

Chromium, Nickel, Cadmium, and Lead Accumulation in Maize, Sunflower, Willow, and Poplar

Lada Kacálková¹, Pavel Tlustoš^{2*}, Jiřina Száková²

¹Department of Biology, University of Hradec Králové, Faculty of Science, Rokitanského 62, 500 03 Hradec Králové, Czech Republic

²Department of Agroenvironmental Chemistry and Plant Nutrition, Faculty of Agrobiolgy, Food and Natural Resources, Czech University of Life Sciences Prague, Kamýcká 129, 165 21 Praha 6 – Suchbát, Czech Republic

Received: 30 November 2012

Accepted: 14 January 2014

Abstract

Our study investigated the accumulation of chromium, nickel, lead and cadmium by maize (*Zea mays* L.), sunflower (*Helianthus annuus* L.), willow (*Salix x smithiana* Willd.), and poplar (*Populus nigra* L. x *P. maximowiczii*), and the relationship between the contaminants in soil and in plants. The experiment was performed in contaminated soil (former waste incineration plant) at the Hradec Králové (Czech Republic) site. Plant and soil samples were collected from three plots with different risk element contents (higher amounts of Cd, Cu, Hg, Zn, Cr, Ni, and Pb). The total and available soil metal concentrations in soil were investigated. Only a low portion of risk elements were available for plants (6% Ni, 14% Cd, 1.3 % Pb, and less than 1% of Cr). Chromium, nickel, and lead showed a similar trend to element accumulation where the highest amount was found in plant roots, higher in herbs than in trees (6.83 mg Cr·kg⁻¹, 5.04 mg Ni·kg⁻¹, and 7.76 mg Pb·kg⁻¹). The highest cadmium concentration was found in leaves of willow (1.87 mg Cd·kg⁻¹) and roots of willow (3.05 mg Cd·kg⁻¹). The correlation between the concentration of risk elements in soil and in plants was the highest in the case of lead reaching up to R= 0.89. Results also indicated that translocation of Cr, Ni, Cd, and Pb from roots to aboveground biomass of willow and poplar was low (89-98% of risk elements was retained in roots). The highest translocation from plant roots to aboveground biomass of maize and sunflower was found in the case of Cd and Pb (57 and 83% of Cd, 56 and 76% of Pb). The behaviour of these elements concerning soil and plants differed among fields with unknown history of contamination and type of contaminants.

Keywords: chromium, nickel, lead, cadmium, plants

Introduction

Heavy metal levels in the biosphere have accelerated rapidly since the onset of the industrial revolution, which poses major environmental problems, including the damages to the earth's surface and cultivated land pollution [1].

Although the main attention of concern is on risk elements such as cadmium, zinc, lead, mercury, etc., other elements such as chromium, nickel, etc. must also be taken into account.

Anthropogenic inputs of chromium include a range of industrial activities, alloys, chrome plating, dyes and pigments, textiles, leather tanning, and wood preserving [2]. These anthropogenic activities have led to widespread envi-

*e-mail: tlustos@af.czu.cz

ronmental Cr contamination, causing its increased bioavailability and biomobility. Soil Cr values range from 10 to 50 mg·kg⁻¹ dw [3]. Cr is toxic to higher plants at 100 mg·kg⁻¹ dw of plant content. Uptake and accumulation of Cr by various crops are well documented. Plant Cr contents vary between 0.006-18 mg·kg⁻¹ dw [4].

The generally accepted Ni values in plant tissues are between 0.5-5 mg·kg⁻¹ of dry weight (dw). Soil Ni contents vary between 5-150 mg·kg⁻¹ dw [5]. Anthropogenic activities further release Ni into the soil through various sources such as smelting, burning of fossil fuel, vehicle emissions, disposal of household debris, municipal and industrial wastes, metal mining, fertilizer application, and organic manures [6]. The overall uptake of Ni by plants depends on the concentration of Ni²⁺ in soil solution, plant metabolism, the acidity of soil or solution, the presence of other metals, and organic matter composition [7].

Owing to the rapid expansion of industrialization and the heavy use of chemical fertilizer, pesticides, and herbicides in agriculture, cadmium pollutants have been considered one of the most serious environmental problems worldwide [8]. Cadmium phytoaccumulation is an important research topic, as food and particularly vegetables constitute a major source of human exposure to Cd [9] and also because plants are, through phytoextraction, a potentially efficient means of remediating soils contaminated with this trace metal [10]. Availability of Cd to plants is regulated by pH, soil organic matter, and redox potential [11].

With rapid industrial development around the world, the inputs of Pb to agricultural soils have been occurring through the combustion of gasoline containing Pb additives; the fugitive emissions from nonferrous metal, the widespread uses of fertilizers, herbicides, and pesticides; and the additions of sewage sludge to the soil [12]. The background concentration of Pb in uncontaminated soil fluctuated within the range of 10-50 mg·kg⁻¹; however, in soil with low-level contamination, Pb concentration can be expected to range from 30 to 100 mg·kg⁻¹ [13]. Pb is not an essential element [14] and is considered relatively unavailable for living organisms due to immobilization in the soil and limited transport from roots to plants [15].

Plant species respond differently to heavy metal pollution. Although a relationship exists between heavy metal accumulation and plant tolerance, many plant species grow on contaminated soils and do not accumulate metals [16]. Gorchach and Gambus found that maize is the most resistant to increased heavy metals concentration in the soil, followed by wheat, barley, sunflower, and hemp [17]. Tree species have a range of characteristics that make them possible candidates for application in phytoremediation approaches. In particular, *Salix* spp. has been suggested as they are fast to propagate, achieve high annual biomass production, and generally possess a high tolerance against metal pollution [18, 19]. Besides hyperaccumulators there are plant species like *Salix viminalis* (which takes up a large portion of Cd and Zn), *Brassica juncea* (Pb), *Lolium perenne* (Pb), *Zea mays* (Pb), *Helianthus annuus* (Pb, Cu), or others, characterized by high content of heavy metals in biomass and good remediation capacity [20, 21]. The potential of

willow for phytoremediation has mainly been established through pot experiments or growth chamber experiments [22, 18]. Besides that, studies such as that of Nowack et al. has shown that the determination of metal bioavailability based on laboratory tests of sampled soils is not representative for field soils and in situ conditions [23].

The most important soil variable affecting the bioavailability of heavy metals (particularly Cd and Pb) is soil pH. The higher pH reduces the availability of Cd and Pb [24]. Some authors suggest that the available soil metal concentration predicts metal transfer from soil to crops better than does total metal concentration. Thus soil quality criteria for trace metals should be based on the bioavailable pool of the elements to ensure adequate environmental protection [9]. There are still paucity reports on soil-to-plant transfer factors based on the bioavailable risk elements in the research articles.

The aims of this investigation were as follows:

- (1) To assess and compare the heavy metal (Cd, Cr, Ni, and Pb) accumulation by high biomass production plants – maize (*Zea mays* L.), sunflower (*Helianthus annuus* L.) and fast-growing trees – willow (*Salix x smithiana* Willd.) and poplar (*Populus nigra* L. x *P. maximowiczii*) in soils contaminated with these elements
- (2) To find a relationship between the contaminants in soil and in plants at specific locations characterized by intensive and heterogeneous soil element contents.

Experimental Procedures

Sampling Site

Field experiments were conducted for three consecutive years in the vegetation periods (May-September) of 2007-09 at the area of a former waste incineration plant with heterogeneous soil contamination in a suburb of Hradec Králové, Czech Republic. History and inorganic and organic contamination of this area were described by Kacálková et al. [25] and Kacálková and Tlustoš [26]. Three plots with different risk element contamination were chosen for preparation of growing fields (signed – F3, F4, and F5 according to Kacálková et al. [25]). This was compared to other samples among three fields with elevated risk element contents having a higher risk elements content. Soil type is loamy Luvisol. The soil had unknown sources of contamination and high heterogeneity compared with other studies. The soil from F5 can be described as weakly acidic with a higher organic carbon content, while sampling fields F4 and F3 have a neutral pH with much lower organic carbon content. High soil Cox value at sampling field F5, could be the cause of uncontrolled storage of organic compounds and materials intended for burning. Neutral soil pH was found almost at all places with the exception of sampling field F5 where the experimental field shows acid soil reaction (pH CaCl₂ – 6.09).

Soil subsamples were taken from 0-20 cm depth within the circle of 3 m diameter. Samples were homogenized, dried at laboratory temperature (22°C) and passed through a 2 mm sieve. Plant residues were removed before sieving.

Each plot was planted out and sown with these experimental plants: willows – W (*Salix x smithiana* Willd.), poplars – P (*Populus nigra* L. x *P. maximowiczii*), maize – M (*Zea mays* L.) and sunflower – S (*Helianthus annuus* L.). Plants were treated ordinarily (watering, weeding) and fertilized by the addition of 30 g·m⁻² NH₄NO₃ for three vegetation periods. Then in autumn before senescence, these plants were harvested, aboveground biomass of fast growing trees and whole biomass of maize at first and second years, whole biomass of all plants at third year. Fresh plant biomass was weighed, separated to leaves and twigs (trees) and roots, leaves, stalks and seeds (maize and sunflower), and after drying at 60°C dry mass was measured.

Soil and Plant Analysis

All chemical analyses were provided in analytical laboratories of the Department of Agroenvironmental Chemistry and Plant Nutrition at Czech University of Life Sciences Prague in order to determine the total contents of risk elements. Total element concentrations in soil were determined in digests obtained by two-step decomposition as follows: 0.5 g of sample was decomposed by dry ashing in a mixture of oxidizing gases (O₂+O₃+NO_x) in an Apion Dry Mode Mineralizer (Tessek, CZ) at 400°C for 10 h; the ash was then decomposed in a mixture of HNO₃ + HF, evaporated to dryness at 160°C, and dissolved in diluted aqua regia [27].

For the determination of mobile portions of elements in soils, extraction with a 0.11 mol·l⁻¹ solution of CH₃COOH at a ratio of 1:20 (w/v) for 16 hours was applied [28]. Each extraction was provided in three replicates. For the centrifuging the extracts, a Hettich Universal 30 RF (Germany) device was used. The reaction mixture was centrifuged at 3,000 rpm for 10 minutes at the end of each extraction procedure, and the supernatants were kept at 6°C prior to measurement. Element concentrations in the digests and extracts were determined by ICP-OES (Varian, VistaPro, Australia).

Plant samples were decomposed using the dry ashing procedure as follows: An aliquot (~1 g) of the dried and powdered aboveground biomass or roots were weighed to 1 mg into a borosilicate glass test-tube and decomposed in a mixture of oxidizing gases (O₂+O₃+NO_x) at 400°C for 10 hours in Dry Mode Mineralizer Apion (Tessek, Czech Republic). The ash was dissolved in 20 ml of 1.5% HNO₃ (electronic grade purity, Analytika Ltd., Czech Republic) and kept in glass tubes until analysis [29]. Aliquots of the certified reference material RM NCS DC 73350 poplar leaves (purchased from Analytika, CZ) were mineralized under the same conditions for quality assurance of the total element contents in experimental plants.

The total contents of Cd, Cr, Ni, and Pb in soil and plant digests were determined by optical emission spectroscopy with inductively coupled plasma (ICP-OES) with axial plasma configuration, Varian, VistaPro, equipped with autosampler SPS-5 (Australia). Measurement conditions were for all lines: power 1.2 kW, plasma flow 15.0 l·min⁻¹, auxiliary flow 0.75 l·min⁻¹, and nebulizer flow 0.9 l·min⁻¹.

Hg content was determined by atomic absorption spectrometry using a single-purpose AMA-254 analyzer (Altec, Czech Republic). Exchangeable values of pH were measured in a 1:20 (w/v) 0.01 mol·l⁻¹ CaCl₂ soluble extract at 20±1°C, and active pH values in water extract (ratio 1:3, v/w) at 20±1°C. The used pH meter was WTW pH 340 i set.

Statistical Analysis

Statistical analyses were performed using the software Statistica 10 (analysis of variance – ANOVA, followed by the Tukey HSD test – $\alpha = 0.05$ for multiple comparisons) and Microsoft Office Excel 2007 (standard deviation).

Regression Analysis

Regression analysis (r value and regression equation) was performed using Microsoft Office Excel 2010. Correlation coefficients (r values) were evaluated according to Snedecor and Cochran (1967). Correlation coefficients at the 95% levels of significance for 9 degrees of freedom have to be larger than 0.602.

Results and Discussion

Soil Contamination

Total and available average contents of chosen risk elements in soils are given in Table 1. Regulation No. 13/1994 Collection of Laws of Ministry of the Environment provides maximum admissible risk element contents in soil as follows: 1 mg·kg⁻¹ Cd, 200 mg·kg⁻¹ Cr, 80 mg·kg⁻¹ Ni, and 140 mg·kg⁻¹ Pb. The cadmium content in the three soil samples exceeded the maximum soil content. Chromium did not exceed the admissible content. Nickel was in a higher concentration than the limit at one sampling point and so lead was at two sampling points. The highest concentration of the chosen risk elements was in the area of sampling point F5.

The total metal concentration was high, but only a limited portion of them was available for plants (6% Ni, 14% Cd, 1.3% Pb, and less than 1% of Cr). These results can be explained with the consideration of the chemical properties of the soils [25], especially heterogeneous soil contamination. Total soil heavy metal concentration is commonly used to indicate the degree of contamination, although extractable concentration provides a more suitable chemical evaluation of the amount of metals available for plant uptake [30]. The solubility of metals in the soil is predominantly controlled by the pH, the amount of metal, the cation exchange capacity, the organic carbon content, and the oxidation state of the mineral components [31]. The concentration of bioavailable soil Cd (in contrast to total Cd) is the key factor for uptake and in certain concentration intervals it may be proportional to accumulated Cd [32]. From our results, in the case of cadmium and lead, we found that increasing pH caused increasing available Cd and Pb content in soil (plot F5 with pH =

Table 1. Total and available (0.11 mol·L⁻¹ acetic acid extractable) metal concentration in soil (mg·kg⁻¹).

	Plot No.	Cr	Ni	Cd	Pb
Total	F3	40±4.3	31±3.3	2.0±0.5	44±3.2
	F4	39±1.7	23±2.1	1.6±0.02	193±7.4
	F5	141±5.2	122±8.4	6.8±0.6	167±9.3
Available	F3	<0.005	1.88±0.3	0.28±0.01	0.55±0.01
	F4	<0.005	1.55±0.1	0.23±0.1	0.44±0.001
	F5	<0.005	8.06±1.0	0.08±0.01	0.01±0.001

Table 2. Chromium concentration in plants (mg·kg⁻¹). The values marked by the same letter did not significantly differ at $\alpha = 0.05$ within individual columns, where small letters indicate the differences among the individual fields and capital letters indicate the differences in element contents within one plant.

	Maize			
	Roots	Stalks	Leaves	Grain
F3	4.08±3.68 ^{ba}	0.89±0.2 ^{aa}	1.16±0.45 ^{aa}	1.04±0.93 ^{aa}
F4	3.43±1.78 ^{ba}	0.75±0.31 ^{aa}	2.12±0.69 ^{ba}	0.5±0.25 ^{aa}
F5	3.12±1.26 ^{ba}	0.61±0.35 ^{aa}	1.08±0.73 ^{aa}	0.57±0.42 ^{aa}
	Sunflower			
	Roots	Stalks	Leaves	Grain
F3	2.99±0.45 ^{ca}	0.23±0.07 ^{aa}	1.23±0.66 ^{ba}	2.32±0.49 ^{cb}
F4	7.02±3.98 ^{db}	0.25±0.16 ^{aa}	1.14±0.84 ^{ba}	1.93±0.3 ^{cb}
F5	6.83±2.93 ^{db}	0.71±0.53 ^{aa}	0.52±0.18 ^{aa}	0.57±0.12 ^{aa}
	Willow			
	Roots	Stalks	Leaves	
F3	0.42±0.07 ^{ba}	0.51±0.45 ^{aa}	0.39±0.08 ^{aa}	
F4	0.89±0.47 ^{aa}	0.45±0.42 ^{aa}	0.46±0.37 ^{aa}	
F5	2.89±1.04 ^{ba}	0.34±0.18 ^{aa}	0.17±0.08 ^{aa}	
	Poplar			
	Roots	Stalks	Leaves	
F3	0.6±0.02 ^{aa}	0.15±0.05 ^{aa}	0.2±0.17 ^{aa}	
F4	1.06±0.12 ^{ba}	0.16±0.05 ^{aa}	0.67±0.73 ^{aa}	
F5	0.74±0.17 ^{aa}	0.31±0.26 ^{aa}	0.78±0.09 ^{aa}	

6.1 and plot F4 and F3 with pH = 7.4). This is in agreement with Shuman [33], who described that increasing pH reduced availability of the metals.

Contents of Chromium, Nickel, Cadmium, and Lead in Plants

All the plant species tested in the experiment were grown on contaminated soil on three sampling points with

different risk element concentrations in soil (F3, F4, and F5), showing no visible symptoms of toxicity. Tables 2-5 show risk element concentrations in particular parts of cultivated plants. Chromium, nickel, and lead showed the identical tendency, respectively, the highest amount was found in plant roots, higher in herbs than in trees (6.83 mg Cr·kg⁻¹, 5.04 mg Ni·kg⁻¹, and 7.76 mg Pb·kg⁻¹). These results corresponded to those of Angelova et al. [34]. They also found the highest amount of Pb, Zn, Cd, Cu in roots of flax, hemp, and cotton compared to aboveground part of plants. Higher ability to accumulate these elements showed sunflower compared to maize and grains of sunflower and maize contained similar concentrations like stalks. Cadmium accumulation in herbs was similar like Cr, Ni, and Pb. However the highest Cd content in trees was found in leaves (often significantly higher than in roots).

A significantly higher amount of chromium (Table 2) was found in sunflower roots (7.02 mg Cr·kg⁻¹). Lower Cr amounts in sunflower (*Helianthus annuus* L.) roots (3.21 mg·kg⁻¹) were found by Mani et al. in sewage-irrigated alluvial soils [35]. Our results are in agreement with Mohanty and Patra [36]. They used para grass (*Brachiaria mutica*) in situ experiment and Cr bioaccumulation in roots, which was nearly 1,000 times higher than in shoots. Leaves accumulated chromium in higher concentrations than stalks (from 0.17 to 1.23 mg Cr·kg⁻¹), significantly higher in the case of sunflowers. However, Sawidis et al. [37] found higher Cr accumulation in the bark of *Platanus orientalis* L. and *Pinus nigra* L. compared to the leaves. Higher concentrations of chromium in grains of maize and sunflower than in stalks was found (up to 2.32 mg Cr·kg⁻¹). The usual range for plants is 0.1-0.5 mg Cr·kg⁻¹ [6]. The Cr concentration values in plants from our experiment were often higher than normal range. Chromium concentration decreases in order: roots > leaves > stalks > grains in case of maize, roots > leaves > grains > stalks in case of sunflower, and roots > leaves > stalks in case of trees.

Nickel was translocated to stems and, especially, to leaves and grains (Table 3). A significantly higher amount of nickel was found in sunflower roots (5.04 mg Ni·kg⁻¹). However, willow leaves demonstrated the ability to accumulate this element in higher concentrations than in other tested plants – up to 3.88 mg Ni·kg⁻¹ in leaves. Fargašová and Beinrohr [38] also found higher nickel accumulation in the shoots than in the roots of *Salix alba*. Leaves of

Table 3. Nickel concentrations in plants ($\text{mg}\cdot\text{kg}^{-1}$). The values marked by the same letter did not significantly differ at $\alpha = 0.05$ within individual columns where the small letters indicate the differences among the individual fields and capital letters indicate the differences in element contents within one plant.

	Maize			
	Roots	Stalks	Leaves	Grain
F3	3.91±2.7 ^{ba}	0.85±0.28 ^{ba}	1.2±0.39 ^{ba}	0.94±0.63 ^{ba}
F4	4.09±2.15 ^{ca}	0.73±0.2 ^{ba}	1.13±0.23 ^{ba}	1.06±0.14 ^{ba}
F5	2.77±0.76 ^{ba}	0.79±0.13 ^{ba}	0.92±0.4 ^{ba}	1.12±0.48 ^{ba}
	Sunflower			
	Roots	Stalks	Leaves	Grain
F3	3.04±0.86 ^{ba}	1.32±0.5 ^{ba}	1.2±0.18 ^{ba}	1.92±0.06 ^{ba}
F4	4.04±1.53 ^{ba}	0.91±0.52 ^{ab}	1.34±0.56 ^{ba}	1.54±0.08 ^{ba}
F5	5.04±0.93 ^{ba}	1.36±0.2 ^{ac}	3.35±1.33 ^{bb}	1.89±0.27 ^{ab}
	Willow			
	Roots	Stalks	Leaves	
F3	3.06±0.21 ^{bb}	0.81±0.59 ^{ba}	1.93±1.44 ^{ba}	
F4	2.36±0.91 ^{ba}	0.94±0.2 ^{ba}	2.59±0.91 ^{ba}	
F5	3.1 ±0.52 ^{ba}	1.41±0.78 ^{ba}	3.88±0.57 ^{bb}	
	Poplar			
	Roots	Stalks	Leaves	
F3	0.83±0.09 ^{ba}	0.59±0.52 ^{ba}	0.6±0.43 ^{ba}	
F4	1.78±0.12 ^{bb}	0.51±0.14 ^{ba}	1.26±0.48 ^{ba}	
F5	0.98±0.22 ^{ba}	0.58±0.29 ^{ba}	1.1±0.43 ^{ba}	

Fraxinus excelsior accumulate higher Ni concentration – 27.04 $\text{mg}\cdot\text{kg}^{-1}$ [39]. Grains of sunflower accumulated 2.32 $\text{mg}\cdot\text{kg}^{-1}$. The accepted Ni values in plant tissues are in the range of 0.5-5 $\text{mg}\cdot\text{kg}^{-1}$ dw [6]. Our results corresponded with this study. The order of the plant parts according to diminishing Ni concentrations is: roots > leaves > grains > stalks in herbs and roots > leaves > stalks in trees.

Cadmium was found in higher concentrations in trees than in herbs (Table 4). Translocation from plant roots to leaves was higher in the case of trees. Cadmium transport to aboveground parts of *Populus deltoides* x *Populus nigra* also was declared by Fuzhong et al. [40]. Significantly higher concentration was in willow roots – 3.05 $\text{mg}\cdot\text{kg}^{-1}$ but only at one sampling site. Concentrations of this element were higher in leaves of willow and poplar than in roots at the next two areas. A significantly higher concentration in willow leaves was 1.97 $\text{mg}\cdot\text{kg}^{-1}$. The amount of uptaken cadmium in plant parts decreases in the order: roots > leaves ~ stalks > grains in herbs, roots > leaves > stalks in willow and leaves > stalks > roots in poplar.

Significantly higher amounts of lead were found in sunflower roots (9.76 $\text{mg}\cdot\text{kg}^{-1}$) and maize roots (8.45 $\text{mg}\cdot\text{kg}^{-1}$) (Table 5). Maize leaves contained up to 4.40 mg

Table 4. Cadmium concentrations in plants ($\text{mg}\cdot\text{kg}^{-1}$). The values marked by the same letter did not significantly differ at $\alpha = 0.05$ within individual columns where the small letters indicate the differences among the individual fields and capital letters indicate the differences in element contents within one plant.

	Maize			
	Roots	Stalks	Leaves	Grain
F3	0.73±0.2 ^{ba}	0.4±0.5 ^{ba}	0.12±0.1 ^{ba}	0.09±0.1 ^{ba}
F4	0.65±0.3 ^{ba}	0.18±0.1 ^{ba}	0.19±0.1 ^{ba}	0.16±0.3 ^{ba}
F5	0.62±0.4 ^{ba}	0.27±0.1 ^{ba}	0.18±0.1 ^{ba}	0.12±0.1 ^{ba}
	Sunflower			
	Roots	Stalks	Leaves	Grain
F3	0.55±0.3 ^{ba}	0.56±0.3 ^{ba}	0.66±0.3 ^{ba}	0.59±0.2 ^{ba}
F4	1.09±0.1 ^{ba}	0.39±0.1 ^{ba}	0.49±0.1 ^{ba}	0.31±0.1 ^{ba}
F5	0.59±0.1 ^{ba}	0.44±0.1 ^{ba}	0.74±0.3 ^{ba}	0.51±0.2 ^{ba}
	Willow			
	Roots	Stalks	Leaves	
F3	3.05±0.3 ^{bb}	1.29±0.3 ^{ba}	1.87±0.7 ^{ba}	
F4	0.72±0.3 ^{ba}	1.16±0.3 ^{ba}	1.97±0.4 ^{ba}	
F5	1.36±0.1 ^{ba}	1.35±0.3 ^{ba}	1.38±0.4 ^{ba}	
	Poplar			
	Roots	Stalks	Leaves	
F3	0.69±0.1 ^{ba}	0.94±0.3 ^{ba}	0.99±0.5 ^{ba}	
F4	0.77±0.1 ^{ba}	0.57±0.2 ^{ba}	1.03±0.2 ^{ba}	
F5	0.66±0.1 ^{ba}	0.94±0.3 ^{ba}	1.18±0.2 ^{ba}	

$\text{Pb}\cdot\text{kg}^{-1}$. Markert [41] suggested that uptake of Pb is probably passive and translocation from roots to other parts of the plant is low, but aerial deposition and foliar uptake may contribute significantly to leaf concentration. Common concentrations of Pb in plants are lower, than 10 $\text{mg}\cdot\text{kg}^{-1}$ [42]. Allen [43] considered 3 $\text{mg}\cdot\text{kg}^{-1}$ of Pb as a common natural level for plants. Lead levels in above-ground plant parts from our experiment did not exceed these values. The concentration of Pb in plant parts decreases in the order: roots > leaves > stalks > grains in herbs and roots > leaves > stalks in trees.

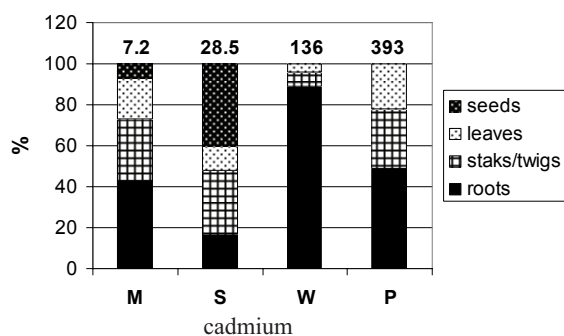
The Effect of Soil Element Content on Element Accumulation in Plants

In our experiment the relationship between total and available concentration of risk elements in soil and risk element concentrations in plants is observed (Table 6). Whatmuff [44], Sun et al. [45], Han et al. [46], and McBride [24] found that increasing concentrations of heavy metals in soil increased plant uptake. Regression analysis showed that the closest relationship (R value from 0.14 to 0.89) was found in the case of lead. R value, evaluated

Table 5. Lead concentrations in plants ($\text{mg}\cdot\text{kg}^{-1}$). The values marked by the same letter did not significantly differ at $\alpha = 0.05$ within individual columns where the small letters indicate the differences among the individual fields, and capital letters indicate the differences in element contents within one plant.

	Maize			
	Roots	Stalks	Leaves	Grain
F3	3.75±1.34 ^{cA}	0.57±0.14 ^{aA}	1.2±0.3 ^{aA}	0.16±0.02 ^{aA}
F4	8.45±5.9 ^{dB}	0.61±0.25 ^{aA}	1.51±0.61 ^{bA}	0.39±0.16 ^{aA}
F5	4.12±4.2 ^{cA}	3.4±4.04 ^{cB}	4.4±3.44 ^{cB}	1.07±0.83 ^{aA}
	Sunflower			
	Roots	Stalks	Leaves	Grain
F3	1.22±0.3 ^{bA}	1.1±0.58 ^{bA}	1.2±0.32 ^{bA}	0.66±0.09 ^{aA}
F4	2.33±2.05 ^{bA}	1.45±0.76 ^{bB}	1.27±0.37 ^{bA}	1±0.14 ^{bA}
F5	9.76±11.5 ^{dD}	3.8±0.62 ^{cA}	3.4±0.7 ^{cB}	0.3±0.06 ^{aC}
	Willow			
	Roots	Stalks	Leaves	
F3	0.83±0.09 ^{aA}	0.82±0.47 ^{aA}	1.01±0.45 ^{aA}	
F4	3.37±1.95 ^{cB}	0.78±0.5 ^{aA}	1.95±0.88 ^{bA}	
F5	2.96±0.63 ^{bA}	2.05±0.93 ^{bA}	1.95±0.92 ^{bA}	
	Poplar			
	Roots	Stalks	Leaves	
F3	1.39±0.28 ^{aA}	0.52±0.15 ^{aA}	0.51±0.28 ^{aA}	
F4	1.64±0.45 ^{bB}	0.57±0.28 ^{aA}	1.24±0.32 ^{aA}	
F5	1.33±0.22 ^{aB}	1.01±0.88 ^{aA}	1.62±0.86 ^{bA}	

according to Snedecor and Cochran [47], were significant in sunflower seeds and willow roots for total risk element contents in soil, leaves and seeds of sunflower, and twigs of willow and poplar for available risk element contents in soil. No significant correlation was found for other risk elements, except one r value for nickel and two values for chromium. The reason could be great heterogeneity of soil risk element contamination. Our results are partly in accord with Orroño et al. [48] in terms of pot experiment with



Pelargonium hortorum in a soil spiked with Cd, Cr, Cu, Pb, Ni, and Zn found out that soil-extractable Cr, Ni, and Pb were related to their concentration in plants, but there was no correlation between soil and plants for Cd, Cu, and Zn. Lead concentrations in plants were positively correlated with increasing total lead content in soil. However, the same concentration responded negatively to available content in soil. Available cadmium and total chromium were negatively correlated with increasing soil concentration.

For example, Vandecasteele et al. [49] reported that willow (*Salix fragilis* and *Salix viminalis*) foliar Cd concentrations were strongly correlated with soil Cd concentrations. In our experiment we found negative correlation in willow leaves for total Cd content in soil and positive correlation for available Cd content in soil with $R=0.6$ (the highest value for available Cd).

The unique element correlation to plants or between plants was not demonstrated. The reason may be different types of contamination and unknown sources of contamination.

Risk Element Removal by Plants

Relative removal of cadmium, lead, chromium, and nickel within three years at plot F5 (with highest amount of these risk elements) are displayed in Figs. 1 and 2. Metal removal is influenced by two factors – the content of the element in particular aboveground plant parts is the first one and the yield of aboveground dry biomass is the second. Biomass of investigated plants increased in the order: maize < sunflower < willow < poplar. Also, the elements removal followed the mentioned order, biomass plays more a important role in the risk element removal than element concentration.

The highest cadmium removal was found by roots of fast growing trees (from 49% to 89%). The reason for the highest risk element removal by roots of willow and poplar could be the higher yield of root biomass. Tree twigs removed higher parts of cadmium than leaves. Also, maize removed the highest amount of cadmium by roots (43%), and minimal was found by seeds (7%). However, a different situation was in the case of sunflower. Cadmium removal by seeds was 40% and by roots and leaves was only 17% and 12%. Cadmium concentrations in sunflower seeds correlated positively with seed weight.

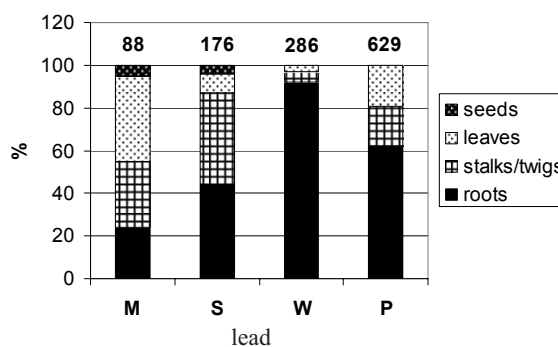


Fig. 1. Percentage removal of risk elements by herbs (with total removal in $\mu\text{g}/\text{plant}$). (M – maize, S – sunflower, W – willow, P – poplar)

Table 6. Results of regression analysis (R value) of total and available soil and plant element concentrations.

		Roots		Stalks/twigs		Leaves		Seeds	
		Total	Available	Total	Available	Total	Available	Total	Available
Cd	M	0.02	0.03	0.01	0.05	0.11	0.21	0.27	0.26
	S	0.42	0.46	0.01	0.09	0.38	0.43	0.17	0.28
	W	0.01	0.19	0.13	0.18	0.23	0.60	*	*
	P	0.33	0.39	0.30	0.39	0.01	0.05	*	*
Ni	M	0.33	0.32	0.01	0.01	0.31	0.31	0.12	0.12
	S	0.44	0.45	0.27	0.27	0.72	0.72	0.35	0.34
	W	0.11	0.12	0.47	0.47	0.50	0.51	*	*
	P	0.40	0.38	0.04	0.04	0.12	0.13	*	*
Pb	M	0.25	0.52	0.18	0.48	0.25	0.58	0.35	0.62
	S	0.20	0.49	0.35	0.87	0.37	0.89	0.14	0.72
	W	0.68	0.37	0.24	0.66	0.49	0.33	*	*
	P	0.18	0.19	0.16	0.37	0.30	0.67	*	*
Cr	M	0.10	*	0.31	*	0.33	*	0.14	*
	S	0.20	*	0.64	*	0.35	*	0.89	*
	W	0.83	*	0.12	*	0.48	*	*	*
	P	0.20	*	0.42	*	0.22	*	*	*

*not analyzed

(M – maize, S – sunflower, W – willow, P – poplar)

Higher root removal than aboveground biomass of trees and sunflower was recorded for Pb (92% – willow, 62% – poplar, 44% – sunflower). In our experiment, root and stalk Pb concentrations in sunflower were not correlated positively with the root and stalk weights. It is reported that the process of Pb taken up from soils by roots is strongly governed by plant factors, and different plant species own different properties in Pb uptake and distribution. Some are root-accumulator, storing Pb in the roots, transporting little to aboveground parts, and others are shoot-accumulators, storing greatly more Pb in the shoots [50].

Chromium and nickel were accumulated mainly in roots of all tested plants (98.7% Cr and 92% Ni by willow, 70% Cr and 66% Ni by poplar, 50% Cr and 43% Ni by maize, 65% Cr and 47% Ni by sunflower). Also, Shams et al. [51] reported that *Zea mays* is not an effective plant for Cr phytoextraction from soil. Higher amounts of Cr and Ni were removed by maize leaves compared to stalks and seeds (27% Cr and 23% Ni), and by sunflower stalks compared to leaves and seeds (17% Cr and 31% Ni).

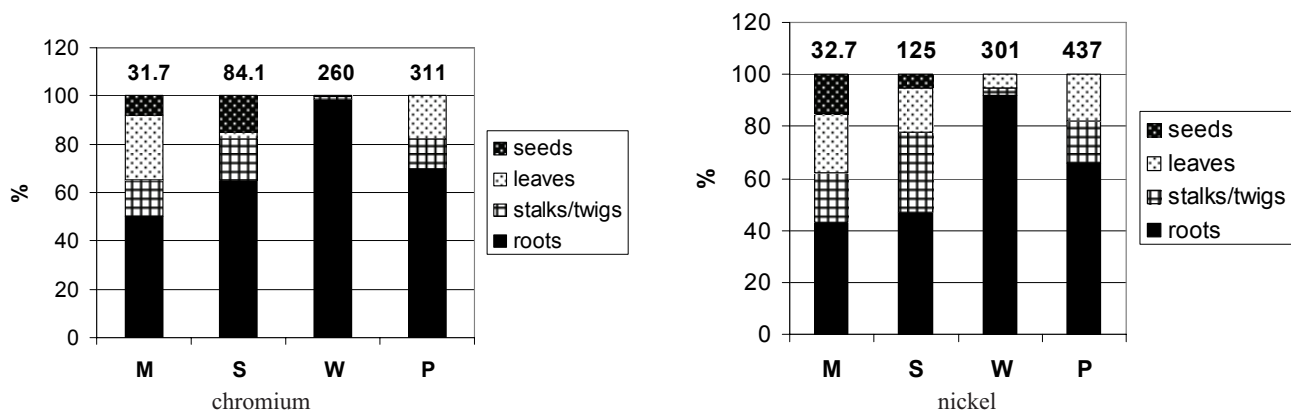


Fig. 2. Percentage removal of risk elements by trees (with total removal in µg/plant). (M – maize, S – sunflower, W – willow, P – poplar)

Acknowledgements

This research was supported by specific research project No. 2122/2011 from the University of Hradec Králové and GAAV IAA600110902.

References

1. GISBERT C., ROS R., DE HARO A. A plant genetically modified that accumulates Pb is especially promising for phytoremediation. *Biochem Biophys Res Comm* **303**, 440, **2003**.
2. KIMBROUGH D.E., COHEN Y., WINER A.M., CREE-LAM L., MABUNI C. A critical assessment of chromium in the environment. *Crit. Rev. Environ. Sci. Technol.* **29**, 1, **1999**.
3. SHANKER A.K., CERVANTES C., TAVERAC H.L., AVUDAINAYAGAMD S. Chromium toxicity in plants. *Environ. Int.* **31**, 739, **2005**.
4. ZAYED A.M., TERRY N. Chromium in the environment: factors affecting biological remediation. *Plant Soil* **249**, 139, **2003**.
5. KABATA-PENDIAS A., PENDIAS H. Trace Elements in Soils and Plants. CRC Press, LLC (Third Ed.); Boca Raton: Florida, pp. 1-403, **2001**.
6. SALT D.E., KATO N., KRAMER U., SMITH R.D., RASKIN I. The role of root exudates in nickel hyperaccumulation and tolerance in accumulator and nonaccumulator species of *Thlaspi*. In: *Phytoremediation of contaminated soil and water*, pp. 189-200 (TERRY N., BANUELOS G.): CRC Press LLC: London, **2000**.
7. CHEN C., HUANG D., LIU J. Functions and toxicity of nickel in plants: recent advances and future prospects. *Clean* **37**, 304, **2009**.
8. FOLGAR S., TORRES E., PÉREZ-RAMA M., CID A., HERRERO C., ABALDE J. *Dunaliella salina* as a marine microalga highly tolerant to but a poor remover of cadmium. *J. Hazard. Mater.* **165**, 486, **2009**.
9. PAN J., PLANT J., VOULVOULIS N., OATES C., IHLENFELD C. Cadmium levels in Europe: implications for human health. *Environ. Geochem. Health* **32**, 1, **2010**.
10. KOOPMANS G.F., RÖMKENS P.F.A.M., FOKKEMA M.J., SONG J., LUO Y.M., JAPENGA J., ZHAO F.J. Feasibility of phytoextraction to remediate cadmium and zinc contaminated soils. *Environ. Pollut.* **156**, 905, **2008**.
11. DEGRYSE F., BROOS K., SMOLDERS E., MERCKY R. Soil solution concentration of Cd and Zn can be predicted with a CaCl₂ soil extract. *Eur. J. Soil Sci.* **54**, 149, **2003**.
12. DAVIES B.E. Lead. In: *Heavy Metals in Soils*, pp. 177-196 (ALLOWAY B.J.), Blackie and Son; Glasgow: UK, **1990**.
13. McLAUGHLIN M.J., PARKER D.R., CLARKE J.M. Metals and micronutrients-food safety issues. *Field Crop. Res.* **60**, 143, **1999**.
14. MARKERT B. Presence and significance of naturally occurring chemical elements of the periodic system in the plant organism and consequences for future investigations on inorganic environmental chemistry in ecosystems. *Vegetation* **103**, 1, **1992**.
15. TURPEINEN R., SALMINEN J., KAIRESALO T. Mobility and bioavailability of lead in contaminated boreal forest soil. *Environ. Sci. Technol.* **34**, 5152, **2000**.
16. GABBRIELLI R., PANDORFINI T., VERGNAMO O., PALANDRI M.R. Comparison of two serpentine species with different nickel tolerance strategies. *Plant Soil* **122**, 271, **1990**.
17. GORLACH E., GAMBUS F. A study of the effect of sorption and desorption of selected heavy metals in soils on their uptake by plants. *Zeszyty Problemowe Postepow Nauk Rolniczych* **398**, 47, **1992**.
18. MEERS E., VANDECASTEELE B., RUTTENS A., VANGRONSVELD J., TACK F.M.G. Potential of five willow species (*Salix* spp.) for phytoextraction of heavy metals. *Environ. Exp. Bot.* **60**, 57, **2007**.
19. VYSLOUŽILOVÁ M., TLUSTOŠ P., SZÁKOVÁ J., PAVLÍKOVÁ D. As, Cd, Pb and Zn uptake *Salix* spp. clones grown in soils enriched by high loads of these elements. *Plant Soil Environ.* **49**, (5), 191, **2003**.
20. SCHMIDT U. Enhancing phytoextraction: The effect of chemical soil manipulation on mobility, plant accumulation, and leaching of heavy metals. *J. Environ. Qual.* **32**, 1939, **2003**.
21. BRICKER T.J., PICHTEL J., BROWN H.J., SIMMONS M. Phytoextraction of Pb and Cd from a superfound soil: effects of amendments and croppings. *J. Environ. Sci. Health* **36**, 1597, **2001**.
22. GREGER M., LANDBERG T. Use of willow in phytoextraction. *Int. J. Phytoremed.* **1**, (2), 115, **1999**.
23. NOWACK B., KOEHLER S., SCHULIN R. Use of diffusive gradients in thin films (DGT) in undisturbed field soils. *Environ. Sci. Technol.* **38**, 1133, **2004**.
24. McBRIDE M. Toxic metals in sewage sludge-amended soils: has proportion of beneficial use discounted the risks? *Adv. Environ. Res.* **8**, 5, **2003**.
25. KACÁLKOVÁ L., TLUSTOŠ P., SZÁKOVÁ J. Phytoextraction of cadmium, copper, zinc and mercury by selected plants. *Plant Soil Environ.* **55**, (7), 295, **2009**.
26. KACÁLKOVÁ L., TLUSTOŠ P. The uptake of persistent organic pollutants by plants. *Cent. Eur. J. Biol.* **6**, (2), 223, **2011**.
27. SZÁKOVÁ J., TLUSTOŠ P., BALÍK J., PAVLÍKOVÁ D., VANĚK V. The sequential analytical procedure as a tool for evaluation of As, Cd and Zn mobility in soil. *Fres. J. Anal. Chem.* **363**, 594, **1999**.
28. QUAVAUILLER P., URE A., MUNTAU H., GRIEPINK B. Improvement of analytical measurements within the BCR-program – Single and sequential extraction procedures applied to soil and sediment analysis. *Int. J. Environ. Anal. Chem.* **51**, 129, **1993**.
29. MIHOLOVÁ D., MADER P., SZÁKOVÁ J., SLÁMOVÁ A., SVATOŠ Z. Czechoslovakian biological certified reference materials and their use in the analytical quality assurance system in a trace element laboratory. *Fresenius J. Anal. Chem.* **345**, 256, **1993**.
30. PRETUZZELLI G. Recycling wastes in agriculture: heavy metals bioavailability. *Agric. Ecosyst. Environ.* **27**, (1-4), 493, **1989**.
31. HENRY J.R. In an Overview of Phytoremediation of Lead and Mercury. NNEMS Report: Washington, D.C., pp. 3-9, **2000**.
32. MORAGHAN J.T. Accumulation of cadmium and selected elements in flax seed grown on a calcareous soil. *Plant Soil* **150**, 61, **1993**.
33. SHUMAN L.M. Fractionation method for soil microelement. *Soil Sci.* **140**, (1), 11, **1985**.
34. ANGELOVA V., IVANOVA R., DELIBALTOVA V., IVANOV K. Bio-accumulation and distribution of heavy metals in fibre crops (flax, cotton and hemp). *Industrial Crops and Products* **19**, 197, **2004**.

35. MANI D., SHARMA B., KUMAR CH., PATHAK N., BALAK S. Phytoremediation potential of *Helianthus annuus* L. in sewage-irrigated indo-gangetic alluvial soils. *Int. J. Phytoremed.* **14**, (3), 235, **2012**.
36. MOHANTY M., PATRA H.K. Phytoremediation potential of paragrass – an in situ approach for chromium contaminated soil. *Int. J. Phytoremed.* **14**, (8), 796, **2012**.
37. SAWIDIS T., BREUSTE J., MITROVIC M., PAVLOVIC P., TSIGARIDAS K. Trees as bioindicator of heavy metal pollution in three European cities. *Environ. Poll.* **159**, 3560, **2011**.
38. FARGAŠOVÁ A., BEINROHR E. Metal-metal interactions in accumulation of V^{5+} , Ni^{2+} , Mo^{6+} , Mn^{2+} and Cu^{2+} under- and above-ground parts of *Sinapis alba*. *Chemosphere* **36**, 1305, **1998**.
39. AKSOY A., DEMIREZEN D. *Fraxinus excelsior* as a Biomonitor of Heavy Metal Pollution. *Pol. J. Environ. Stud.* **15**, (1), 27, **2006**.
40. FUZHONG W., WANQUIN Y., ZHANG J., LIQIANG, Z. Cadmium accumulation and growth responses of a poplar (*Populus deltoids* x *Populus nigra*) in cadmium contaminated purple soil and alluvial soil. *J. Hazard. Mater.* **177**, 268, **2010**.
41. MARKERT B. Plant as a biomonitors: Indicators for heavy metals in the terrestrial environment. VCH Weinheim: New Your/Basel/Cambridge, pp. 395-401, **1993**.
42. KABATA-PENDIAS, A., PIOTROWSKA M. Zanieczyszczenie Gleb i Roślin Uprawnych Pierwiastkami Sładowymi, CBR-opracowanie Problemowe; Warszawa: Poland, pp. 2-28, **1984**.
43. ALLEN S.E. Analysis of Ecological Materials, 2nd ed. Blackwell Scientific Publications: Oxford, **1989**.
44. WHATMUFF M.S. Applying biosolids to acid soil in New South Wales: are guideline soil metal limits from other countries appropriate? *Aust. J. Soil Res.* **40**, 1041, **2002**.
45. SUN Y., ZHOU Q., LIU W., AN J., XU Z., WANG L. Joint effects of arsenic and cadmium on plant growth and metal bioaccumulation: a potential Cd-hyperaccumulator and As-excluder *Bidens pilosa* L. *J. Hazard. Mater.* **165**, 1023, **2009**.
46. HAN Y.L., YUAN H.Y., HUANG S.Z., GUO Z., XIA B., GU J. Cadmium tolerance and accumulation by two species of *Iris*. *Ecotoxicology* **16**, 557, **2007**.
47. SNEDECOR G.W., COCHRAN W.G. Statistical Methods. Sixth Edition; The Iowa State University Press, pp. 1-593, **1967**.
48. ORROÑO D.I., SCHINDLER V., LAVADO S.R. Heavy metal availability in *Pelargonium hortorum* rhizosphere: Interactions, uptake and plant accumulation. *J. Plant Nutr.* **35**, (9), 1374, **2012**.
49. VANDECASTEELE B., MEERS E., VERVAEKE P., DE VOS, B., QUATAERT P., TACK F.M.G. Growth and trace metal accumulation of two *Salix* clones on sediment-derived soils with increasing contamination levels. *Chemosphere* **58**, 995, **2005**.
50. SHALLARI S., SCHWARTZ C., HASKO A., MOREL J.L. Heavy metals in soils and plants of serpentine and industrial sites of Albania. *Sci. Total Environ.* **209**, 133, **1998**.
51. SHAMS K.M., TICHY G., FISCHER A., SAGER M., PEER T., BASHAR A., FILIP K. Aspects of phytoremediation for chromium contaminated sites using common plants *Urtica dioica*, *Brassica napus* and *Zea mays*. *Plant Soil* **328**, 175, **2010**.

