a part of the biochemical cycling of elements. Several factors control the processes of mobility and availability of elements like pH, organic matter and redox conditions; between these pH is considered to be the most important and easily manageable one^[8]. Heavy metals occur in different species ac-

ording to whether they are external or internal bound to various soil components or in the liquid phase.

The ability of a plant to hyperaccumulate any metal may enhance some ability to accumulate other metals^[4, 23]. Some metals may interact competitively for accumulation (e.g., Zn and Ni in calamine and serpentine soils)^[4]. Suggested common mechanisms of absorption and transport of several metals by *Thlaspi* species. They observed high uptake by the roots for all metals studied. Zn, Cd, Co, Mn and Ni were readily transported to the shoot, whereas, Al, Cr, Cu, iron (Fe) and Pb were predominantly immobilized in the roots.

In this study a total of 12 plant samples of 4 plant species were collected from 12 locations in the investigated area. Concentrations of Fe, Cu, Zn, Mn, Ni and Al in soil and plant biomass were provided in TABLES 2, 3, 4, 5, 6 and 7. Total copper concentration ranges from 10.58 to 284.4 mg Kg^{-1} with the maximum concentration being in the root of *Citrullus colonthisis* from the site 4 (TABLE 2). The obtained result for *citrullus colonthisis* is in contradict with that obtained by^[24] who found that the uptake of copper is low for the same plant. No plant species accumulate copper above 1000 mg Kg^{-1} . Also the root of *Artemisia judaica* contained significant amount concentration of copper 80 mg Kg^{-1} while *Hyosymus muticus* and *Cleome droserifolia* contain normal concentration from copper ranging from 10.58 to 21.4 mg Kg^{-1} which agreed to those found in the literature obtained by^[11] (1.1 to 33.1 mg Kg^{-1})^[21]. Reported that the range of Cu contents in 50 medici-

TABLE 2 : Copper concentrations in soil and plant samples (mg kg^{-1}).

Scientific name	Site	Roots	Shoots	Soil
	1	13.27	2.32	42
<i>H. muticus</i>	2	11.78	1.53	32
	3	10.58	0.59	57
	4	284.4	63.5	320
	5	19.29	3.3	18
<i>C. colonythisis</i>	6	19.37	2.8	17
	7	21.4	5.18	61
	8	20.6	5.19	84
<i>C. droserifolia</i>	9	15.82	4.9	40
	10	80	19.3	300
<i>A. judaica</i>	11	63.5	11.3	280
	12	59	8.72	144

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nally important leafy materials growing in India were 17.6–57.3 mg kg⁻¹.

Zinc is an essential element to all plants; the mean concentration in normal plants (aboveground tissue) is 66 mgKg⁻¹[19]. The physiological activities of the plant influence Zn absorption and the interactions with many elements like Fe, Mn and Cu also affect Zn uptake^[10]. The concentration of zinc in tulsi (*Ocimum sanctum*) and neem (*Azadirachta indica*) leaves, which are widely used in Indian Ayurvedic medicine, were 140 and 10 mg g⁻¹, respectively^[26]. The zinc concentration in the investigated plant species ranges from non detectable to 607.5 mg kg⁻¹ (TABLE 3). Like copper the maximum value was found in the root of *C. Colonythisis* from site 4. Also no plant species accumulated Zn above 1000 mgkg⁻¹. The root of *Artemisia judaica* contained a significant concentration of Zinc ranging from 70.24 to 188.5 mgkg⁻¹. Similar to copper Zn concentration were greater in the root than that found in the shoots.

TABLE 3 : Zinc concentrations in soil and plant samples (mg kg⁻¹)

Scientific name	Site	Roots	Shoots	Soil
<i>H. muticus</i>	1	-	-	34
	2	-	-	42
	3	-	-	48
	4	607.5	86.25	922
<i>C. colonythisis</i>	5	104.5	49	340
	6	112.5	59.25	450
	7	40.5	18.925	71
<i>C. droserifolia</i>	8	34.5	14.5	86
	9	24.25	8.75	59
	10	188.5	13.925	280
<i>A. judaica</i>	11	70.25	5.9	340
	12	99.25	29	210

The concentration of total manganese ranged from 32.5 to 1123.75mgkg⁻¹ (TABLE 4) where the highest concentration was found in the root of *Hyocymus muticus* species. Both *Citrullus colonthisis* and *Artemisia judaica* contain normal concentration from manganese which is nearly in the same range obtained by^[21] who reported that the range of Mn in their study to be 10.5–81.6 mg kg⁻¹. While *Cleome droserifolia* contain higher concentration from manganese which is higher than the value obtained by^[24]. In their study.

TABLE 4 : Manganese concentrations in soil and plant samples (mg kg⁻¹)

Scientific name	Site	Roots	Shoots	Soil
<i>H. muticus</i>	1	1123.75	450.235	7689
	2	402	160	799
	3	324	113.75	815
	4	46.75	15	33.6
<i>C. colonythisis</i>	5	59.75	18.5	46.8
	6	35.5	13.75	25.8
	7	443	140	425
<i>C. droserifolia</i>	8	81	33.25	288
	9	80.75	27.25	322
	10	59.75	20.3	425
<i>A. judaica</i>	11	27.25	10.4	312
	12	32.5	8.5	238

Hyosymus muticus contain higher concentration from manganese (1123.7 mgkg⁻¹) >1000 mgkg⁻¹.^[24] stated that the plant is selective in accumulating Mn.

The nickel concentration in the investigated plant ranges from undetectable to 53.75 mgKg⁻¹(TABLE 5). Both *Hyosymus muticus* and *Cleome droserifolia* posses the ability to uptake nickel. The concentration in *Hyosymus muticus* is higher than that in *Cleome droserifolia* species but, both plants are under 1000 mg Kg⁻¹. The concentration in the root is higher than that in shoot. The concentration of iron ranges from 67 to 2840 mg Kg⁻¹, where the highest concentration was found in the root of *Hyosymun muticus*. Both *Hyosymus muticus* and *Cleome droserifolia* show the ability to accumulate Fe in their root part with a concentration exceeds 1000 mg Kg⁻¹[26]. Reported that the concentration of Fe in tulsi (*Ocimumsanctum*) and neem (*Azadirachta indica*) leaves which are widely used in Indian Ayurvedic medicine were 129 and 355 mg g⁻¹, respectively. The suggested Fe requirement for animals range between 30 and 100 p.p.m. and the maximum tolerable level for cattle is suggested as 1000 ppm. (National Research Council^[17]).

Aluminum (Al) is not regarded as an essential nutrient, but low concentrations can sometimes increase plant growth or induce other desirable effects. The Aluminum concentration in roots ranges between 875 and 3825 mg Kg⁻¹. All investigated, plant species show the ability to uptake aluminum with concentration nearly to be >1000 mg Kg⁻¹. The highest concentration was

TABLE 5 : Nickel concentrations in soil and plant samples (mg kg⁻¹)

Scientific name	Site	Roots	Shoots	Soil
<i>H. muticus</i>	1	50.75	7.2	25
	2	43	5	29.6
	3	53.75	13	15.3
	4	-	-	45
<i>C. colonythisis</i>	5	-	-	77
	6	-	-	83
	7	16	4.75	64
<i>C. droserifolia</i>	8	11.85	2.03	139
	9	49	11.85	64
	10	-	-	48
<i>A. judaica</i>	11	-	-	31
	12	-	-	22

TABLE 6 : Iron concentrations in soil and plant samples (mg kg⁻¹).

Scientific name	Site	Roots	Shoots	Soil
<i>H. muticus</i>	1	2840	781.25	21546
	2	2540	575.75	19888
	3	2180	387.5	22145
	4	974.25	169.75	15234
<i>C. colonythisis</i>	5	287.5	15.3	4056
	6	342.75	22.5	2546
	7	2083.2	350.2	12457
<i>C. droserifolia</i>	8	1202.7	200.75	11546
	9	3140	335	14256
	10	423	44.3	8254
<i>A. judaica</i>	11	128	33.5	1254
	12	67	12	980

TABLE 7 : Aluminum concentrations in soil and plant samples (mg kg⁻¹)

Scientific name	Site	Roots	Shoots	Soil
<i>H. muticus</i>	1	3825	635	18225
	2	2825	675	19380
	3	2400	456	16258
	4	1325	875	8254
<i>C. colonythisis</i>	5	1500	462.5	12354
	6	2125	475	14050
	7	1255	200	7658
<i>C. droserifolia</i>	8	935	197.5	8574
	9	875	100	5687
	10	2340	475	11234
<i>A. judaica</i>	11	1600	225	10800
	12	1075	200	8954

found in the root of *H. muticus* while the lowest was found in *C. droserifolia* (TABLE 7). The concentration of root is higher than that in shoot as what observed in the previous elements.

Mechanisms for metal tolerance

Plant species that are naturally high in heavy metals develop a strategy to tolerate heavy metals by unrestricted absorption and, as a result, accumulate high concentrations from heavy metal in their tissue. The hyper-accumulation strategies requires some mechanism(s) to detoxify the metals. It is widely accepted that detoxification of metal ions within plant tissues must depend on chelation by appropriate ligands. The anionic species of organic acids, such as citrate, malate, and malonate, are commonly found in high concentrations in the leaves of *Alyssum* spp.^[22] has pointed out that these anions tend to be present constitutively in these plants in substantial amounts and cannot account for the metal-specificity or species variability of Ni hyper-accumulators^[2]. Suggested that the Ni hyper-accumulation trait in *Alyssum* was associated with the ability of the root system to produce substantial amounts of histidine as a Ni complexion legend

Accumulation and translocation of metals in plant

In this study none of the plant species showed metal concentration > 1000 mg Kg⁻¹ in the leaf part (TABLE 2, 3, 4, 5, 6 and 7) in other word none of them are hyper accumulator^[3], however the ability of these plants to tolerate and accumulate heavy metals may be useful tool for phyto-stabilization. Both bio-concentration factor (defined as the concentration of metal in the root to that in the leaf) and translocation factor (defined as concentration of metal in leaf to that in root) estimates a plant's potential for phyto-remediation purpose.

The process of phyto-extraction generally requires the translocation of heavy metals to the easily harvestable plant part. By comparing the BCF and TF we can compare the ability of different plant to take up metals from soil and translocating them to shoots, tolerant plant tend to restrict soil root and root- shoot transfer and therefore have much less accumulation in their part while hyper-accumulator plant take up and translocation metals into their aboveground part^[7]. Stated that when plant

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TABLE 8 : Average BACs and TFs for investigated plant species.

Scientific name	Bioaccumulation Factor					Translocation Factor						
	Cu	Zn	Ni	Mn	Fe	Al	Cu	Zn	Ni	Mn	Fe	Al
<i>H. muticus</i>	0.28	-	2.36	0.35	0.12	0.17	0.12	-	0.16	0.38	0.23	0.23
<i>C. colonythisis</i>	1.05	0.41	-	1.34	0.09	0.14	0.18	0.38	-	0.34	0.1	0.4
<i>C. droserifolia</i>	0.28	0.46	0.37	0.74	0.16	0.14	0.26	0.41	0.23	0.35	0.15	0.16
<i>A. judaica</i>	0.30	0.45	-	0.12	0.06	0.16	0.19	0.62	-	0.33	0.18	0.17

TABLE 9 : Normal and toxic concentrations for the screened heavy elements

HM	Non-toxic concentration (mg kg ⁻¹)	Toxic concentration (mg kg ⁻¹)	Hyperaccumulation limit (mg kg ⁻¹)
Cu	5–30 ^e	20–100 ^e	1000 ^{bl} .
Zn	27–150 ^e	100–400 ^e	10,000 ^{al} and ^{bl}
Ni	0.1–5 ^e	10–100 ^e	1000 ^f
Mn	30–300 ^e	400–1000 ^e	10,000 ^d
Fe	5–250 ^e	–	10,000 ^d
Al	3–6 ^e		

^aVasquez et al. (1992); ^bHenry (2000); ^cKabata-Pendias (2001); ^eÁlvarez et al. (2003); ^fWong (2003).

had low TF and BCF values, their ability to accumulate and translocate heavy metals is limited (TABLE 8).

among all investigated, plants *Hyosymus muticus* had the highest BCF greater than 1 for Ni (BCF + 2.36; TABLE 8), while the total concentration in the plant was < 1000 mg Kg⁻¹, *C. colonythisis* plant species have the highest BCFs for Cu and Mn which is higher than one where BCF are 1.05 and 1.34 respectively (TABLE 8). Although the concentration of the Mn in *C. colonythisis* is < 1000 (TABLE 4) but its BCF is higher than one which complies with (kabata and pendias 2001) who stated that the concentration of the substrate plays an important role in the value of the BCF.

The BCFs for the Fe, Al and Zn were less than one in all the plant species where the average BCF for these plant are 0.06-0.12, 0.14-0.17 and 0.41-0.46 respectively in spite of the concentration of Al was mostly > 1000 mg Kg⁻¹ in all plants as well as Fe in *Hyosymus muticus* and *C. droserifolia*. Similar to Mn the high soil concentration decrease the value of BCF for those plant.

Though none of the plant samples were metal hyper-accumulator some observation were noted. Based on the average BCFs of all plants, Ni (0.68) is the highest one followed by Mn (0.63), Cu (0.48), Zn (0.33), Al (0.15) and finally Fe (0.11). Regarding to the average

Tf's of all plant samples the plants were most efficient in trans-locating Zn and Mn with value of 0.35 followed by Mn (0.35), Al (0.24), Cu (0.19), Fe (0.17) and finally Ni (0.1). Among the screened elements, the plants growing in the investigated area were efficient of uptaking Ni, Mn and Zn while low translocation of Fe and Ni from their root to shoot^[31]. Discussed restriction of metal uptake by plant from contaminated soil and the presence of exclusion mechanism in such plant specie. In general all heavy metals occurred at elevated level in plant part collected from the site, normal and phyto-toxic concentration were reported in TABLE (9). These results may show that plant species growing on the contaminated site with heavy metals were tolerant of these metals. Restriction of upward movement from root into shoot can be considered as one of the tolerance mechanism^[31].

Although no metal hyper-accumulator were found, heavy metal tolerant species with high BCF and low TF can be use phyto-stabilization of contaminated site together with a vegetative cover. Example of such plant in this study includes *C. colonythisis* for Cu and Mn and *Hyosymus muticus* for Ni (TABLE 8). Phyto-stabilization minimizes migration of contaminant in soil (Susaral et al. 2002), this phenomena use the ability of plant root to change environmental condition via root exudates. Plant can immobilize heavy metal through absorption and accumulation by roots, adsorption onto roots or precipitation within rhizosphere. This technique reduces metals mobility and leaching onto ground water and reduces metal bioavailability for entry into food chain.

An important advantage of this technique is that the disposal of the metal-laden plant materials is not required (Suarla et al. 2002). Using metal tolerant plant species for stabilizing contamination soil may also improve condition for natural attenuation. Although metals accumulated in roots are considered relatively stable

as far as release to environment is concerned so many studies are needed to regard the turn over of nutritive roots and the potential release of metals from decomposing roots^[33].

CONCLUSION

This study was conducted to screen native plants growing on an arid area to determine their potential for metal accumulation. Only species with both BCFs and TFs greater than one have the potential to be used for phyto-extraction. Within all the investigated plant species screened no plant samples were identified as metal hyper-accumulator. However *Hyosymus muticus* and *Citrullus colonythisis* have BCF greater than one where *Hyosymus muticus* was effective in taking up Ni while *Citrullus colonythisis* was effective in taking up Cu and Mn. So it can be ended that *Hyosymus muticus* and *Citrullus colonythisis* was considered as the most promising species for phyto-excluder of heavy metals in contaminate site.

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