Plants Role in Reducing Heavy Metals from Polluted Soil Leachate

**ABSTRACT**

**Aims** In the past few decades, more attention has been paid to clean up soils polluted with heavy metals by plants. A serious problem in this way is the amount of heavy metals uptake by plants. This study was conducted to evaluate the effectiveness of 3 local plants of Mazandaran province, Iran, in reducing and controlling the soil’s heavy metals.

**Instrument & Methods** The removal amount of three heavy metals (lead, zinc and cadmium) by native plants (maize, velvetleaf and wild amaranth) was investigated in alkaline (pH=8) and acidic (pH=5) soils and also using three substances such as EDTA, ammonium citrate and phosphate. The concentrations of these metals in leachate were measured by using atomic absorption spectrometry method.

**Findings** Lead, cadmium and zinc levels in leachate in treatments with plants were less than unplanted ones. The concentrations of these metals in the produced leachate of treatments with acidic soils were higher than those with alkaline soils. In the treatments of soil polluted with additives, treatments containing ammonium phosphate and EDTA had the lowest and highest concentrations of heavy metals, respectively. Concentrations of these metals in treatments without plants were higher than those with plants.

**Conclusion** Increasing of soil pH is effective on stabilization of heavy metals in soil. Ammonium phosphate plays an important role in stabilizing and EDTA and ammonium citrate increase the mobility of lead, zinc and cadmium in soil and groundwater.

**Keywords** Metals, Heavy; Soil; Plants; Water Pollutants, Chemical
Introduction
One of the most important environmental concerns of the current world is soil pollution with heavy metals. The main sources of heavy metals include industrial activities such as mining, metallurgy and use of products containing lead such as colors, gasoline and pesticides \[1-3\]. Unlike organic compounds, heavy metals are undegradable in nature and can remain in the soils for decades and even centuries \[4, 5\]. Soils polluted with heavy metals can have long-term effects on environment and health \[6, 7\].

Most common methods for removing heavy metals from polluted environments including the use of physical and chemical methods in site and off site but these methods are complex, costly and time-consuming \[8, 9\]. Therefore, bioremediation methods are useful due to the characteristics such as simplicity, low cost and lack of complexity \[10, 11\]. Plants can play effective role on preventing the entrance of soil heavy metals into groundwater using various mechanisms such as absorption and accumulation of heavy metals in the underground or air body parts of plants and their stabilization and absorption on the surfaces and particles of soil through discharge of organic or inorganic compounds secreted from plants \[12-14\].

Most studies have been performed on the removal of lead and cadmium by using phosphorus-containing treatments such as artificial and natural apatite and hydroxyapatite \[2\], phosphate rock \[3, 4\] salts dependent on phosphate \[5\], di-ammonium phosphate \[6\], phosphoric acid and EDTA \[9\]. Zinc can combine with organic ligands through using hydroxides, carbonates, phosphates, sulfides, molybdate and several anions forming the complex \[7\]. Since the plants can play an effective role in preventing the infiltration of soil heavy metals into the underground waters, this study was conducted to evaluate the effectiveness of 3 local plants of Mazandaran province, Iran, in reducing and controlling the soil’s heavy metals.

Instrument & Methods
This descriptive cross-sectional study where done at 2004.

**Soil sampling and preparation:** Approximately 1000kg of unpolluted, intact and pristine soil of remote areas around Babol City, Iran, was collected and transferred to the Health Research Center of Tehran University of Medical Sciences and was kept in a special pool. Then, a certain amount of sample soil and agents that enhanced plant growth, which were a kind of a natural organic fertilizer, called peat as 10% in soil, were thoroughly mixed with solution prepared with specific concentrations of each heavy metals (50, 10 and 150ml/kg for lead, cadmium and zinc, respectively) using a concrete mixer machine and transferred into 10-liter plastic pots. Soil samples were saturated with moisture and close to natural soil by flooding with water to a depth of 2 to 3cm in each pot. All treatments containing polluted soil were released in the open air for more than 6 months to stabilize the soil elements and heavy metals and to close the artificial pollution conditions with natural ones (Figure 1).

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Ins</th>
<th>Agr</th>
<th>Hgw</th>
<th>Bkg</th>
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<tbody>
<tr>
<td>Size (%)</td>
<td></td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>Clay</td>
<td>19</td>
<td>25.5</td>
<td>20.5</td>
<td>27.5</td>
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<tr>
<td>Silt</td>
<td>46.6</td>
<td>41</td>
<td>44</td>
<td>43</td>
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<tr>
<td>Sand</td>
<td>34.5</td>
<td>33.5</td>
<td>35.5</td>
<td>29.5</td>
</tr>
<tr>
<td>Cation exchange capacity (meq/100gr)</td>
<td>11-15.5</td>
<td>18-23.5</td>
<td>14-16.5</td>
<td>23-27</td>
</tr>
<tr>
<td>pH</td>
<td>6.5-7.5</td>
<td>7.1-7.4</td>
<td>7-7.3</td>
<td>6.8-7.2</td>
</tr>
<tr>
<td>Organic carbon (mg/kg)</td>
<td>41-53</td>
<td>22.5-29</td>
<td>25-36.6</td>
<td>18.5-21</td>
</tr>
</tbody>
</table>

**Planting, growing and harvesting of plants:** After preparing the seeds of plants, they were transferred into the small plots and the displacement and planting of seedlings into the 10-liter plastic pots were made after their growing. Normally, 4 to 5 seedlings were planted per pot. Preparation and transport of soil and planting the plants were done in each of the treatments as completely random block design. Then the plants were planted on soils polluted with lead, cadmium and zinc in acidic (pH=5) and alkaline (pH=8) conditions. The locations and lineup of each treatment were alternatively changed every week for preparing the same spatial and environmental conditions (the sun, the temperature and humidity, rainfall and wind direction). Irrigation was manually carried out at least 3 times a week.
Determination of heavy metals in leachate: Samples were collected in good quality screw capped high density pre-sterilized polypropylene bottles, each 1000ml capacity, labeled properly and analyzed in laboratory for trace metals by Atomic Absorption Spectrometer (AAS). Preservation and analysis of leachate samples were based on Standard Methods proposed by American Public Health Association [15]. Nitric acid was added and evaporated to 10ml. Then it was transferred to a 125ml conical flask. 5ml of conc. Nitric acid and 10ml of Perchloric acid (70%) were added. Then it was heated gently, till white dense fumes of HClO4 appeared. The digested samples were cooled at room temperature, filtered through Whatmann No. 1 filter paper and transferred to 50ml balloons then reached the volume with distilled water in order to prepare each sample resulted from leachate out of pots. To measure the concentrations of lead, cadmium and zinc in the samples, Perkin-Elmer Model 603 atomic absorption spectrophotometer (Perkin-Elmer Company; USA) was used in analysis laboratory of Faculty of Science of Tehran University. The sensitivity of the used device was about 1mg/l with using a flame photometer (Perkin-Elmer Company; USA) and it was up to 1 microgram per liter in the case of using graphite furnace. The wavelength to read lead, cadmium and zinc, was set at 283, 228 and 213nm, respectively.

Statistical analysis: Statistical analysis for multiple comparisons was performed with one-way ANOVA (with a significance level of 95%) using SPSS 16 software.

Findings
The mean concentrations of Pb, Cadmium and zinc in leachate produced from planted and unplanted treatments in free of additives and polluted alkaline soils (pH=8) and acidic soils (pH=5) were significantly reduced. A significant difference was seen between the mean concentrations of Pb, Cd and Zn in the treatment and control groups. The mean concentrations of Pb, Cd and Zn in leachate produced from acidic soils were significantly more than alkaline soils in samples with and without plants (Figure 2).

Figure 2) Mean concentrations of lead, cadmium and zinc in leachate derived from A: alkaline soils (pH=8); and B: acidic soils (pH=5)
Figure 3) Mean concentrations of lead (A), cadmium (B) and zinc (C) in leachate resulted from cultures containing additive chemicals.

The mean concentration of lead in leachate generated from the samples with EDTA, ammonium citrate and ammonium phosphate were significantly different between samples without plants and samples with plants. The lead concentration was minimal in leachate produced from soil containing ammonium phosphate in samples with and without plants. In addition, there was a statistically significant difference between the mean concentrations of lead in leachate resulted from samples containing EDTA with treatments including ammonium citrate and phosphate (p<0.05). There was a significant difference between the mean concentrations of lead in treatments containing ammonium citrate and phosphate in samples with and without plants (Figure 3A).
The highest and lowest concentrations of cadmium in leachate of case and control samples were concerned the treatments containing EDTA and ammonium phosphate, respectively and significantly different (p<0.05). The mean concentration of cadmium in leachate from soils with ammonium citrate in samples with and without plants showed that difference was significant (p<0.05). Difference between mean concentrations of cadmium in leachate of soils containing ammonium phosphate was not statistically significant (Figure 3B).

The mean concentration of zinc in leachate derived from the soils containing EDTA in samples free from plants was 73.2mg/l and in samples with plants was 52.5mg/l. This difference was statistically significant (p<0.05). According to the results, there were significant differences between the mean concentrations of zinc in leachate of soils with EDTA and soils with ammonium citrate and phosphate in unplanted samples (Figure 3C).

**Discussion**

Today, the extensive researches have been performed on the use of chemical chelating agents to increase the availability and mobility of lead adsorbed on the soil surface [13]. The results of this study indicated that the use of the chemical elements and compounds causes to increase the mobility and transport of various heavy metals in the soil depths and finally to pollute the groundwater. On the other hand, many researchers have found that despite the use of chemical chelating agents in some soils, metal concentrations are poor in groundwater of these areas [4]. However, another study performed on sandy soils polluted with lead demonstrated that the groundwater pollution with high levels of lead was due to the use of EDTA [1, 4].

These results suggest that the different characteristics of the soil of studied regions in terms of physical and chemical properties such as the size and percentage of clay, the CEC, percentage of organic carbon and soil acidity were very effective on keeping and stabilization of lead in soil and also on preventing of its movement into the groundwater and on its pollution control [16]. The mean concentration of lead was the lowest one in leachate of treatments with ammonium phosphate. One of the main reasons of this, in addition to the physical and chemical characteristics of the studied soils was the effect of soil phosphate material on lead compounds and producing insoluble compounds of pyromorphite and chloropyromorphite lead in soil in this case. There was a significant difference between the mean concentrations of lead in treatments containing EDTA in case and control groups (p<0.05), while this difference in treatments with ammonium phosphate was not statistically significant.

The results of studies have shown that the extent of groundwater pollution in polluted areas with sandy soils is higher than clay soils [17, 18]. Also, the extent of groundwater pollution was double in acidic soils with mentioned conditions [19]. Studies conducted by researchers around the world have shown that cadmium in soil can absorb and stabilize through phosphor and its compounds on the surface or within the soil particles [20].

The role of physical and chemical properties of the studied soil must be considered important on stabilizing and adsorption of cadmium on soil surfaces. The high concentration of zinc in leachate generated from control treatments was mainly due to the increase of solubility and availability of zinc compounds within the soil [21]. Therefore, this metal came out of soil with leachate after passing through the pores of the soil. Scientists have conducted extensive research on the effects and roles of vegetation covers on the various pollution controls including groundwater pollution control. It is known that the performance of different chelating agents was depended to the type of plant and heavy metals [10]. Our results showed that soil conditions (physical and chemical properties) were had an important role on stabilization and control of pollutions in soil and leachate.

Few plants (just 3) have been used for investigation of plant role in the immobilization of heavy metals through to underground water in the study which is a limitation. It is necessary to use some big plants and trees for investigation of cleanup of contaminated soils with heavy metals and the other pollutants.
Conclusion

Increasing of soil pH is effective on stabilization of heavy metals in soil. Ammonium phosphate plays an important role in stabilizing and EDTA and ammonium citrate increase the mobility of lead, zinc and cadmium in soil and groundwater.

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