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Growth and metal accumulation ability of plants in soil polluted with Cu, Zn and Pb

Dalė Pečiulytė,

Jūratė Repečkienė,

Loreta Levinskaitė,

Albinas Lugauskas

Institute of Botany, Žaliųjų ežerų 49, LT-08406 Vilnius, Lithuania

Algirdas Motuzas

Lithuanian University of Agriculture, Studentų 11, LT-53361 Kaunas-Akademija, Lithuania

Igoris Prosyčevas

Kaunas University of Technology, K. Donelaičio 73, LT-44029 Kaunas, Lithuania

The article draws on the search of plants able to grow in metal-contaminated soil and accumulate them in their biomass. The soil samples were collected from 10–20 cm and 25–56 cm deep layers and saturated with a mixture of Cu, Zn and Pb acetates to a full sorption capacity under laboratory conditions. Maize (*Zea mays* L.) and vetch (*Vicia sativa* L.) were grown in vegetative pots for three weeks. A negative effect on the length of shoots and roots of maize and vetch was observed, especially in the soil from of 25–56 cm deep layers, which is not rich in humus. However, in a 10–20 cm layer of metal-treated soil vetch roots grew well, and a significant increase of their biomass (up to 128.57%) was established. The biomass of plant shoots under the effect of heavy metals decreased by 63.78–79.21% (maize) and 14.47–92.57% (vetch) in the different soil layers. The results demonstrated that the maize plants growing in contaminated soils had accumulated higher Cu contents (338 mg kg^{-1}), while vetch plants accumulated a higher Zn content (365 mg kg^{-1} dry soil). *Zea mays* L. could be important for remediation of metal-contaminated soils.

Key words: lead, copper, zinc, plants, soil remediation

INTRODUCTION

Soils contaminated with heavy metals cause many environmental and human health problems calling for an effective technological solution. Many sites around the world remain contaminated, because it is expensive to clean them up by available technologies. Phytoremediation is considered to be an innovative, economical, and environmentally compatible solution for remediation of heavy metal contaminated sites (Thompson, 1995; Comis, 1996; Lombi et al., 2001; Wang et al., 2003; Bennet et al., 2003; O'Connor et al., 2003). Heavy metals may be bound or accumulated by particular plants, which may increase or decrease the mobility and prevent the leaching of heavy metals into groundwater. Growing plants can help to reduce heavy metal pollution. The advantage of this technique is evident as the cost of phytoremediation is much less than the traditional *in situ* and *ex situ* processes; plants can be easily monitored to ensure proper growth; and valuable metals can be reclaimed and reused through phytoremediation. The metals most commonly associated with phytoremediation are lead, cadmium, zinc, nickel, or radioactive isotopes such as uranium or cobalt (Comis, 1996; Lombi et al., 2001). Plants in metal-contaminated soils have been used only since the 1970s (Cunningham, Lee, 1995). Specifically, several subsets of metal phytoremediation have been developed. They include: (1) phytostabilization, in which plants stabilize the pollutants, thus rending them harmless; (2) phytoextraction, in which heavy metal hyperaccumulators, high-biomass, metal-accumulating plants and appropriate soil amendments are used to transport and concentrate metals from the soil into above-ground shoots which are harvested with conventional agricultural methods; (3) phytovolatilization, in which plants extract volatile metals from soil and volatilize them from the foliage (Cunningham et al., 1995).

Plants play an important role in all subsets of phytoremediation; however, it is necessary to use plants that could tolerate high levels of heavy metals. There are known some tolerant plants such as *Polygonum hydropiper* L., *Rumex acetosa* L. (Wang et al., 2003), *Lolium perenne* cv Elka (O'Connor et al., 2003), *Brassica juncea* (L.) Cern. (Bennet et al., 2003), *Thlaspi caerulescens* J. Presl, C. Presl, *Zea mays* L. (Lombi et al., 2001) or *Vetiveria zizanioides* (L.) Nash. ex Small (Greenfield, 1989). Vetiver grass, due to its unique morphological and physiological characteristics, is known for its effectiveness in erosion and sediment control, in addition to its tolerance of heavy metal contamination (Grimshaw, 1989; Trough, Kaker, 1998). Plants called hyperaccumulators are preferred, because they take up 100 times the concentration of metals over other plants (Cunningham et al., 1995). They accumulate toxic metals through their roots and transport them to the stems or leaves. Researchers hope that these metal-scavenging plants, called hyperaccumulators, could be grown in contaminated soils and harvested like hay (Lombi et al., 2001; Bennet et al., 2003; O'Connor et al., 2003). The metal could then be recovered and recycled when burned and the ash collected (Comis, 1996). In addition, the plant root system and its growth rate are very important factors which may improve the remediation procedure.

The aim of the investigation was to search for the plants possible to grow in metal-contaminated soil and accumulate metals in their biomass.

CONDITIONS AND METHODS

Moraine sandy loam on clay loam *Calcari-Endohypogleyic* (*LVg-n-w-*cc) was selected for the investigation. The experimental plot (10 × 10 m) was 350 m away from the roadside of the Vilnius–Kaunas–Klaipėda highway (Kaunas distr., Giraitė). Soil samples taken from 10–20 cm and 25–56 cm layers were used for the investigation. Soil from 10–20 cm was dark greyish with yellow shade (10YR 4/2), fluffy dampish sandy loam with a medium-crumby structure, and soil from 25–56 cm layer was brownish yellow (10TR 5/6), hard dampish sandy light loam with a medium-nutty structure. The soil pH was 6.9 (Lugauskas et al., 2005). Under laboratory conditions soil samples from each layer were treated with a mixture of Zn, Pb and Cu acetate solutions (1 g of each metal/l) until full sorption capacity was achieved. Control treatment was performed by wetting soil with distilled water in the same amount as a metal solution.

Measurements of metals were conducted using an atomic absorption spectrometer (Perkin–Elmer M403). Soil samples for detection of the total metal content were treated with HF, HNO_3 acids. The content of heavy metals in the soil treated with the metals and the control soil is given in Table 1.

Table 1. **Content of copper, lead and zinc in the soil before phytoremediation**

Soil layer	Variants of trial	Concentration (mg/kg)		
		Cu	Pb	Zn
10–20 cm	Control	4.80	7.80	2.10
10–20 cm	Saturated with metals	980.0	545.20	929.40
25–56 cm	Control	5.60	9.10	6.10
25–56 cm	Saturated with metals	875.5	560.40	805.80

For the accumulation of metals from soil, maize (*Zea mays* L.) and vetch (*Vicia sativa* L.) were used. Plants were grown in the plastic, 0.5 l capacity vegetative pots (five seeds in each pot in three replicates) for three weeks. The plants were watered as often as it was needed to maintain the constant moisture level. Stem height, root length, shoot and root weight of each plant grown were determined. After that plant biomass was prepared for atomic sorption spectrophotometry, and the content of the metals accumulated in dry biomass was estimated.

The data are presented as the mean and standard deviation of three replicates.

RESULTS AND DISCUSSION

The background metal contamination of the soil used in the current investigation was low (Table 1), however, the ability of soil to accumulate heavy metals from a mixture of Zn, Pb and Cu acetate solutions was high. After full sorption capacity of soil was achieved, the concentration of heavy metals in the samples increased 125–196, 60–62 and 134–464 times for Cu, Pb and Zn, respectively. The sorption capacity of the soil samples taken from 10–20 cm and 25–56 cm layers was very similar, but plant growth characteristics differed significantly.

After three weeks of growth in soil saturated with metals, a negative effect on the length of shoots and roots of maize and vetch was observed. The average height, length and green biomass of shoots and roots is shown in Table 2. The growth of plants was particularly weak in the 25–56 cm soil layer, which was not rich in humus. The maize roots were 28.0 cm in the metal-saturated soil, *versus* 145.0 cm in the control soil. The length of vetch roots was similar in metal-treated and control soil variants, but a significant increase of their biomass in the 10–20 cm layer was observed (9.6 g in the polluted soil *versus* 4.2 g in the control soil). The reason for such difference was the high level of lead accumulation by vetch roots (results are not presented).

The biomass of the plant seedlings was significantly smaller in the metal-contaminated soil in comparison with control. The biomass of the aerial parts of the plants grown in the soil saturated with metals decreased by 63.78% and 14.47% (maize and vetch, respectively) in the 10–20 cm layer and by 79.21% and 92.57% (maize and vetch, respectively) in the 25–56 cm layer (Fig. 1).

Visual evidence of metal toxicity to maize plants was present in each variant of the polluted soil (Fig. 2). The plants grew better in the 10–20 cm layer soil, which was rich in humus. It could be supposed that a high organic matter content improved the plants' resistance to heavy metals and their growth in metal-polluted soil.

Table 2. **Growth of vetch (*Vicia sativa* L.) and maize (*Zea mays* L.) plants in soil from two layers saturated with Cu, Pb and Zn and control soil (average of 15 replicates: 5 plants × 3 pots)**

Plants	Soil samples	Shoot height, cm	Root length, cm	Shoot weight*, g	Root weight, g
Maize plants	10–20 cm layer (control)	31.2 ± 5.4	169.4 ± 41.8	69.3 ± 13.8	35.9 ± 16.0
	10–20 cm layer (saturated with metals)	15.6 ± 4.8	29.2 ± 10.4	25.1 ± 6.7	25.1 ± 6.1
	25–56 cm layer (control)	22.5 ± 3.1	145.0 ± 47.6	38.0 ± 10.5	34.6 ± 6.9
	25–56 cm layer) (saturated with metals	5.8 ± 2.3	28.0 ± 13.4	7.9 ± 3.0	15.1 ± 7.0
Vetch plants	10–20 cm layer (control)	45.3 ± 5.6	67.8 ± 26.0	31.8 ± 5.7	4.2 ± 2.6
	10–20 cm layer (saturated with metals)	38.3 ± 6.4	69.2 ± 17.2	27.2 ± 5.0	9.6 ± 2.6
	25–56 cm layer (control)	43.3 ± 6.2	13.4 ± 5.6	26.9 ± 6.8	5.0 ± 1.8
	25–56 cm layer (saturated with metals)	3.1 ± 2.3	10.0 ± 0	2.0 ± 1.7	1.0 ± 0.3

* Weight of green biomass.

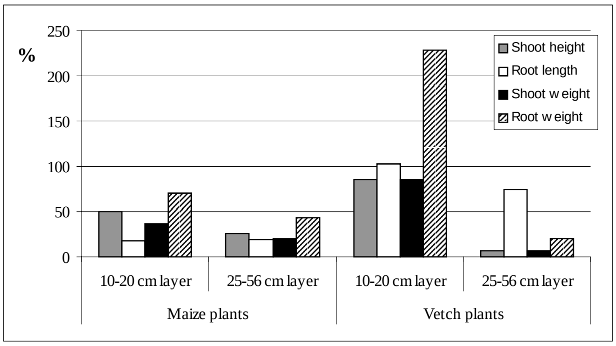

Fig. 1. Growth of maize and vetch plants in metal-contaminated soil from different layers (control – 100%)

a b c d

Fig. 2. Effect of heavy metals on the development of maize plants: control: 10–20 cm soil layer (a); 25–56 cm soil layer (b); soil saturated with metals: 10–20 cm soil layer (c); 25–56 cm soil layer (d)

The concentrations of metals in plants serve to indicate the metal contamination status of the site, and also reveal abilities of various plant species to take up and accumulate metals from polluted soil (Wang et al., 2003). As mentioned above, plants that accumulate 100 mg of metals per 1 kg of their biomass are preferred for phytoremediation (Cunningham et al., 1995). Substantial differences in the accumulation of Cu, Pb and Zn were observed between the two plant species investigated in the current study. Heavy metal uptake by maize and vetch plants is expressed as the metal content accumulated by plants during three weeks of growth in metal-contaminated and control soils and is shown in Fig. 3. The strategies for uptake heavy metal by the two plant species were different: over a short period time, the plants took up significant quantities of Pb, Cu and Zn, however, maize plants growing in contaminated soils had accumulated higher a Cu content (338 mg kg^{-1}), while vetch plants accumulated a higher Zn content (365 mg kg^{-1} dry wt). The content of metals accumulated in plants growing in the soil from the 10–20 cm layer containing organic mater did not differ from that in the plants growing on the soil from the 25–56 cm layer with a low nutrient concentration (Fig. 3). *Zea mays* L. growing in the soil from the upper layer had accumulated 340 mg kg^{-1} of Zn, and *Vicia sativa* L. growing in the same soil had accumulated high levels of both Zn and Cu (384 mg kg^{-1} and 327 mg kg^{-1}, respectively). It should be noted that plants growing in the soil from the 25–56 cm layer accumulated less Pb than plants growing in the upper 10–20 cm soil layer. Plant root weight also differed depending on the soil variant and Pb content accumulated by the plants, especially by vetch plants growing in the soil from the 10–20 cm layer (Figs. 1 and 3).

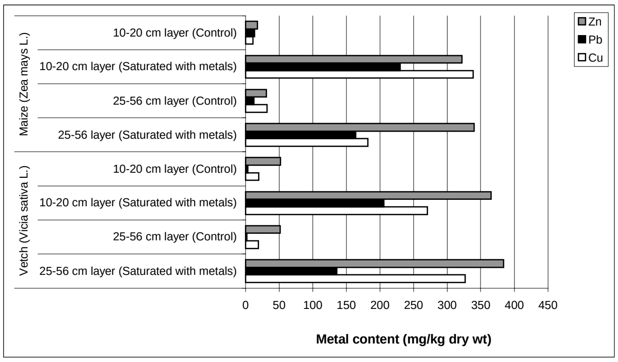

Fig. 3. Metal content in maize and vetch plants after three weeks of growth: control soil samples from two layers and soil samples saturated with Cu, Pb and Zn from two layers

The results demonstrate an enhanced metal accumulation potential of maize and vetch plants in polluted soil. These plants could be used for phytoremediation of soils contaminated by Cu and Zn, with a lower effect in soils contaminated with Pb, which affects the vetch root growth due to a high Pb accumulation in its roots. The mobility mechanism of heavy metals through soil presents a great interest to both environmental and soil scientists, mostly because of groundwater contamination through metal leaching (Alloway, 1990; Jensen et al., 2000; Dube et al., 2001). Metal mobility depends not only on the chemical properties of the metals but mostly on the physical and chemical properties of the soil, such as soil organic matter content, clay fraction content, mineral composition, pH, etc. Taken together, all these factors determine the binding ability of the soil. The reaction of plants to heavy metals in the soil from the different soil profiles in the current investigation varied. Despite the high metal pollution level, the ability of maize and vetch to accumulate Cu, Pb and Zn was evident. The most conspicuous characters of maize plants include their fast growth, high biomass, strong root system, and a high level of metal tolerance. Therefore, *Zea mays* L. could be important for stabilization of metal-contaminated soils. Field and glasshouse investigations conducted by Wang et al. (2003) on the responses of field pea, fodder vetch, maize, wheat and rapeseed to the heavy metals in soil showed that Pb accumulation was high in wheat, the highest concentration of Cu was found in fodder vetch and of Zn in maize. Our results, as well as the recent summary of heavy metal phytoremediation, indicate that the process could significantly decrease contamination. Such plants could be used for remediation, especially of soils rich in humus which provides for a good plant growth and reduces the toxic effect of metals.

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Dalė Pečiulytė, Jūratė Repečkienė, Loreta Levinskaitė, Albinas Lugauskas, Algirdas Motuzas, Igoris Prosyčevas

AUGALŲ GEBĖJIMAS AUGTI IR KAUPTI METALUS Cu, Zn IR Pb UŽTERŠTAME DIRVOŽEMYJE

Santrauka

Tyrimo tikslas – parinkti augalus, galinčius augti sunkiaisiais metalais užterštoje dirvoje ir kaupti juos savo biomasėje. Bandymams naudoto dirvožemio mėginiai paimti iš 10–20 ir 25–56 cm gylio ir prisotinti vario, švino ir cinko acetatų tirpalu. Vegetatyviniuose induose 3 savaites auginti kukurūzai (*Zea mays* L.) ir vikiai (*Vicia sativa* L.). Nustatyta, kad augalai silpniau augo dirvožemyje iš 25–56 cm gylio, nes jame yra mažiau humuso. Kukurūzų šaknys buvo 12–28% trumpesnės dirvožemyje, paveiktame metalais, lyginant su kontroliniu dirvožemiu. Vikių šaknys dirvožemyje su metalais augo gerai, jų biomasė buvo didesnė 128,7%, palyginti su kontrole. Kukurūzų antžeminės dalies svoris metalais užteršto dirvožemio kai kuriuose sluoksniuose sumažėjo 63,7–79,2%, o vikių – 14,5–92,6%. Nustatyta, kad kukurūzai sukaupia daugiau Cu (365 mg kg^{-1}), o vikiai – Zn (365 mg kg^{-1} sauso dirvožemio). Dėl gebėjimo sukaupti sunkiuosius metalus, stiprios šaknų sistemos, greito augimo kukurūzai gali būti naudojami variu, cinku ir švinu užterštam dirvožemiui apvalyti.

Raktažodžiai: švinas, varis, cinkas, augalai, dirvožemio remediacija