**Environmental Resources Research**
Vol. 9, No. 1, 2021

GUASNR

# Screening of native plant species for phytoremediation potential in Pb-Zn mines in Iran

**M. Kazemi**
M.Sc. graduated, Payame Noor University, Department of Environment,
Tehran Shargh branches, Tehran, Iran

*Received: January 2020; Accepted: September 2020*

**Abstract**

Mining activity is one of the major sources of heavy metal pollution of soil. Most mines are abandoned without any remediation and cause contamination of soil and water. Phytoremediation is an environment-friendly technology for the remediation of contaminated sites. In this work, native plant species were identified that can tolerate high concentrations of heavy metals and are useful for phytoremediation in an abandoned Pb-Zn mine in the north of Iran. Twelve plant species and the corresponding soils were collected and analyzed for As, Cd, Co, Cr, Cu, Mo, Pb, Sb, Ni and Zn contents using inductively coupled plasma mass spectroscopy (ICP-MS). In order to measure the bioavailability of heavy metals for plants uptake, DTPA extraction of heavy metals were determined. Then, the physiochemical characteristics of the soil samples were measured and the translocation factors (TFs) and bio-concentration factors (BFs) were determined. The soil samples were alkaline, and exhibited low electrical conductivity, high cation-exchange capacity, moderate organic carbon content, and clay loam texture. All samples exceeded the soil toxicity thresholds for AS, Cd, Pb, Sb, and Zn. The results indicate that *Stachys byzantina* has the ability to accumulate significant amounts of Pb in its shoot. The average concentrations of Pb in the soil, shoot, and root were 15472mg $kg^{-1}$, 1797 mg $kg^{-1}$, and 371 mg $kg^{-1}$, respectively, with TF value of 4.8 and BF value of 0.1. Therefore, *S. byzantina* may have the potential to function as a Pb hyper-accumulator and merits further investigation.

***Keywords*:** Phytoremediation; Pb hyper-accumulator; Heavy metals; *Stachys byzantina*; Pb-Zn mine tailing

## Introduction

Industrial mining activities are one of the significant sources of contamination and their deleterious effects impact soils, water bodies, wildlife habitats, and quality of life far from their location. Contaminated soil threatens the human health as heavy metals can be transferred into the food chain, especially when these sites are converted to agricultural land or pastureland (Dickinson, 2017). Therefore, it is necessary to reclaim the contaminated sites and reduce the risk of transfer of contaminants along the food chain.

Phytoremediation is a technique wherein metal tolerant plants are used to decontaminate the environment. Some plants, called hyper-accumulators, can accumulate significant quantities of heavy metals in their tissues, and are suitable for phytoremediation. The metal accumulation ability of hyper-accumulator plants is approximately 100–1000-fold higher than that of normal plants growing in soil with normal concentrations of metals, and 10–100-fold higher than that of plants growing in metal-contaminated soils (McGrath, Zhao, & Lombi, 2002). Hyper-accumulators are described as plants can accumulate more than 10000 mg $kg^{-1}$ of Zn or Mn, or more than 1000 mg $kg^{-1}$ of Co, Cu, Cr, Pb, or Ni, or more than 100 mg $kg^{-1}$ of Cd or other rare metals, measured in dry weight (Baker and Brooks, 1989; cited in

*Corresponding author; maryam.kazemi2@yahoo.com

Koleli et al., 2015). Moreover, hyper-accumulators are characterized by a shoot-to-root metal concentration quotient, i.e., translocation factor (TF) of more than one (Baker, 1981), and a shoot-to-soil metal concentration quotient, i.e., bio-concentration factor (BF), of more than one (Brooks, 1998).

In a study on the Angouran Pb-Zn mine in Iran, Chehregani, Noori, & Yazdi (2009) reported *Noea mucronata* as a Pb hyper-accumulator; after two years of phytoremediation using plant colonies of *N. mucronata*, the concentrations of Pb in the experimental pots decreased considerably. Boojar and Tavakkoli (2011) investigated two dominant plant species, *Zygophyllum fabago* and *Peganom harmala*, that naturally colonized the region around the Angouran Pb-Zn mine in Iran. They measured the accumulation of Pb, Zn, and Cu, and the chlorophyll and biomass contents of the plant tissues in metal-contaminated and non-contaminated sites. As markers of oxidative stress, the malondialdehyde and dityrosine contents of the plant tissues were measured. However, none of the plants absorbed significant amounts of the elements in the abovementioned sites, and the contents of chlorophyll, biomass, malondialdehyde, and dityrosine in the two plant species did not vary markedly between the two zones studied. In another study carried out by Mahdavian, Ghaderian, and Torkzadeh-Mahani (2015), in a Pb-Zn mine in Yazd, a city in the center of Iran with a semi-arid climate, 40 plant species and the associated soil samples were collected and analyzed for Pb, Zn, and Ag; TF and BF values also were calculated; they tentatively used 1 mg $kg^{-1}$ in plant dry maters as a threshold for Ag hyper-accumulator; some of the plant species including *Colchicum schimperi, Londesia eriantha, Lallemantia royleana, Bromus tectorum, Hordeum glaucum*, and *Thuspeinantha persica* reached this threshold, with Ag concentrations in shoots ranged between 0.5-3.8 mg $kg^{-1}$, TF values above one (1.3.-4.2), and BF values almost above one (0.7-3.1). Hesami, Salimi, and Ghaderian (2018) also researched a Pb-Zn mine in Isfahan-Iran to find locally adapted species for phytoremediation purposes, 69 vascular plant species and associated soils were collected; concentrations of the soluble, exchangeable and total amount of Pb, Zn, Cd, Ca, and Mg in the soil samples were measured; for plants concentration of Pb, Zn and Cd in shoots and roots were measured; TF and BF factors were calculated; they also measured Ca/Mg ratio as a factor that helps plants to uptake less Pb and Zn and reduces toxicity in plants. In a survey conducted on cultivated rice (*Oryza sativa)*, Kim, Yang, & Lee (2002), showed a high concentration of Ca blocked Pb uptake into roots and high concentration of both Ca and Mg block Cd uptake in roots which reduce toxicity caused by high Pb and Cd in plants.

Still, the search for hyper-accumulator species compatible with the surrounding environment is necessary. The selection of true hyper-accumulators is essential for the remediation of contaminated soils. It is desirable that hyper-accumulator plants be identified and examined in their natural habitats (van der Ent, Baker, Reeves, Pollard, & Schat, 2013). The use of native plants for phytoremediation reduces the risk of invasive species that could threaten indigenous plant diversity (Mohanty, Dhal, Patra, Das, & Reddy, 2010). In the present work, the aim was to determine native plant species that can tolerate high concentrations of heavy metals in their above-ground tissues.

## Materials and methods

### *Site description and collection of samples*

The study was carried out in a former Pb-Zn mine that had been mined from 1923 until 1971. The mine has nine holes at different elevations, all of which had been abandoned for over 40 years; mining activity at the site was resumed in 2017. The Marjanabad Pb-Zn mine is located at a high elevation in the Manjil region in the Guilan Province of Iran (Fig. 1). The geographic coordinates of the sampling site are as follows: longitude, 49° 28′ 18″; latitude, 36° 46′ 57″; and altitude, 1700 m above sea level. The region generally has a dry climate with average annual precipitation of about 165 mm. The average annual temperature is about 17.5°C. The warmest month is August with a maximum temperature of about 38.5°C. By contrast, January is usually the coldest month with a minimum temperature of about -5°C.

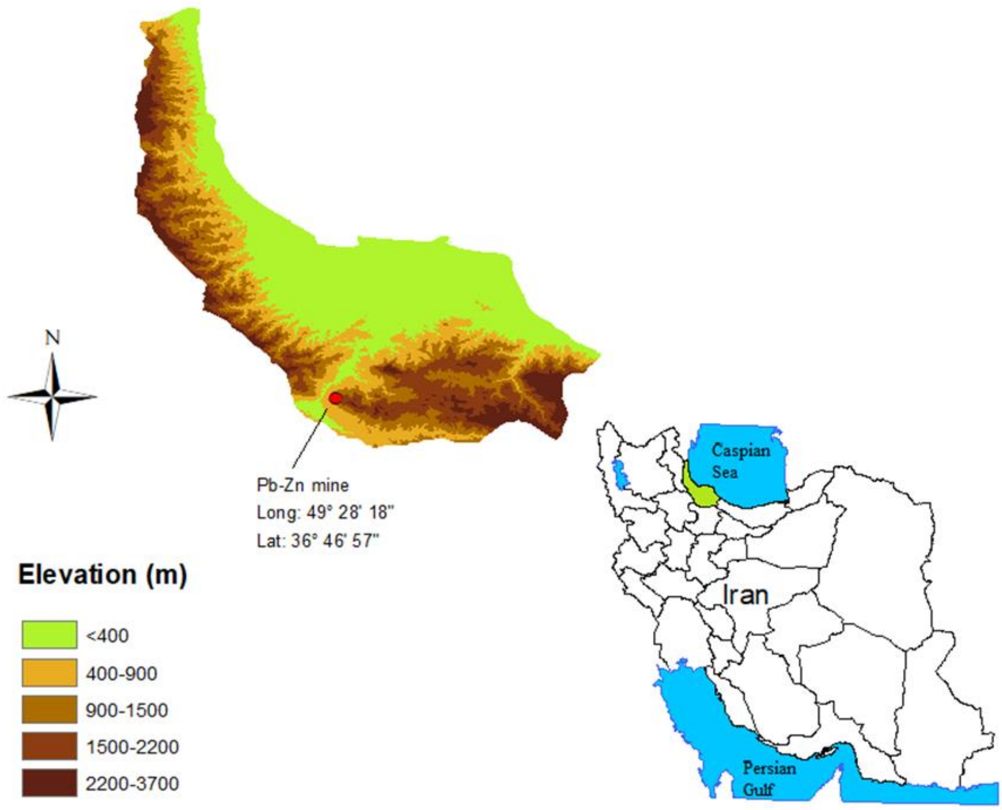


**Figure 1.** Location of survey area in Guilan province

The mine is placed in hill forest and dominated by trees and shrubs, such as olive, cypress, beech, oak, hornbeam, checker tree, juniper, ilex, and hawthorn, that have grown owing to the flow of groundwater. There is a village downstream with a population of around 2600 people. The main job of villagers is agriculture and animal husbandary, and the chief agricultural product of the region is olive.

In the course of field survey, an open area was found at the top of the mountain that had undergone significant changes. The soil there had been extensively excavated, the trees had been cut, and consequently, herbs were growing well in the region. Therefore, the study was conducted there and one sample site was chosen. In order to have a general vision of physiochemical characteristics of soil, four soil samples were randomly collected and placed in plastic bags and sent to the laboratory for further analysis. Specimens of plants, shrubs and trees that were more common were randomly collected. Sampling was carried out in August 2017. Healthy, mature plants of the same physiological age and appearance were collected. Plants were avoided that were affected by diseases, insect damage, and physical or chemical injury. A total of twelve plant samples which were the most common species in the area were collected, including five herbaceous plant species belonging to five different families, and three species of shrubs, and four species of trees. For trees and shrubs only leaves were taken with a clean scissor, and for herbaceous species whole plants were excavated and stored in paper bags. Three replicates of each plant were collected, two of which were employed for the purpose of identification and one for the measurement of heavy metal contents. Soil samples were collected around the roots of the plants gathered. Some plants were growing in the same place, so ten soil samples were taken. The plants were divided into roots and shoots. The samples were carefully washed with tap water, rinsed with distilled water, then washed with 0.1 mol $L^{-1}$ HCl to ensure there were no soil particles adhering to the plants, and rinsed with distilled water several times. The samples were dried at 70°C for three days, ground to pass a 2 mm mesh sieve, placed in labeled bags, and transported to the environmental laboratory for further analysis. The soil samples were dried in air, stored in plastic bags, and transported to the environmental laboratory for further analysis.

### *Soil and plant analyses*

In the laboratory, the physical and chemical properties of soil were determined via several methods. The soil acidity (pH) and electrical conductivity (EC) values were measured via the saturated extraction method as described by Warncke (1986), and read using a pH meter and EC meter, respectively, while the soil texture was analyzed via the hydrometer method as described by Gee and Bauder (1986). The cation-exchange capacity (CEC) was determined using the Bower method (Bower, Reitemeier & Fireman, 1952). The organic carbon content was determined using the Walkley and Black procedure (Nelson & Sommers, 1982).

For the analysis of heavy metals in soil, the soil samples were pulverized using a low-Chrome stainless steel mortar and pestle and passed through a 0.075 mm mesh plastic sieve. The total metal contents of the soil samples were determined via multi-acid digestion of 250 mg of soil using HF, HCl, $HClO_4$, and $HNO_3$. The resultant solutions were analyzed via inductively coupled plasma mass spectrometry (ICP-MS) with detection sensitivities of 0.1 mg $kg^{-1}$ for As, Cd and Mo, and 1 mg $kg^{-1}$ for Co, Cr, Cu, Ni, Pb and Zn, and 0.5 mg $kg^{-1}$ for Sb.

Extractable metals were measured by adding 50 ml DTPA (diethylene triaminepenta acetic acid) to 25 g of the pulverized soil and shaking for 2-hr, samples were filtered through Whatman filter paper, and the resulting solution was analyzed by using inductively coupled plasma optical emission spectrometry (ICP-OES). The detection limit was 0.05 mg $kg^{-1}$ for all heavy metals.

For the analysis of heavy metals in the plants, a mixture of $HNO_3/HClO_4$ was added to 250 mg of dry plant tissues. The cell containing the samples and reagents was placed in a microwave, after the plant samples were diluted with, and dissolved in, distilled water. The resultant solutions were analyzed via ICP-MS. The detection limits were 0.01 mg $kg^{-1}$ for As, Cd, Co, Cr, Mo, Ni, Sb and Pb, and 1 mg $kg^{-1}$ for Cu and Zn. Then, the BF and TF values were determined. The BF value was expressed as the quotient of metal concentration in the shoot to that in the soil, and the TF value was expressed as the quotient of metal concentration in the plant shoot to that in the root (Deng, Ye, and Wong, 2004).

## Results

### *Soil properties*

The physiochemical characteristics of the soil samples are presented in Table 1. All the soil_samples were alkaline, and the pH values were approximately 8. The pH value is one of the most important factors controlling the bioavailability of metals in soil. The soil samples were classified based on their EC values as follows: less than 2, non-saline; 2–8, moderately saline; 8–16, very saline; and exceeding 16, extremely saline (Boulding, 1994). The soil samples had low EC values (< 2 dS $m^{-1}$) and were non-saline. The CEC value shows the ability of the soil to retain exchangeable cations, and is an important factor in sites polluted by heavy metals. In general, soils with high CEC can absorb significantly more quantities of heavy metals than those absorbed by soils with low CEC. Almost all the soil samples in the present study had high CEC values (20–33 me.$100^{-1}$). The CEC of soil is primarily driven by its organic matter content. The organic carbon contents of the soil samples were moderate (2–7%), and the texture was mostly clay loam.

**Table 1.** Physiochemical characteristics of soil samples

| Site code | pH | EC | CEC | OC | Sand | Clay | Silt | Soil texture |
|---|---|---|---|---|---|---|---|---|
| | | dS $m^{-1}$ | me.$100^{-1}$ | % | % | % | % | |
| I | 8.2 | 0.58 | 20.5 | 2.47 | 38 | 26 | 36 | Loam |
| II | 8.2 | 0.93 | 26.4 | 2.24 | 25 | 48 | 27 | Clay |
| III | 7.9 | 0.85 | 32.7 | 6.7 | 17 | 31 | 52 | Silty Clay Loam |
| IIII | 8.1 | 1.37 | 25.2 | 7 | 33 | 18 | 49 | Loam |

OC: Organic carbon
EC: Electrical conductivity
CEC: Cation-exchange capacity

### *Heavy metals in soils*

As seen in Table 2, the concentrations of As, Cd, Pb, Zn and Sb in the soils were mostly outside the ranges of maximum allowable concentration (MAC) and trigger action values (TAVs) of trace metals in agricultural soils, as established by Kabata-Pendias and Mukherjee (2007). The average As concentrations in the soils were relatively high and in the range 26–760 mg $kg^{-1}$. The Cd concentrations in the soils varied from a minimum of 0.3 mg $kg^{-1}$ to a maximum of almost 48 mg $kg^{-1}$. The soils were extremely contaminated with Pb and the Pb concentrations were in the range 450–24226 mg $kg^{-1}$. Similarly, the average Zn concentrations were high and varied in the range 138–37100 mg $kg^{-1}$. The concentration of Sb varied from 6 to 153.

**Table 2.** Total metal concentration of soil samples

| Site code | As | Cd | Co | Cr | Cu | Mo | Pb | Sb | Ni | Zn |
|---|---|---|---|---|---|---|---|---|---|---|
| | mg/kg | | | | | | | | | |
| 01 | 590 | 8.6 | 12.4 | 102 | 132 | 1.6 | 15472 | 66.1 | 38 | 4153 |
| 02 | 248 | 48.1 | 4.8 | 50 | 77 | 1.5 | 11231 | 153.5 | 25 | 37100 |
| 03 | 760 | 32.9 | 12.6 | 106 | 99 | 3.8 | 24226 | 70.3 | 67 | 19792 |
| 04 | 26.1 | 0.3 | 16.9 | 107 | 60 | <DL | 2010 | 6.4 | 58 | 148 |
| 05 | 70.4 | 1.2 | 18 | 94 | 35 | 1.4 | 700 | 12.7 | 43 | 748 |
| 06 | 47.3 | 1.6 | 16.2 | 104 | 26 | 2.8 | 1119 | 31 | 38 | 1758 |
| 07 | 153 | 22 | 15.9 | 110 | 43 | 1.7 | 4406 | 74.9 | 54 | 12144 |
| 08 | 107 | 3.1 | 14.6 | 114 | 30 | 2.3 | 498 | 6.8 | 53 | 1229 |
| 09 | 108 | 2.4 | 16.1 | 111 | 31 | 1.9 | 450 | 11.9 | 60 | 947 |
| 10 | 239 | 1.3 | 19.4 | 115 | 48 | 1.2 | 622 | 9.9 | 66 | 242 |
| Range | 26.1 - 760 | 0.3 - 48.1 | 4.8 - 19.4 | 50 - 115 | 26 - 132 | 0 - 3.8 | 450 - 24226 | 6.4 - 153.5 | 25 - 67 | 148 - 37100 |
| MAC | 15 - 20 | 1 - 5 | 20 - 50 | 50 - 200 | 60 - 150 | 4 - 10 | 20 - 300 | 10 | 20 - 60 | 100 - 300 |
| TAV | 10 - 65 | 2 - 10 | 30 - 100 | 50 - 450 | 60 - 500 | 5 - 20 | 50 - 300 | 10 - 20 | 75 - 150 | 200 - 1500 |

<DL: Below detection limit; MAC, ranges of Maximum Allowable Concentration and TAV, Trigger Action Value for trace metals in agricultural soils (mg/kg) (A. Kabata-Pendias & Mukherjee, 2007).

DTPA extraction of heavy metal was used to find out the bioavailability of heavy metals for plants uptake. The concentrations of DTPA-extractable for As, Co, Cr, Cu, Mo, Sb, and Ni were considerably low or undetectable (Table 3). The available Cd concentrations were less than 10 mg $kg^{-1}$, the available Pb concentration ranged from 120 to 1552 mg $kg^{-1}$, and the available Zn concentration varied between 2.5 to 573 mg $kg^{-1}$.

**Table 3.** Available metal concentrations in soil samples

| | mg/kg | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| Site code | As | Cd | Co | Cr | Cu | Mo | Pb | Sb | Ni | Zn |
| 01 | <DL | 1.14 | <DL | <DL | 2.26 | <DL | 1552 | <DL | 0.44 | 95 |
| 02 | <DL | 6.6 | <DL | <DL | 1.64 | <DL | 138 | 0.1 | 0.38 | 573.2 |
| 03 | <DL | 5.02 | <DL | <DL | 2.38 | <DL | 940 | <DL | 0.54 | 270 |
| 04 | 0.1 | <DL | 0.62 | <DL | 3.5 | <DL | 892 | <DL | 0.36 | 2.6 |
| 05 | 0.2 | 0.4 | <DL | <DL | 1.6 | <DL | 208 | <DL | 2.2 | 28.8 |
| 06 | 0.1 | 1.04 | 0.82 | <DL | 1.02 | <DL | 234 | <DL | 1.16 | 91.2 |
| 07 | 0.1 | 9.84 | 0.3 | <DL | 0.5 | <DL | 242 | <DL | 0.7 | 469.6 |
| 08 | 0.2 | 2.04 | 1.56 | <DL | 1.08 | <DL | 130 | <DL | 1 | 112 |
| 09 | 0.2 | 1.56 | 2.16 | <DL | 1.08 | <DL | 122 | <DL | 1.06 | 92.4 |
| 10 | 0.2 | <DL | 0.56 | <DL | 1.52 | <DL | 120 | <DL | 0.7 | 2.8 |

<DL: Below detection limit

***Heavy metals in plants***

The plant species collected were identified based on the references provided by Flora of Iran (Ghahreman, 1978-2003), and the trees and shrubs were identified based on Trees and Shrubs of Iran (Mozaffarian, 2004). The names of the plant species and their families as well as the heavy metal concentrations in different plant parts are summarized in Table 4. The calculated BF and TF values are presented in Table 5.

As seen in Table 4, the concentrations of As, Pb, and Zn in *Stachys byzantina*, *Thlaspi hastulatum*, and *Urtica dioica* were in the ranges deemed 'excessive' or at 'toxic levels' for plants based on the classification established by Kabata-Pendias (2001).

In *S. byzantina*, the average concentrations of Pb in the shoot and root were quite high, with the former exceeding 1000 mg $kg^{-1}$ in dry weight. The average concentrations of As in the shoot and root were significant although not high enough for the species to qualify as an As hyper-accumulator. The TF values of the plant species were more than one except Cu, which indicated good ability to translocate most elements studied from the root to shoot, although its BF values for all the elements investigated were less than one. In the case of *T. hastulatum*, the average concentrations in the root of the almost all elements studied were higher than those in the shoot, except Mo which is slightly higher in the shoot. None of the elements were present in adequately high levels for the plant to qualify as a hyper-accumulator. Its TF and BF values were less than one for almost all elements. *T. hastulatum* appears to be an excluder since the heavy metal concentrations in its roots are higher than those in its shoots. In the case of *U. dioica*, As, Pb and Zn were in excessive concentration levels, but none of these values were high enough for the species to qualify as a hyper-accumulator. Its TF values were more than one for all the elements studied, except Cr while the BF values were less than one for all elements studied.

The concentrations of all studied elements in *Colchicum speciosum* and *Carduus accanthoides* were not significant and were lower than the toxic levels established by Kabata-Pendias (2001). None of the trees and shrubs accumulate significant amount of heavy metals. Almost all the heavy metals in trees and shrubs including *Crataegus microphylla*, *Quercus castaneifolia*, *Sorbus torminalis*, *Junperus communis*, *Carpinus betulus*, *Fagus orientalis*, *Ilex spinigera* were generally below the toxic levels in plants established by Kabata-Pendias (A. e. Kabata-Pendias, 2001).

**Table 4**. Metal concentration (mg kg$^{-1}$) in plant samples

| Site code | Family | Scientific name | Plant parts | As | Cd | Co | Cr | Cu | Mo | Pb | Sb | Ni | Zn |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | mg/kg | | | | | | | | | |
| 01 | *Lamiaceae* | *Stachys byzantina* | Shoot | 56.53 | 0.26 | 0.42 | 1.44 | 21 | 0.20 | 1797.5 | 1.08 | 7.74 | 1145 |
| | | | Root | 15.45 | 0.26 | 0.18 | 0.84 | 27 | 0.11 | 371.13 | 0.93 | 2.86 | 361 |
| 02 | *Cruciferae* | *Thlaspi hastulatum* | Shoot | 14.64 | 0.95 | 0.08 | 0.49 | 10 | 0.57 | 314.63 | 0.68 | 1.27 | 527 |
| | | | Root | 19.52 | 1.35 | 0.12 | 0.78 | 11 | 0.45 | 562.07 | 1.46 | 3.57 | 861 |
| 03 | *Urticaceae* | *Urtica dioica* | Shoot | 16.36 | 0.38 | 0.09 | 0.21 | 9 | 0.83 | 292.47 | 2.34 | 0.87 | 933 |
| | | | Root | 4.32 | 0.30 | 0.02 | 0.31 | 7 | 0.15 | 71.61 | 1.78 | 0.72 | 158 |
| 04 | *Asteraceae* | *Carduus accanthoides* | Shoot | 1.34 | 0.30 | 0.13 | 0.30 | 10 | 0.06 | 7.23 | <DL | 2.12 | 47 |
| | | | Root | 2.26 | 0.32 | 0.16 | 0.79 | 25 | 0.13 | 33.40 | <DL | 4.61 | 47 |
| 05 | *Colchicaceae* | *Colchicum speciosum* | Shoot | 5.36 | 0.05 | 0.42 | 2.19 | 15 | 0.11 | 14.37 | 1.43 | 1.29 | 61.17 |
| | | | Root | 3.49 | 0.04 | 0.17 | 1.32 | 9 | 0.16 | 14.43 | 1.28 | 0.87 | 55.60 |
| 06 | *Fagaceae* | *Fagus orientalis* | Leaves | 0.43 | 0.11 | 0.05 | 0.14 | 13 | <DL | 0.51 | <DL | 3.28 | 92 |
| 07 | *Corylaceae* | *Carpinus betulus* | Leaves | 6.18 | 0.24 | 0.06 | 0.16 | 8 | <DL | 47.45 | 0.69 | 0.92 | 241 |
| 07 | *Cupressaceae* | *Juniperus communis* | Leaves | 2.25 | 0.17 | 0.06 | 0.33 | 6 | 0.06 | 15.65 | 0.18 | 1.46 | 130 |
| 08 | *Aquifoliaceae* | *Ilex spinigera* | Leaves | 0.06 | 0.76 | 0.01 | <DL | <DL | <DL | 1.26 | <DL | 0.27 | 764 |
| 09 | *Rosaceae* | *Crataegus microphylla* | Leaves | 1.63 | 0.22 | 0.07 | 0.14 | 8 | 0.02 | 10.12 | <DL | 1.25 | 91 |
| 10 | *Fagaceae* | *Quercus castaneifolia* | Leaves | 0.52 | 0.18 | 0.05 | 0.21 | 9 | 0.01 | 0.33 | <DL | 2.51 | 29 |
| 10 | *Rosaceae* | *Sorbus torminalis* | Leaves | 0.66 | 0.17 | 0.04 | 0.26 | 8 | 0.02 | 0.23 | <DL | 1.73 | 68 |
| Approximate concentrations of trace elements in mature leaf tissue.(1) | | Sufficient or normal | | 1 - 1.7 | 0.05 - 0.2 | 0.02 - 1 | 0.1 - 0.5 | 5 - 30 | 0.2 - 5 | 5 - 10 | 7 - 50 | 0.1 - 5 | 27 - 150 |
| | | Excessive or toxic | | 5 - 20 | 5 - 30 | 15 - 50 | 5 - 30 | 20 - 100 | 10 - 50 | 30 - 300 | 150 | 10 - 100 | 100 - 400 |

<DL: Below detection limit; <DL: Below detection limit; [1], Approximate concentrations of trace elements in mature leaf tissue generalized for various species (ppm DW) (A. e. Kabata-Pendias, 2001).

**Table 5.** Bioaccumulation Factor (BF[1]) and Translocation Factor (TF[2]) for plants samples

| Site code | Scientific name | | As | Cd | Co | Cr | Cu | Mo | Pb | Sb | Ni | Zn |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | mg/kg | | | | | | | | | |
| 01 | *Stachys byzantina* | Tf | 3.66 | 1.02 | 2.33 | 1.72 | 0.78 | 1.80 | 4.84 | 1.16 | 2.71 | 3.17 |
| | | BF | 0.10 | 0.03 | 0.03 | 0.01 | 0.16 | 0.13 | 0.12 | 0.02 | 0.20 | 0.28 |
| 02 | *Thlaspi hastulatum* | Tf | 0.75 | 0.70 | 0.69 | 0.62 | 0.91 | 1.24 | 0.56 | 0.47 | 0.36 | 0.61 |
| | | BF | 0.06 | 0.02 | 0.02 | 0.01 | 0.13 | 0.38 | 0.03 | 0.00 | 0.05 | 0.01 |
| 03 | *Urtica dioica* | Tf | 3.79 | 1.26 | 4.47 | 0.68 | 1.29 | 5.42 | 4.08 | 1.32 | 1.21 | 5.91 |
| | | BF | 0.02 | 0.01 | 0.01 | 0.00 | 0.09 | 0.22 | 0.01 | 0.03 | 0.01 | 0.05 |
| 04 | *Carduus accanthoides* | Tf | 0.59 | 0.95 | 0.79 | 0.38 | 0.40 | 0.44 | 0.22 | - | 0.46 | 1.00 |
| | | BF | 0.05 | **1.01** | 0.01 | 0.00 | 0.17 | - | 0.00 | - | 0.04 | 0.32 |
| 05 | *Colchicum speciosum* | Tf | 1.54 | 1.19 | 2.46 | 1.66 | 1.67 | 0.68 | 1.00 | 1.11 | 1.48 | 1.10 |
| | | BF | 0.08 | 0.04 | 0.02 | 0.02 | 0.43 | 0.08 | 0.02 | 0.11 | 0.03 | 0.08 |
| 06 | *Fagus orientalis* | BF | 0.01 | 0.07 | 0.00 | 0.00 | 0.50 | - | 0.00 | - | 0.09 | 0.05 |
| 07 | *Carpinus betulus* | BF | 0.04 | 0.01 | 0.00 | 0.00 | 0.19 | - | 0.01 | 0.01 | 0.02 | 0.02 |
| 07 | *Junperus communis* | BF | 0.01 | 0.01 | 0.00 | 0.00 | 0.14 | 0.03 | 0.00 | 0.00 | 0.03 | 0.01 |
| 08 | *Ilex spinigera* | BF | 0.00 | 0.25 | 0.00 | - | 0.23 | - | 0.00 | - | 0.01 | 0.62 |
| 09 | *Crataegus microphylla* | BF | 0.02 | 0.09 | 0.00 | 0.00 | 0.26 | 0.01 | 0.02 | - | 0.02 | 0.10 |
| 10 | *Quercus castaneifolia* | BF | 0.00 | 0.13 | 0.00 | 0.00 | 0.19 | 0.01 | 0.00 | - | 0.04 | 0.12 |
| 10 | *Sorbus torminalis* | BF | 0.00 | 0.13 | 0.00 | 0.00 | 0.17 | 0.02 | 0.00 | | 0.03 | 0.28 |

[1] BF, bioaccumulation factor, (ratio of metal concentration in shoot to that in soil)
[2] TF, translocation factor, (ratio of metal concentration in shoot to that in root)

## Discussion

The goal of this survey was a primary assessment of local plant species useful for phytoremediation. Phytoremediation technique allows the soil to be decontaminated by successive cropping and harvesting. Metal enriched plants can be disposed of as hazardous material or, if economically feasible, can be recycled.

The only plant that exhibited the characteristics of hyper-accumulators was *S. byzantina*, that accumulated a significant amount of Pb in its above-ground tissues (1797 mg $kg^{-1}$), which met the criteria of Pb concentration in dry shoot matter exceeding 1000 mg $kg^{-1}$. Pb is the most common soil contaminant. Hyper-accumulators of Pb are rare mainly owing to the low solubility of Pb for plant uptake from soil (McGrath et al., 2002). Even in contaminated sites, the uptake of Pb by plants are low and mostly less than 50 mg $kg^{-1}$ (Cunningham et al., 1995 cited in McGrath et al., 2002).

*S. byzantine* had the TF value exceeding one (4.84), however, the BF value was less than one (0.12), which could be due to the high level of Pb in the soil (15472 mg $kg^{-1}$). Besides, the soil was alkaline, which rendered significant quantities of elements in the soil unavailable for plant uptake. Metals are more soluble in acidic conditions than in alkaline conditions (Wang, Angle, Chaney, Delorme, & Reeves, 2006; Zeng et al., 2011).

Given the high concentrations of heavy metals in the soil, the BF value is not significant and does not represent the significant ability of the plant to accumulate elements. In agreement with our research, Bech et al. (2012) reported *Senecio* sp. as a Pb hyper - accumulator with Pb concentration in its aerial part exceeding 4000 mg $kg^{-1}$, TF value exceeding one, and BF value of less than one; the average Pb concentration in the soil was more than 13000 mg $kg^{-1}$. Similar observations have been reported from other mining areas. In a study by Rotkittikhun et al. (2006) on an extremely contaminated Pb mining site in Thailand (Pb concentration in the range 6420–142400 mg $kg^{-1}$), 26 of the 48 plant species investigated accumulated more than 1000 mg $kg^{-1}$ of Pb in their shoots, 11 species had more than 1000 mg $kg^{-1}$ of Pb accumulated in their shoots as well as TF values of more than one, while none had a BF value exceeding one. Similar results were obtained by Bech, Duran, Roca, Poma, Sánchez, Barceló, et al. (2012) who did not regard the BF value as a reliable index in the case of soils with extremely high levels of metals; in their study, *Plantago orbignyana* accumulated a significant amount of Zn (8290–11560 mg $kg^{-1}$), with TF value of more than one, although the BF value was less than one mainly owing to the high Zn concentration in the soil (30565 mg $kg^{-1}$).

Additionally, according to Reeves (2006), plants that accumulate remarkable quantities of metals and whose BF values are less than one are interesting candidates for phytoremediation; by contrast, plants that accumulate insignificant quantities of metals but whose BF values are more than one owing to the low concentrations of metals in soil are not useful for phytoremediation. In our study, the only plant that has the BF value more than one is *Carduus accanthoides*. Although it has the BF value more than one (1.01) for Cd, the concentration of Cd in its shoot is so trivial (0.3 mg $kg^{-1}$), and the reason it had the BF value more than one is that the amount of Cd in the soil is negligible (0.3 mg $kg^{-1}$) too. Therefore, the BF value is not a good representative of the extraordinary accumulation of heavy metals in such plants.

## Conclusion

The *S. byzantina* plant species has the potential to function as a Pb hyper-accumulator owing to its significant capacity for the uptake and transfer of Pb to aerial parts as well as its ability to grow in the presence of toxic metals. One of the main limitations of our work was lack of replicate samples, thus further study is necessary to thoroughly assess the entire potential of *S. byzantina*. Our future work will entail additional investigation into this plant in controlled environments too to ensure that it exhibits healthy growth and to determine any symptoms of phytotoxicity owing to the high quantities of heavy metals.

## References

Baker, A.J.M. 1981. Accumulators and excluders -strategies in the response of plants to heavy metals. Journal of Plant Nutrition, 3(1-4), 643-654.

Bech, J., Duran, P., Roca, N., Poma, W., Sánchez, I., Barceló, J., Poschenrieder, C. 2012. Shoot accumulation of several trace elements in native plant species from contaminated soils in the Peruvian Andes. Journal of Geochemical Exploration, 113,106-111.

Bech, J., Duran, P., Roca, N., Poma, W., Sánchez, I., Roca-Pérez, L., and Poschenrieder, C. 2012. Accumulation of Pb and Zn in Bidens triplinervia and Senecio sp. spontaneous species from mine spoils in Peru and their potential use in phytoremediation. Journal of Geochemical Exploration, 123, 109-113.

Boojar, M.M.A., and Tavakkoli, Z. 2011. Antioxidative Responses and Metal Accumulation in Invasive Plant Species Growing on Mine Tailings in Zanjan, Iran. Pedosphere, 21(6): 802-812.

Boulding, J.R. 1994. Description and sampling of contaminated soils: a field guide (2nd Edition ed.). New York: Routledge.

Bower, C.A., Reitemeier, R.F., and Fireman, M. 1952. Exchangeable cation analysis of saline and alkali soils. Soil Science, 73(4), 251-262.

Brooks, R.R. 1998. Plants that hyperaccumulate heavy metals: Cambridge: University Press.

Chehregani, A., Noori, M., and Yazdi, H.L. 2009. Phytoremediation of heavy-metal-polluted soils: screening for new accumulator plants in Angouran mine (Iran) and evaluation of removal ability. Ecotoxicol Environtal Safety, 72(5), 1349-1353.

Deng, H., Ye, Z.H., and Wong, M.H. 2004. Accumulation of lead, zinc, copper and cadmium by 12 wetland plant species thriving in metal-contaminated sites in China. Environmental Pollutant, 132(1), 29-40.

Dickinson, N. 2017. Phytoremediation. In Encyclopedia of Applied Plant Sciences (pp. 327-331).

Gee, G.W., and Bauder, J.W. 1986. Particle-size Analysis1. In A. Klute (Ed.), Methods of Soil Analysis: Part 1—Physical and Mineralogical Methods (pp. 383-411). Madison, WI: Soil Science Society of America, American Society of Agronomy.

Ghahreman, A. (1978-2003). Flora of Iran. Tehran: Research Institute of forest and rangelands.

Hesami, R., Salimi, A., and Ghaderian, S. M. 2018. Lead, zinc, and cadmium uptake, accumulation, and phytoremediation by plants growing around Tang-e Douzan lead–zinc mine, Iran. Environmental Science and Pollution Research, 25(9), 8701-8714.

Kabata-Pendias, A., and Mukherjee, A.B. 2007. Trace Elements from Soil to Human: Springer-Verlag Berlin Heidelberg.

Kabata-Pendias, A.E. 2001. Trace elements in soils and plants (third ed.). Boca Raton: CRC Press.

Kim, Y.-Y., Yang, Y.-Y., and Lee, Y. 2002. Pb and Cd uptake in rice roots. Physiologia Plantarum, 116(3), 368-372.

Koleli, N., Demir, A., Kantar, C., Atag, G. A., Kusvuran, K., and Binzet, R. 2015. Heavy Metal Accumulation in Serpentine Flora of Mersin-Findikpinari (Turkey) – Role of Ethylenediamine Tetraacetic Acid in Facilitating Extraction of Nickel, 629-659.

Mahdavian, K., Ghaderian, S.M., and Torkzadeh-Mahani, M. 2015. Accumulation and phytoremediation of Pb, Zn, and Ag by plants growing on Koshk lead–zinc mining area, Iran. Journal of Soils and Sediments, 17(5), 1310-1320.

McGrath, S.P., Zhao, J., and Lombi, E. 2002. Phytoremediation of metals, metalloids, and radionuclides. In Advances in Agronomy 75, 1-56, Academic Press.

Mohanty, M., Dhal, N. K., Patra, P., Das, B., and Reddy, P.S. 2010. Phytoremediation: a novel approach for utilization of iron-ore wastes. Rev Environ Contam Toxicol, 206, 29-47.

Mozaffarian, V. 2004. Trees and Shrubs of Iran. Tehran: Farhang Moaser.

Nelson, D.W., and Sommers, L.E. 1982. Total Carbon, Organic Carbon, and Organic Matter1. In A. L. Page (Ed.), Methods of Soil Analysis. Part 2. Chemical and Microbiological Properties (pp. 539-579). Madison, WI: American Society of Agronomy, Soil Science Society of America.

Reeves, R.D. 2006. Hyperaccumulation of trace elements by plants. In Phytoremediation of Metal-Contaminated Soils (pp. 25-52).

Rotkittikhun, P., Kruatrachue, M., Chaiyarat, R., Ngernsansaruay, C., Pokethitiyook, P., Paijitprapaporn, A., and Baker, A.J. M. 2006. Uptake and accumulation of lead by plants from the Bo Ngam lead mine area in Thailand. Environmental Pollution, 144(2), 681-688.

van der Ent, A., Baker, A.J.M., Reeves, R.D., Pollard, A.J., and Schat, H. 2013. Hyperaccumulators of metal and metalloid trace elements: Facts and fiction. Plant and Soil, 362(1/2), 319-334.

Wang, A.S., Angle, J.S., Chaney, R.L., Delorme, T.A., and Reeves, R.D. 2006. Soil pH Effects on Uptake of Cd and Zn by Thlaspi caerulescens. Plant and Soil, 281(1-2), 325-337.

Warncke, D.D. 1986. Analyzing greenhouse growth media by the saturation extract methode. HortScience, 21, 223-225.

Zeng, F., Ali, S., Zhang, H., Ouyang, Y., Qiu, B., Wu, F., and Zhang, G. 2011. The influence of pH and organic matter content in paddy soil on heavy metal availability and their uptake by rice plants. Environmental Pollution, 159(1), 84-91.