

## EVALUATION OF PHYTOREMEDIATION POTENTIAL OF SIX WILD PLANTS FOR METAL IN A SITE POLLUTED BY INDUSTRIAL WASTES: A FIELD STUDY IN RIYADH, SAUDI ARABIA

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### Abstract

This study aimed to assess the accumulation content of toxic heavy metals such as Cd, Zn, Cu, Ni and Pb in the soil, shoots and roots of six plants species collected from the second industrial zone of Riyadh, Saudi Arabia. Translocation factor (TF), biological concentration factor (BCF) and bioaccumulation coefficient (BAC) parameters were used to evaluate the of phytoremediation potential of the six studied plants named *Malva parviflora*, *Datura stramonium*, *Citrullus colocynthis*, *Rhazya stricta*, *Phragmites australis* and *Lycium shawii*. Metal concentrations of Cd, Zn, Cu, Ni, and Pb in soils collected from industrial region varied between 19.79, 217, 332, 37.12 and 169.8 mg/kg. The pattern of metal accumulation in studied plants were: Zn > Cu > Pb > Ni > Cd. Results obtained showed significant ( $P \geq 0.05$ ) accumulation in the above ground components of *Malva parviflora*, *Rhazya stricta*, *Phragmites australis* and *Lycium shawii* compared with root. However, the highest accumulation of Cd and Pb was observed in *Phragmites australis* and followed by *Lycium shawii*. The translocation factor of Cd and Pb with most plant species are greater than 1, indicating that these moved more easily in these plants. However, these results also showed that the translocation of Cd and Pb from root to shoot of *Phragmites australis* and *Lycium shawii* plants was higher than other metals. Similar pattern was observed with *Malva parviflora* and *Rhazya stricta*. In conclusion, none of our studied plant species were identified as hyperaccumulator; however, *Phragmites australis* and *Lycium shawii* together with *Malva parviflora* and *Rhazya stricta* showed a highly positive phytoextraction potential for Cd and Pb. whereas, *Datura stramonium* and *Citrullus colocynthis* were found to be suitable for phytostabilization of soils contaminated with Ni and Cu.

### Introduction

Environmental pollution with heavy metals is a global disaster that is related to human activities such as mining, smelting, electroplating, energy and fuel production, power transmission, intensive agriculture, sludge dumping, and melting operations (Igwe & Abia, 2006). All the heavy metals at high concentrations have strong toxic effects and are regarded as environmental pollutants (Chehregani *et al.*, 2005; Sawidis, 2008). The volume of wastewater is increasing day by day due to over population and spreading industrialization (Kahlowan *et al.*, 2006). The global volume of wastewater is more than 1,500 km<sup>3</sup> per year (Anon., 2007). The transfer trend of heavy metals from soil to forage is very high so, the toxicity due to their higher levels can be expected for the grazing ruminants during any time of the year (Asia *et al.*, 2011; Khadijeh *et al.*, 2011). The most common heavy metals at hazardous waste sites are Cadmium (Cd), Chromium (Cr), Copper (Cu), Lead (Pb), Mercury (Hg), Nickel (Ni) and Zinc (Zn) (Anon., 1997). Big cities are constantly generating huge volume of effluent water which needs proper disposal and management (Ghosh & Singh, 2005). Numerous efforts have been undertaken recently to find methods of removing heavy metals from soil, such as phytoremediation (Antonkiewicz & Jasiewicz, 2002; Horsfall & Spiff, 2005; Igwe & Abia, 2006). For chemically polluted lands, vegetation plays an increasingly important ecological and sanitary role (Antonkiewicz & Jasiewicz, 2002). Phytoremediation has recently become a subject of public and scientific interest and a topic of many recent researches (Igwe & Abia, 2006; Horsfall & Spiff, 2005). Moreover

phytoremediation is one of the promising methods for reclamation of soils contaminated with toxic metals by using hyperaccumulator plants (Baker *et al.*, 2000; Ghosh & Singh, 2005; La'zaro *et al.*, 2006). More than 400 plant species belonging to 45 plant families have been identified and reported from temperate to tropical regions with the ability to tolerate and hyperaccumulate trace elements (Baker & Brooks, 1989). These plants have been considered suitable for soil stabilization and extraction of heavy metals (Madejon *et al.*, 2002). Hyperaccumulator plants can play a key role in the fate of the pollutants of contaminated matrixes via their root systems. The identification of metal hyperaccumulators, plants capable of accumulating extra-ordinary high metal levels, demonstrates that plants have the genetic potential to clean up contaminated soil.

The ability of selecting species of plants, which are either resistant to heavy metals, or can accumulate great amounts of them, would certainly facilitate reclamation of contaminated areas (Bizly *et al.*, 2000; Lasat, 2002; Zulfqar *et al.*, 2012). In the process of phytoremediation pollutants are collected by plants and either decomposed to less harmful forms or accumulated in the plant tissues. Thus, phytoremediation is environmentally friendly, inexpensive and can be carried out in polluted places (remediation in situ) plus the product of decomposition does not require further utilization. New candidate of phytoremediating plants are required especially under various environmental conditions.

The use of plant species to decontaminate and remediate polluted soils with heavy metals is non-existence in Saudi Arabia. In this study 6 plant species were investigated for metals such as Cd, Zn, Cu, Ni and Pb from the second industrial zone of Riyadh city in order

to evaluate the total metal accumulation in the soil and plant parts. Plant species that have the great effectiveness for phytoremediation of different heavy metals through phytostabilization and phytoextraction were identified.

### Materials and Methods

**Experimental design:** This study was conducted in the second industrial zone of Riyadh city, the capital of Saudi Arabia. It was established in 2001 and lies between (Lat.47°15', Long.35°45'). It houses more than 250 different industrial units. Two sites were selected for the collection of plant and soil samples. Plant species collected were the most common/dominant species at this site. Six species such as *Malva parviflora*, *Datura stramonium*, *Citrullus colocynthis*, *Rhazya stricta*, *Phragmites australis* and *Lycium shawii* were collected in November and December in 2010. A total of 8-10 plants including roots, shoots of each species were collected from each site and mixed to form a composite sample, placed in labeled bags and transported to our Laboratory for further analysis. Before analysis, from each plant roots and mixture of stems and leaves were carefully removed and washed (for 2-3 minutes approximately) with tap water and with deionized water to remove any soil and surface dust.

Plant species were chosen after taking into account their abundant existence in the field, which indicated the plants' tolerance to different levels of heavy metals. The accumulator plants were identified regarding the concentration of heavy metals in the subjected plants.

**Soil collection and plant harvest:** Soil samples (five replicates), at 0-20 cm depth from rhizosphere of each plant, were taken from each site from where plant sample was rooted. Soil samples (a composite mixture) were air dried at room temperature for two weeks, crushed and pulverized to pass through 2-mm sieve. For each soil sample, soil pH and EC, texture and heavy metals viz., Cd, Zn, Cu, Ni and Pb were measured. Soil pH was determined using portable pH meter (Instruments, Padova, Italy) in a 1: 2.5 soil-water suspension. Total Dissolved Solids and EC was measured using conductivity meter (4010 Conductivity Meter. Hani. Instruments, Padova, Italy). The proportion of percent sand, silt and clay of the soil samples were calculated to determine soil textural class using Bouyous hydrometer method. Plant harvesting was performed by using a shovel and paying attention not to break the roots. Subsequently, the plant fractions (roots, and shoots), were carefully washed with de-mineralized water. Dry biomass of the plant fractions were measured after drying for 48 h at 105°C in an air-forced oven.

**Heavy metals determination in Soil and plant:** The soil analysis procedures used in the present study have been described in more detail elsewhere Westerman (1990). Total content of Cd, Zn, Cu, Ni and Pb in topsoil and in the plant tissues were determined by an inductively coupled plasma emission spectrometry (ICP ICP-AES

(Perkin Elmer, 4300 DV) of a more comprehensive range of 12-20 elements. The standard solutions used for the ICP analytical calibration have their Certificate of Analysis (Certipur Reference Material).

Heavy metal determination was done according to detailed method that described by Sawidis (2008). To estimate the total heavy metals in the plants, samples (roots and shoots) were dried at 105°C for 24 h in acid-washed and reweighed volumetric 100 ml Pyrex conical flasks. The samples were acid-digested in a microwave oven (CEM, MARSX press, USA), according to the EPA method 3051 (Anon., 1995). After the mineralization, the samples were filtered (0.45 mm PTFE) and diluted. The plant samples collected were acid treated following the EPA method 3052 (Anon., 1995) and analyzed by an ICP-OES.

Average values of five replicates were taken for each determination. The precision of analytical procedures was expressed as Relative Standard Deviation (RSD) which ranged from 5-10% and was calculated from the standard deviation divided by the mean. Chemicals, stock solutions and reagents were obtained from Sigma/Fluka/Merck and were of analytical grade. All glassware before use were washed with distilled water, soaked in nitric acid (30%) overnight, rinsed in deionized water and air-dried.

Biological Concentration Factor (BCF) was calculated as metal concentration ratio of plant roots to soil (Yoon *et al.*, 2006). Translocation Factor (TF) was described as ratio of heavy metals in plant shoot to that in plant root (Cui *et al.*, 2007; Li *et al.*, 2007). Biological Accumulation Coefficient (BAC) was calculated as ratio of heavy metal in shoots to that in soil (Cui *et al.*, 2007; Li *et al.*, 2007).

### Results

**Heavy metals in soil:** The basic soil parameters are showed in Table 1 in addition to the acidity pH for the soil from which plants were sampled at the industrial zone. Soil collected from 2 sites was loamy sand and was a dominant soil texture class with pH varied from 5.9-6.6, EC of 2.3 mS/cm.

**Table 1. Basic parameters measured in soil of two sites in the field trial of industrial region in Riyadh City (Saudi Arabia). Each value is an average of five replications.**

Parameter	%	
	Site 1	Site 2
Sand (2- 0.05 mm )	69.54	70.38
Silt (0.02-0.05 mm)	6.27	5.65
Silt (0.02- 0.002 mm)	17.15	16.25
Clay (<0.002 mm)	7.14	7.67
Acidity pH (H <sub>2</sub> O)	5.9	6.6

Total concentrations of heavy metals in the soils from which studied plants were sampled at the studied sites were presented in Table 2. The experimental site of the industrial region of Riyadh city appears to be particularly suitable for studies on phytoextraction and phytostabilization, because of the presence of several metals in the soil layer that can be accessed by the roots of the plants.

**Table 2. Comparative total metal content (mg Kg<sup>-1</sup>) of the soil sample collected from control unpolluted region and industrial region in Riyadh. Each value is an average of five replications (p≤0.05).**

Metal	Soil (mg Kg <sup>-1</sup> )	
	Control region	Industrial region
Cd	0.061 ± 0.002	19.79 ± 1.76
Zn	29.26 ± 2.82	217 ± 2.17
Cu	21.37 ± 1.24	332 ± 6.34
Ni	2.13 ± 0.09	37.12 ± 2.76
Pb	1.12 ± 0.06	169.8 ± 3.73

**Heavy metals accumulation in plants:** Table 3 shows the concentrations of the elements measured in the roots and shoots of the plants of *Malva parviflora*, *Datura stramonium*, *Citrullus colocynthis*, *Rhazya stricta*, *Phragmites australis* and *Lycium shawii* which were collected from the study region.

**Cadmium:** Significant differences between the studied plants (p<0.05) in Cd concentrations and the pattern of Cd concentrations in studied plants were: *Phragmites australis* > *Rhazya stricta* > *Lycium shawii* > *Malva parviflora*. ANOVA reveal a significant effect of plant type on biological concentration factor (BCF) and bioaccumulation coefficient (BAC) of the plant. The shoots of most studied plants showed higher values of Cd and Pb concentrations than those observed in the roots. As a consequence, the translocation factor (shoot/root calculated in terms of concentration), which is typically >1 in the case of accumulator species, was >1 for Cd and Pb elements of our plants.

Total Cd concentration in the shoots of *Phragmites australis*, *Rhazya stricta*, *Lycium shawii* were 23.63, 18.29 and 16.73 mg kg<sup>-1</sup>, respectively. On the other hand the corresponding values for *Malva parviflora*, *Datura stramonium* and *Citrullus colocynthis* were significantly lower and reached to 6.88, 6.23 and 4.78, respectively (p<0.05). In regard to the values of Cd concentration measured in the shoots and roots of the studied plants, *Phragmites australis* and *Rhazya stricta* accumulated more Cd than all other studied plants (p<0.05), and the levels of concentration were significant from the perspective of phytoextraction.

**Zinc:** Zinc concentration in the roots was significantly higher in our studied plants (p<0.05). Their concentration in the roots of *Datura stramonium*, *Lycium shawii*, *Phragmites australis* and *Rhazya stricta*, were 118.8, 89.72, 75.8 and 66.9 mg kg<sup>-1</sup>, respectively (Table 3). Moreover, the Zn concentration in the shoots of the same plants was 49.8, 37.3, 53.7 and 41.8 mg kg<sup>-1</sup>, respectively (Table 3). Such values associate with a translocation factor of 0.42, 0.41, 0.71 and 0.63.

**Copper:** In regard to the concentration of Cu in the shoots and roots of plant species, ANOVA revealed a significant effect of the species on the Cu concentration (p<0.05). On average, the Cu concentration in shoots was about 2 to 4.5 times lower than in the roots, for all studied plants. The concentration of Cu measured in the roots of *Lycium shawii* and *Phragmites australis* were 312 and 297.3 mg kg<sup>-1</sup> (Table 3). The shoot of *Malva parviflora* had the lowest concentration of Cu, 62.4 mg Kg<sup>-1</sup> compared to other studied plants.

**Table 3. The elemental mean values mg Kg<sup>-1</sup> in shoots and roots of six plant species.**

Plant species	Cd		Zn		Cu		Ni		Pb	
	Shoot	Root	Shoot	Root	Shoot	Root	Shoot	Root	Shoot	Root
<i>Malva parviflora</i> ,	6.86	3.19	39.7	87.3	62.4	282	2.21	7.87	11.41	4.73
<i>Datura stramonium</i> ,	6.23	4.13	49.8	118.8	57.8	261.7	3.07	8.32	6.43	4.65
<i>Citrullus colocynthis</i> ,	4.78	2.83	79.8	48.5	68.7	277.5	2.71	5.9	9.34	4.36
<i>Rhazya stricta</i> ,	18.29	9.35	41.8	66.9	78.9	197.3	3.13	7.32	28.63	10.32
<i>Phragmites australis</i>	23.63	5.31	53.7	75.8	123	297.3	4.30	8.21	45.68	10.73
<i>Lycium shawii</i>	16.73	5.27	37.3	89.7	162	312	3.22	6.12	37.34	11.53
<b>Mean</b>	<b>12.76</b>	<b>5.01</b>	<b>50.35</b>	<b>81.20</b>	<b>92.13</b>	<b>271.3</b>	<b>3.11</b>	<b>7.29</b>	<b>23.4</b>	<b>7.72</b>

**Nickel:** Variable concentrations of Ni were observed in our experiment and we found that, roots of *Datura stramonium* and *Phragmites australis* had the highest concentrations of Ni in comparison to the other studied plants.

The shoot/root translocation factor of Ni was not very high and ranged between 0.28-0.53 and showed a significant effect for the plant species (p<0.05). The concentrations of Ni in the roots of *Datura stramonium* and *Phragmites australis* had concentrations that were, respectively, 2.7 and 2 times lower than in the shoots.

**Lead:** The concentration of Pb measured in the *Phragmites australis* roots were nearly 4.2 fold higher than their concentration in *Datura stramonium*, and their concentration in *Lycium shawii* and *Rhazya stricta* were significantly high in comparison with the other plants (p<0.05). As previously mentioned in case of cadmium element the Pb content was higher in shoots of most studied plants than in roots. Moreover, the translocation factor was more than 1 in most studied plants.

**The interspecies comparison of the heavy metals concentrations:** In order to analyze the interspecies differences of plant heavy metal accumulation capacity, the comparison of the elemental mean concentrations between six plants was shown in Table 3. The concentration of over 4/5 elements in *Lycium shawii*, *Phragmites australis* and *Rhazya stricta* were higher than other species (Table 3). However, contents of Cd, Ni and Pb elements in *Malva parviflora*, *Datura stramonium* and *Citrullus colocynthis* were lower than the average levels.

Among our studied plant species screened for Cd, Zn, Cu, Ni and Pb, most of the species were efficient to take up and translocate more than one heavy metal from roots to shoots (Table 4). Variations between TF values were found (Table 4). The highest TF value was found for

*Phragmites australis* and *Lycium shawii* were 4.45 and 3.17 for Cd and 2.26 and 3.24 for Pb.

Most screened plants showed high values of TF >1, especially for *Phragmites australis* and *Lycium shawii*, as well as, *Malva parviflora* and *Rhazya stricta*. As a consequence these plants were efficient in translocation of metals such as Pb and Cd from roots to shoots for Cd and Pb. Plant species such as (Table 4).

Table 4 indicated that all studied plant species screened for total metal concentration showed value of BAC < 1 for all heavy metals except the value of *Phragmites australis* for Cd. It varied between 0.041-1.19. Highest BAC values of Cd were found for *Phragmites australis* (1.19) and *Rhazya stricta* (0.92). Similar pattern were observed for BCF in all studied plants.

**Table 4. Biological concentration factor (BCF), Translocation factor (TF) and biological accumulation coefficient (BAC) of plant species for determined metals.**

Plant Species	Cd			Zn			Cu			Ni			Pb		
	BCF	TF	BAC	BCF	TF	BAC	BCF	TF	BAC	BCF	TF	BAC	BCF	TF	BAC
<i>Malva parviflora</i>	0.16	2.16	0.35	0.40	0.46	0.18	0.85	0.22	0.19	0.21	0.28	0.06	0.03	2.41	0.07
<i>Datura stramonium</i>	0.21	1.51	0.32	0.55	0.42	0.23	0.79	0.22	0.17	0.22	0.37	0.08	0.03	1.38	0.04
<i>Citrullus colocynthis</i>	0.14	1.69	0.24	0.22	1.65	0.37	0.84	0.25	0.21	0.16	0.46	0.07	0.03	2.14	0.06
<i>Rhazya stricta</i>	0.47	2.14	0.92	0.31	0.63	0.19	0.59	0.40	0.24	0.20	0.43	0.08	0.06	2.77	0.17
<i>Phragmites australis</i>	0.27	4.45	1.19	0.35	0.71	0.25	0.90	0.41	0.37	0.22	0.53	0.12	0.06	2.26	0.27
<i>Lycium shawii</i>	0.27	3.17	0.85	0.41	0.42	0.17	0.94	0.52	0.49	0.17	0.53	0.09	0.07	3.24	0.22

## Discussion

Large number of factors control metal accumulation and bioavailability associated with soil and climatic conditions, plant genotype and agronomic management, including: active/passive transfer processes, sequestration and speciation, redox states, the type of plant root system and the response of plants to elements in relation to seasonal cycles (Kabata-Pendias & Pendias, 1984).

Heavy metals contamination of arable soil showed several problems, including phytotoxic effects of certain elements such as Cd, Pb, Zn and Cu, which are well known as micronutrients and cause several phytotoxicities if critical endogenous levels are exceeded (Mengel & Kirkby, 2001; Chehregani *et al.*, 2005). Another and even a more serious problem is posed by the up taking of potentially noxious elements through food or forage plant species and their being transferred to the food chain and, finally, to humans (Kloke, 1980).

The analysis of data showed that the interspecies concentrations of most elements were significantly different. Accordingly the biological characters for each species, such as living form, morphology, in addition to, the climatic condition had a great effect on the accumulation capacity of the element in plant. We concluded that, even, plants were collected from the same region, the uptake efficiency of the elements was significantly varied, this could be regarded as the effect of plant species, climatic condition, as well as, the nature of metal element on its efficiency.

The total mean concentrations are all higher when compared with the value reported by Ruling, 1994 in European countries and other countries (Percy, 1983, Onianwa & Ajayi, 1987, Pignata *et al.*, 2002).

According to the results of this study, the plants presented below can be regarded as heavy metal accumulators while they are different regarding their accumulating ability (Tables 3 and 4). The study also concludes that *Phragmites australis* could be considered as Cd accumulator plant especially its content in their shoot reached the highest concentration between the six studied plants. Also *Lycium shawii* and *Rhazya stricta* could be regarded as Cd accumulator plants (Tables 3).

High root to shoot translocation of these metals indicated that these plants have vital characteristics to be used in phyto-extraction of these metals as indicated by Ghosh & Singh (2005) and La'zaro *et al.*, (2006). Plant species with slow plant growth, shallow root system and small biomass production are not generally preferred for phytoremediation. These two species had high biomass and based on high TF values could have enormous potential to be used for phyto-extraction of Cd and Pb than other species which also showed TF > 1 for different metals. Yoon *et al.*, 2006 showed that plant species with high TF values were considered suitable for phytoextraction generally requires translocation of heavy metals in easily harvestable plant parts i.e., shoots.

According to Ghosh & Singh (2005) phyto-extraction is a process to remove the contamination from soil without destroying soil structure and fertility. Great metal accumulation may be attributed to well develop

detoxification mechanism based on sequestration of heavy metal ions in vacuoles, by binding them on appropriate ligands such as organic acids, proteins and peptides in the presence of enzymes that can function at high level of metal ions (Cui *et al.*, 2007) and metal exclusion strategies of plant species (Ghosh & Singh, 2005). In general the results indicated that none of the plant species were identified as hyperaccumulator because all species accumulated Cd, Zn, Cu, Ni and Pb less than 1000 mg/kg (Baker & Brooks, 1989). However, based on BCFs, TFs, and BAC values plant species were identified which have the potential for phytostabilization and phytoextraction. Most of the species were efficient to take up and translocate more than one metal from roots to shoots. Based on highest TF value *Phragmites australis* and *Lycium shawii* can be used for phytoextraction of Cd and Pb.

The study concludes that *Phragmites australis* could be considered as accumulator plant for most studied metals. This is the first report focusing on their ability as metal accumulator. Moreover, *Lycium shawii* and *Rhazya stricta*, which were more current in the polluted area, were selected as a good metal accumulator, especially a good for Cd and Pb.

### Conclusion

This study concluded that Al-Kharj industrial area appears to be particularly suitable for studies on phytoextraction, because of the presence of several metals in the soil layer that can be accessed by the roots of the plants. The results of this study also indicated that there is an increasing need for further research on the mechanisms whereby such plants are able to survive in contaminated soils. Furthermore, studies are needed to determine the growth performance, biomass production and metal accumulation of these species in metal contaminated soils for their better management and conservation.

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