



RESTORATION OF CONTAMINATED SOIL SITES WITH THE AID OF FAST GROWING TREES

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Soil is basic means of agricultural production and subsequently the source of food for human population. Soil degradation and contamination is concerned of an issue worldwide. Several remediation techniques have been developed and applied to restore contaminated land and to keep soil properties on the sufficient level for the food production. Success of designed procedure is affected by metal presence and level of contamination. In experiments to extract metals from moderately and heavily metal-contaminated sites were investigated along with procedure to study the influence of organic fertilizers application on biomass production and a heavy metal uptake as well. The ability to accumulate risk elements in plant tissues has been proved, cadmium showed the best performance. Furthermore, a significant influence of biomass production on the total uptake of risk elements was proved. Organic fertilization had an adverse effect on the metal uptake. Extremely contaminated soils cannot directly be remediated by plants itself; they mainly need sufficient chemostabilization treatment before planting of trees.

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Introduction

Soil contamination with heavy metals is a major environmental problem nowadays and affect not only the soil quality but also several other environmental parameters.¹ Restoration of contaminated soils using conventional technologies requires considerable economic resources. For this reason, an interest has developed to have cost effective decontamination techniques. Phytoremediation is one of them. This method provides acceptable economic and environmental aspects for cleaning up contaminated soils.² Phytoremediation is methods using plants for restoration of main functions of contaminated soils and is established on several principles: stabilization methods like phytostabilization and phytoimmobilization or removal methods as phytoextraction, phytodegradation and phytovolatilization.³ Significant role in development of remediation methods plays selection of appropriate plant species for phytoremediation.⁴ Adriano (2001)⁵ categorized plants into several groups according to their ability to accumulate risk elements: metal excluders, indicators and accumulators (metallophytes). Metallophytes are plant species that able to grow in contaminated areas and accumulate heavy metals in aboveground biomass. Nevertheless, these plants are able to produce less biomass than other species.⁶ Therefore, appropriate group of plants for phytoremediation techniques are fast growing trees that accumulate relatively lower amounts of heavy metals but provide a high biomass production. Generally higher

removal of risk elements by fast growing trees and utilization of their biomass production provide economic return of contaminated land.⁷ According to pot experiments of Vysloužilová et al.⁸ willows planted in two soils differing in level of contamination confirm their ability to accumulate Zn and Cd in plant tissues. All tested willow clones planted in highly contaminated Fluvisol from the alluvium of the Litavka River suffer from chlorosis, partial leaf fall and reduction in the yield of aboveground biomass. Nevertheless, willow clones are able to provide high biomass production and accumulate substantial amounts of Cd in aboveground biomass in medium contaminated Cambisol.

The aims of this study were to evaluate the effects of two phytoremediation techniques: 1) phytoextraction in medium contaminated soil and 2) chemophytostabilization in highly contaminated soil.

First, we investigated ability of selected clones of willows and poplars to accumulate Cd and Pb in their branches grown on moderately contaminated soil in sewage sludge and control treatments in field conditions and second, the effect of alkaline and phosphorus additives to stabilize Cd and Pb in heavily contaminated soil and observe changes in yield and in Cd and Pb content in willow in model pot conditions.

Materials and Methods

Soil samples.

Two soils differing in level of contamination and soil type were used in our study. Both contaminated soils are located near the Příbram town and this region belongs to the most polluted areas in the Czech Republic.⁹ 1) Weakly acidic soil

“Podlesi” with medium contamination was tested in the field experiment and was characterized as follows: modal Cambisol, CEC 166 mmol kg⁻¹, C_{org} 4.1 %, 8.3 mg Cd kg⁻¹, 218 mg Zn kg⁻¹, 1214 mg Pb kg⁻¹. At the study site, the mean annual precipitations are 700 mm and mean annual temperature is 6.5 °C (Příbram meteorological station). 2) Slightly acidic soil “Litavka”, highly contaminated, was used in the pot experiment and was characterized as follows: Fluvisol, CEC 55 mmol kg⁻¹, C_{org} 3.6 %, 53.8 mg Cd kg⁻¹, 6172 mg Zn kg⁻¹, 3305 mg Pb kg⁻¹.

Field experiment

The field experiment in “Podlesi” was started in year 2008. The each treatment (control; sewage sludge, 7.5 kg m⁻² soil) used in the experiment was established in four replications. The plot of the each treatment was 7.5 m × 3.9 m. The experimental plots were planted with cuttings of fast growing trees from family *Salicaceae* (genus *Populus* – poplars and genus *Salix* – willows) (*S. schwerinii* × *S. viminalis* – VB1; *S. smithiana* – VB2) and two clones of poplars (*P. nigra* × *P. maximowiczii* – TP1; *P. nigra* – TP2) were used in each treatment. Planting distance between each 20 cm long cutting was 1.3 m × 0.25 m. After four years of planting in February 2012, branches (8 plant samples from each clone for each treatment) were harvested, measured, checked for fresh and dry biomass (dried at 60°C), ground and decomposed by dry-ash procedure.¹⁰ Total Cd and Pb contents were determined by inductively coupled plasma with optical emission spectroscopy (ICP-OES) (Varian VistaPro, Varian, Australia).

Pot experiment

In pot experiment, two different groups of soil additives (alkaline additives, phosphorus additives) were used as the metal stabilization agents in highly contaminated soil “Litavka”. From alkaline additives were chosen highly soluble quick lime and poor soluble dolomite, from phosphorus additives fast P release superphosphate and slow P release rock phosphate. At the beginning of the experiment in 2010, air-dry soil was treated with 0.3 g N, 0.16 g P, and 0.4 g K per kg of the soil. All soil additives (quick lime – 7.3 g kg⁻¹ soil, dolomite – 21.6 g kg⁻¹ soil, superphosphate – 4.2 g kg⁻¹ soil and rock phosphate – 2.9 g kg⁻¹ soil) corresponding to individual treatments were thoroughly mixed with the experimental soil and than one 20 cm long cutting of willow (*S. smithiana* – VB2) was planted in each pot (containing 5 kg soil).

Four replications were used for each treatment (control, quick lime, dolomite, superphosphate, and rock phosphate). The pots were kept in an outdoor weather-controlled vegetation hall. After five months of plantation leaves and twigs were harvested, checked for fresh and dry biomass, ground and decomposed by dry procedure as well as soil samples were collected and analyzed. Soil samples were extracted with 0.01 mol L⁻¹ CaCl₂ (1:10 w/v) for 6 hours.¹¹ Plant-available Cd and Pb concentrations in soil extracts and total plant Cd and Pb contents were determined by ICP-OES.

Data analyses.

The total element contents in plants, dry biomass and plant-available element concentrations in soil from individual treatments in field or pot experiments were evaluated by ANOVA (STATISTICA 9.0 software).

Results

Field experiment

Tested clones of fast growing trees planted in medium contaminated soil “Podlesi” showed high potential to accumulate Cd (Fig. 1). Both willow clones significantly accumulated more Cd compared to poplar clones. The highest amount of Cd accumulated willow clone VB1 in control treatment (54.2 mg kg⁻¹) and the lowest amount of Cd poplar clone TP1 (16.3 mg kg⁻¹). All tested clones in sewage sludge treatment accumulated less Cd compared to control treatment.

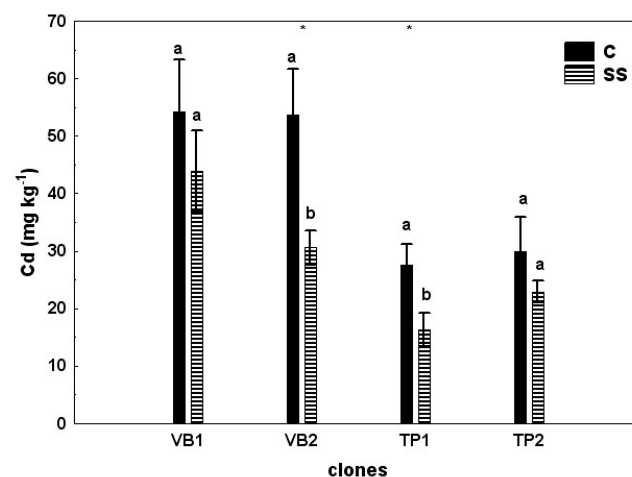


Figure 1. Cd content in branches of willow and poplar clones planted in medium contaminated soil treated with different treatments (C – control, SS – sewage sludge). Error bars represent standard error of the mean (SE). Using Tukey post-hoc test, treatments with the same letter are not significantly different within each clone.

Low mobile Pb was accumulated in branches of fast growing trees less compared to Cd (Fig. 2). Poplar clones accumulated higher Pb contents than Cd ones. The highest amount of Pb accumulated poplar clone TP1 in control treatment (34.4 mg kg⁻¹). The lowest amount of Pb accumulated poplar clone TP2 in sewage sludge treatment (16.8 mg kg⁻¹). Poplar clone TP2 was in Pb accumulation ability similar as willow clones. As in the case of Cd, fast growing trees planted in control treatment accumulated more Pb compared to sewage sludge treatment.

Biomass production is one of the significant parameters for evaluating ability of plants for phytoextraction. Decreasing trend in biomass production was in order TP1>VB2>TP2>VB1, see Fig. 3. Nevertheless, a decrease in biomass production was significantly different only between poplar clones TP1 and TP2.

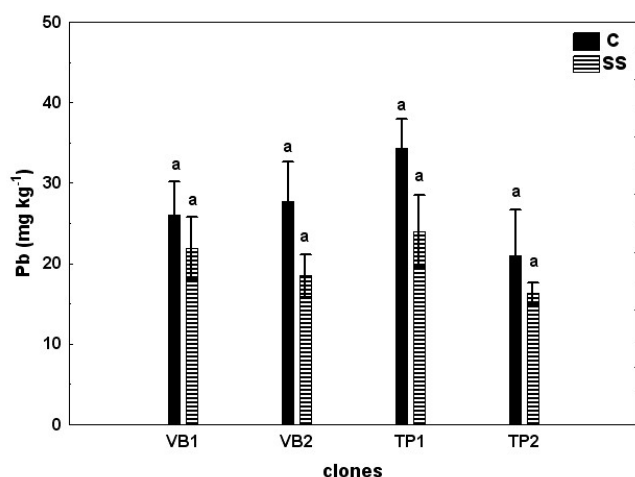


Figure 2. Pb content in branches of willow and poplar clones planted in medium contaminated soil treated with different treatments (C – control, SS – sewage sludge). Error bars represent standard error of the mean (SE). Using Tukey post-hoc test, treatments with the same letter are not significantly different within each clone.

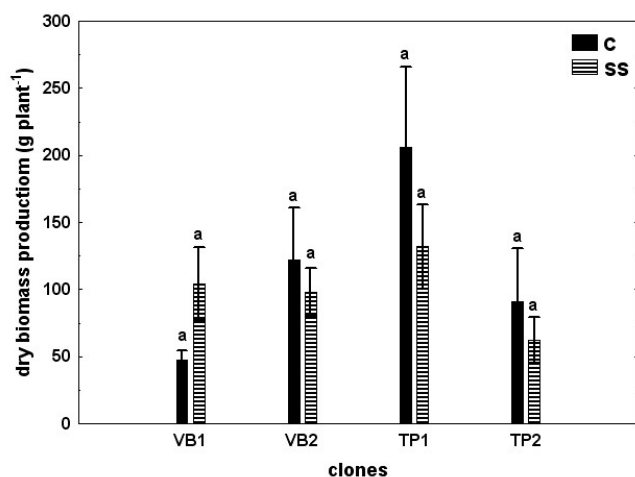


Figure 3. Dry biomass of willow and poplar clones planted in medium contaminated soil treated with different treatments (C – control, SS – sewage sludge). Error bars represent standard error of the mean (SE). Using Tukey post-hoc test, treatments with the same letter are not significantly different within each clone.

Biomass production of fast growing trees was higher in control treatment compared to sewage sludge treatment. It is interesting because it was conversely to previous years. It can be explained by several effects: 1) deficiency of nutrients for growth of fast growing trees in sewage sludge treatment after four years of start of field experiment, 2) competition with overgrown weeds, especially docks and 3) partial mortality of fast growing trees in sewage sludge treatment. In field conditions is very difficult to identify specific effects.

Total removal of risk elements plays the most important role in phytoremediation and is calculated by multiplication of plant element content and dry aboveground biomass. Figure 4 represents removal of Cd and figure 5 removal of Pb by plants. From both graphs, is obvious effect of biomass production on removal of both elements.

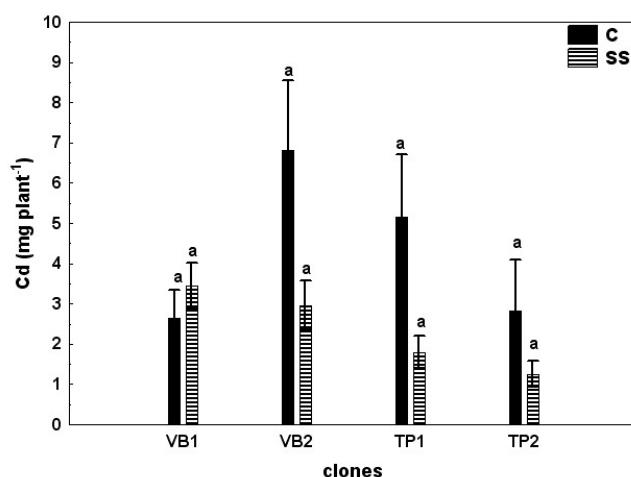


Figure 4. Removal of Cd by selected willow and poplar clones planted in medium contaminated soil treated with different treatments (C – control, SS – sewage sludge). Error bars represent standard error of the mean (SE). Using Tukey post-hoc test, treatments with the same letter are not significantly different within each clone.

Total removal of Cd was the highest by willow clone VB2, although the highest Cd content in branches was in willow clone VB1. The order of decreasing removal of Cd followed the order: VB2>TP1>VB1>TP2 with significantly different between clones VB2 and TP2.

Pb content in branches decreased in the order: TP1>VB1>VB2>TP2. Nevertheless, removal of Pb decreased in order: TP1>VB2>TP2>VB1. We can conclude that removal of Pb has similar trend as biomass production.

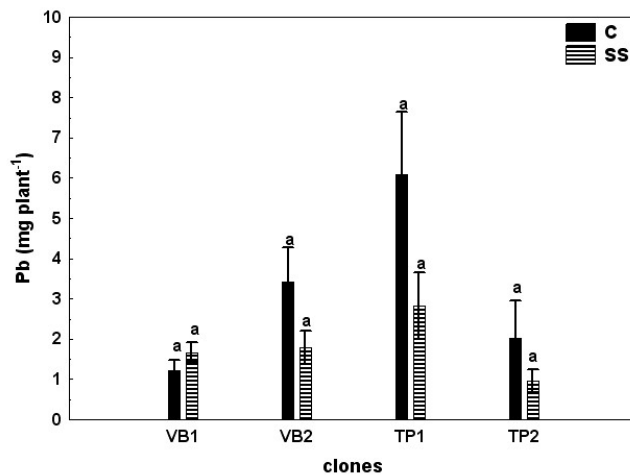


Figure 5. Removal of Pb by selected willow and poplar clones planted in medium contaminated soil treated with different treatments (C – control, SS – sewage sludge). Error bars represent standard error of the mean (SE). Using Tukey post-hoc test, treatments with the same letter are not significantly different within each clone.

Fast growing trees planted in sewage sludge treatment accumulated less Cd and Pb compared to control treatments. It can be related with low biomass production and low content of both elements in aboveground biomass which can be probably affected by higher sorption surface caused by organic matter derived by sewage sludge. Nevertheless, differences among treatments were not significantly different.

Pot experiment

Addition of both alkaline additives (quick lime, dolomite) significantly reduced plant-available Cd concentrations compared to the control (see Table 1). In the case of phosphorus additives, Cd immobilization was established only in the rock phosphate treatment. Slight increase in plant-available Pb concentrations after all soil additives applications was not significantly different.

Table 1. Mean plant-available Cd and Pb concentrations (\pm standard error of the mean; SE) in highly contaminated soil treated with different treatments (control, quick lime, dolomite, superphosphate, and rock phosphate) after five months of planting willow clone VB2. Using Tukey post-hoc test, treatments with the same letter are not significantly different.

Treatment	Element (mg kg^{-1})	
	Cd	Pb
control	3.11 ± 0.09^a	0.14 ± 0.04^a
quick lime	0.27 ± 0.04^b	0.26 ± 0.05^a
dolomite	2.36 ± 0.06^c	0.23 ± 0.08^a
superphosphate	3.51 ± 0.07^d	0.19 ± 0.05^a
rock phosphate	2.75 ± 0.10^c	0.24 ± 0.10^a

Quick lime, dolomite and superphosphate applications increased Cd content in leaves compared to the control but not significantly (see Fig. 6). In contrast, rock phosphate decreased Cd content in leaves. From comparison of plant-available Cd concentrations in soil, we can conclude that there is an indirect correlation after alkaline additives application and direct correlation after phosphate additives application. Otherwise, there was no effect of tested additives on decrease Cd content in twigs compared to the control.

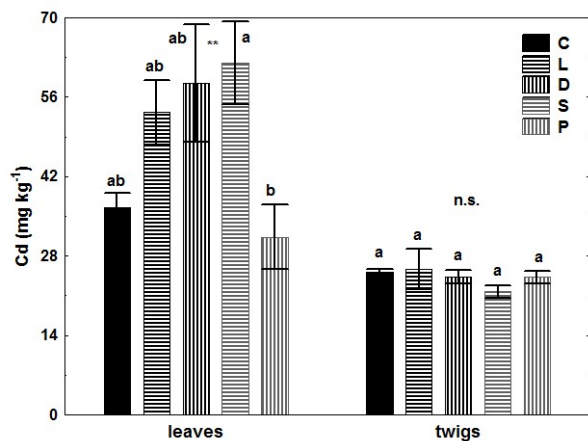


Figure 6. Cd content in leaves and twigs of willow clone VB2 planted in highly contaminated soil treated with different treatments (C – control, L – quick lime, D – dolomite, S – superphosphate, P – rock phosphate). Error bars represent standard error of the mean (SE). Using Tukey post-hoc test, treatments with the same letter are not significantly different within each organ of plant.

In most of treatments (quick lime, dolomite, and rock phosphate application), an increase in Pb content in leaves and twigs was not significantly different compared to the control (see Fig. 7) as in the case of plant-available Pb concentrations in soil (see Table 1).

We observed a decreasing trend in dry production of willow after all additives application compared to the control (see Fig. 8). Nevertheless, in the cases of quick lime

and superphosphate applications, a decrease in dry aboveground biomass production was not significantly different.

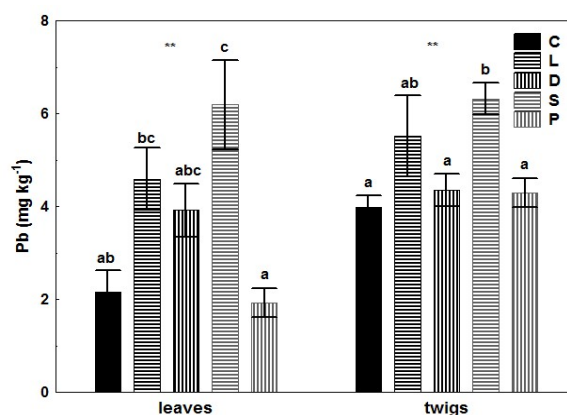


Figure 7. Pb content in leaves and twigs of willow clone VB2 planted in highly contaminated soil treated with different treatments (C – control, L – quick lime, D – dolomite, S – superphosphate, P – rock phosphate). Error bars represent standard error of the mean (SE). Using Tukey post-hoc test, treatments with the same letter are not significantly different within each organ of plant.

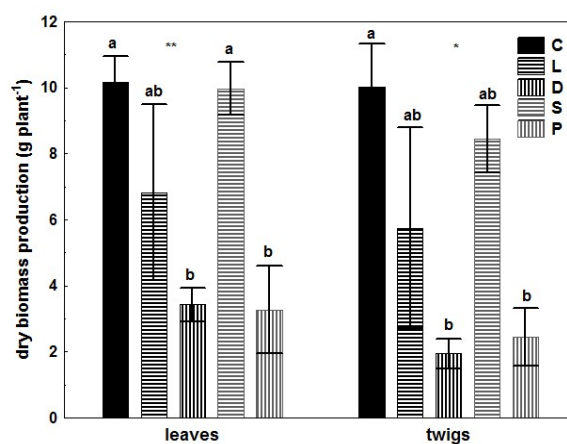


Figure 8. Dry biomass production of willow clone VB2 (leaves, twigs) planted in highly contaminated soil treated with different treatments (C – control, L – quick lime, D – dolomite, S – superphosphate, P – rock phosphate). Error bars represent standard error of the mean (SE). Using Tukey post-hoc test, treatments with the same letter are not significantly different within each organ of plant.

Discussion

Field experiment.

Cadmium is mobile element occurring in large portion in exchangeable and plant-available fractions.¹² Both willow clones accumulated more Cd in branches as compared to Pb; similar results were obtained by Vysloužilová et al.¹³ Willows confirmed their ability as Cd accumulators, and it was also observed by Dickinson and Pulford.¹⁴ On the other hand, poplars confirmed their ability as Pb accumulators, and this was also observed in study of Fischerová et al.¹⁵ Lead is low mobile element. Only small portion of Pb in soil is occurred in easily mobilisable fraction. In the case of Pb, efficiency of phytoextraction was very low in medium contaminated soil “Podleší”; similar results see also in Komárek et al.¹⁶

Cd and Pb contents in branches of fast growing trees were negatively affected after sewage sludge application. According to Chaney et al.¹⁷ fertilization and other agronomic activities necessary for high biomass production can negatively affect risk elements uptake. Liming and organic fertilization can reduce mobility and availability these elements by plants. Ability of organic matter to bond risk elements can reduce phytoextraction in low and moderately contaminated soils. Quality of organic matter, especially ratio of fulvic and humic acids can play an important role in reduction of risk elements uptake by plants. Fulvic acids create soluble chelate and humic acids with high ability to bond heavy metals create insoluble complexes especially in acid conditions.¹⁸ Cadmium in low content is bond in sewage sludge on fulvic acids. On the other hand, Pb is bonded especially in humic acids.¹⁹

High biomass production of fast growing trees is important medium for phytoextraction.²⁰ In our study was confirmed that risk element contents in branches and especially yield of biomass play an important role in removal of Cd and Pb by fast growing trees; it was also reported by Hammer et al.²¹ In this study, total removal of elements from soil with high pH has similar trend as biomass production.

Pot experiment.

The highest decrease in plant-available Cd concentrations after quick lime application is connected with high increase in soil pH; see other studies.^{22, 23} On the other hand, an increase in plant-available Cd concentrations after superphosphate applications is probably because of decrease of soil pH, observed also by Wang et al.²⁴ There is no positive effect on decrease of plant-available Pb concentrations after all soil additives application because of generally low Pb mobility in soil.²⁵

There is an increasing trend in accumulation Cd and Pb contents in leaves of willow clone VB2 after first vegetation period. Nevertheless, the results were not significantly different. So it is necessary to monitor these clones of willow in the same conditions for more vegetation periods for equitable evaluating soil additives treatments. We can conclude that all tested additives (quick lime, dolomite, superphosphate and rock phosphate) negatively affected dry biomass production of willow clone VB2 after the first vegetation period in pot experiment. The results of dry biomass production were surprising. Stabilization of plant-available concentration in soil especially after quick lime application would give good possibilities for health growth of willows⁸. The results were probably affected by premature leaf fall because of Fe deficiency. Therefore, it will be necessary to treat willows by Fe fertilizers during the next vegetation periods.

Conclusion

Results of both experiments confirm that high biomass production plays a crucial role in total removal of both toxic elements from contaminated soils. Application of organic matter did not affect biomass production for longer period in

field experiment but limited element uptake, with addition of additives into extremely contaminated soil was not successful in improvement biomass production, however decreased metal availability in soil.

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Reference

- ¹Santos F. S., Magalhaes M. O. L., Mazur N., Amaral Sabrinho N. M. B., *Sci. Agric.*, **2007**, *64*, 506.
- ²Nascimento C. W. A., Xing B., *Sci. Agric.*, **2006**, *63*, 299.
- ³Wenzel, W.W., Lombi, E., Adriano, D., *Biogeochemical processes in the rhizosphere: Role in phytoremediation of metal-polluted soils. In heavy metal stress in plants - from molecules to ecosystems. Prasad, N.; Hagemeyer, J. (Ed) Heidelberg, Springer Verlag, 1999.*
- ⁴Salt, D.E., Blaylock, M., Kumar, P.B.A.N., Dushenkov, V., Ensley, B.D., Chet, I., Raskin, I., *Nat. Biotechnol.*, **1995**, *13*, 468.
- ⁵Adriano, D. C., *Trace elements in terrestrial environments: Springer Verlag, New York, 2001.*
- ⁶Frančová K., Macek T., Demnerová K., Macková M., *Chem. Listy*, **2001**, *95*, 630.
- ⁷Laureysens I., De Temmerman L., Hastir T., Van Gysel M., Ceulemans R., *Environ. Pollut.*, **2005**, *133*, 541.
- ⁸Vysloužilová M., Tlustoš P., Száková J., *Plant, Soil Environ.*, **2003A**, *49*, 542.
- ⁹Šichorová K., Tlustoš P., Száková J., Kořínek K., Balík J., *Plant, Soil Environ.*, **2004**, *50*, 525.
- ¹⁰Miholová D., Mader P., Száková J., Slámová A., Svatoš Z., *Fresenius J. Anal. Chem.*, **1993**, *51*, 256.
- ¹¹Novozamsky J., Lexmond T.M., Houba V.J.H., *Int. J. Environ. Anal. Chem.*, **1993**, *51*, 47.
- ¹²Száková, J., Tlustoš P., Pavlíková D., Balík J., *Chem. Pap.*, **2003**, *57*, 167.
- ¹³Vysloužilová M., Tlustoš P., Száková J., Pavlíková D., *Plant, Soil Environ.*, **2003B**, *49*, 191.
- ¹⁴Dickinson N. M., Pulford I. D., *Environ. Int.*, **2005**, *31*, 609.
- ¹⁵Fischerová Z., Tlustoš P., Száková J., Šichorová K., *Environ. Pollut.*, **2006**, *144*, 93.
- ¹⁶Komárek M., Tlustoš P., Száková J., Chrástný V., *Environ. Pollut.*, **2008**, *151*, 27.
- ¹⁷Chaney R. L., Li Y-M, Angle J. S., Baker A. J. M., Reeves R. D., Brown S. L., Homer F. A., Malik M., Chin M., *Improving metal-hyperaccumulators wild plants to develop commercial phytoextraction systems: Approaches and progress. In Phytoremediation of Contaminated Soil and Water, eds. N. Terry and G.S. Bañuelos, CRC Press, Boca Raton, 1999.*
- ¹⁸Tlustoš P., Pavlíková P., Balík J., *Mechanismus příjmu rizikových prvků rostlinami a jejich hromadění v biomase. Česká zemědělská univerzita v Praze, 2006.*
- ¹⁹Hanč A., Tlustoš P., Száková J., Balík J., *Chem. Listy*, **2007**, *101*, 807.
- ²⁰Pulford I. D., Watson C., *Environ. Int.*, **2003**, *29*, 529.
- ²¹Hammer D., Kayser A., Keller C., *Soil Use Manage.*, **2003**, *19*, 187.

²²Mühlbachová G., Tlustoš P., *Plant Soil Environ.*, **2006**, 52, 345.

²³Vondráčková, S., Hejzman, M., Tlustoš, P., Száková, J., *Polish J. Environ. Stud.*, **2012**, In press.

²⁴Wang, B., Xie, Z., Chen, J., Jiang, J., & Su, Q., *J. Environ. Sci.*, **2008**, 20, 1109.

²⁵Miretzky, P., & Fernandez-Cirelli, A., *Environ. Chem. Lett.*, **2008**, 6, 121.

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