Palacký University Olomouc
Faculty of Science, Department of Ecology and Environmental Sciences

The Effect of Different Pollution Levels in Fluvisols on the Accumulation of Heavy Metals in Carrots (*Daucus carota L.)* and Spring Barley (*Hordeum vulgare L.)*

RNDr. Jan Kočař

Ph.D. thesis

Olomouc, December 2011

supervisor:	**prof. Dr. Ing. Bořivoj Šarapatka, CSc.** Palacký University Olomouc Faculty of Science Department of Ecology and Environmental Sciences
consultant:	**Ing. Michaela Smatanová, Ph.D.** Central Institute for Supervising and Testing in Agriculture in Brno

Abstract

This thesis deals with the effect of different pollution levels of soils with an accumulation of heavy metals by two plants. Fluvisols from the Morava river floodplain area are used. The soil samples are taken from three different localities (yearly, irregularly and rarely flooded). Homogenized sludge from a municipal wastewater treatment plant is artificially introduced into the samples. Two types of accumulative plants are used: the Rubina variety of Daucus carota and the Amulet variety of Hordeum vulgare. In the following analysis of changes in soil characteristics are observed, i.e., mainly the heavy metal concentrations of cadmium, lead, zinc, copper and nickel in soils before the experiment, after finishing each cycle and in the plants themselves. Analysis show exceedingly high levels of heavy metals in fluvisols with almost all elements, with the exception of lead. Zinc and nickel have the highest concentrations. The yield of both plants was influenced by the amount of applied sludge, with the same effect as a fertilizer. On the other hand the high sludge pH level partly limited introduction of most heavy metals into the plants. Total increases of heavy metals in the plants were monitored and evaluated by using transfer factors. They are compared at all locations including the differences between the controls and soil with the sludge, and total transfer factors focused on various elements. The research showed almost the same accumulation abilities of heavy metals except cadmium by both plants. As expected there is a higher accumulation potential in the root of D. carota and in the straw of H. vulgare.

Key words

accumulation, fluvisols, heavy metals, transfer factors, carrot, barley

Contents

1. Introduction

Heavy metal pollution of soils is increasingly becoming a global problem with the development of industry, mining activity, waste water irrigation and the application of sewage sludge, even if it is relatively localized at present. The soil-plant system is the fundamental building block of the geosphere and biosphere. Therefore, heavy metal pollution of the soil has an important influence not only on the yield and quality of crops, but also on the quality of the atmospheric and the aquatic environment, and even on the health of human beings via the food chains (Lasheen 2000, Yamagata 1970).

An Important source of heavy metal soil contamination can become industrial compost, which is primarily produced from sewage sludge. At present we do not have enough knowledge of the negative effects of repeated compost application (with different levels of heavy metals) on soil and plants (Vana 2003).

Traditional solutions such as disposal of contaminated soil in landfills account for a large proportion of the remediation operations at the present. However, some of the remediation techniques currently in use will probably lose economic favour and public acceptance in the near future (Lombi 2000).

Heavy metals can be translocated to plant parts aboveground. The metal-rich plant material may be safely harvested and removed from the site without extensive excavation, disposal costs, and loss of topsoil associated with traditional remediation practices (Blaylock 1997).

Contaminated sites often support characteristic plant species, some of which are able to accumulate high concentrations of heavy metals in their tissue (Baker 1989).

Most plants that survive in polluted soils do so by either, avoiding heavy metals, or, hyperaccumulating them in their tissues. Such plants are uncommon (Cunningham 1996) and, to date, approximately 400 hyperaccumulator species have been identified, according to an analysis of field-collected speciments (Kramer 1997).

Besides the limited distribution of hyperaccumulators in the wild, such plants also tend to be contaminant specific. No plant species has yet been found that will demonstrate a wide spectrum of hyperaccumulation (Watanabe 1997).

The success of the phytoremediation process is dependent on adequate plant yield and high metal concentrations in plant shoots. Plants must produce sufficient biomass while accumulating high concentrations of heavy metals. Hyperaccumulator plants possess an ability to take up abnormally high amounts of heavy metals in their shoots (Chaney 1997; Shen 1997). However, most hyperaccumulators species are not suitable for phytoremediation application in the field due to their small biomass and slow growth (Huang 1996; Banuelos 1997).

1.1 Aims

The aims of the thesis are:

- The analyzation and comparison of the soil samples taken from six locations (flooded annually, irregularly and rarely) located on the north and south of the city Olomouc.

- The evaluation of the content of potentially hazardous elements – heavy metals Cd, Cu, Ni, Pb and Zn – in samples of fluvisols from Morava river.

- The selection and description of physical and chemical properties of fluvisols and in the plants before the experiment and after finishing of each cycle.

- The research of the effect of different pollution levels of soils with an accumulation of heavy metals by Carrots (Daucus carota L.) and Spring Barley (Hordeum vulgare L.).

- Using of homogenized sludge from a municipal wastewater treatment plant artificially introduced into the samples as a fertilizer.

- The comparison of the yield of both plants and possibilities of influencing by the amount of applied sludge.

- The monitoring of total increases of heavy metals in the plants evaluated by using transfer factors.

- The characterisation of the risk of contamination of plant production by hazardous substances.

- The evaluation that will be carried out according to legislative regulations and using the results of the research, regarding the load of hazardous substances on soil.

1.2 Hypotheses

The hypotheses can be following:

- The soil samples taken from surroundings of Olomouc city will show higher values of heavy metals caused by the floods from 1997.

- There is some phytoextraction potential of experimental plants grown on fluvisols and their values of transfer factors will be higher than some selected hyperaccumulator plants.

- The heavy metals from floods and sewage sludge can accumulate in soil and in plants and can produce harmful effects in animals and humans through food chain.

- Higher pH of sewage sludge from wastewater treatment plant can influence (limit) the transfer of heavy metals into the plants and on the other side can possitively influence the plant´s yield.

2. Heavy metal pollution

Soil and its uses have been neglected or have been taken for granted for centuries. The human race has been disposing of waste via the soil for as long as we have been on the earth.

Agriculturally based civilisations have used human and animal waste to fertilise the soil and when populations were small this was no problem in fact it was the natural scheme of things but as the human race increased in number the pressure on the soil to produce ever more food be it arable or dairy has put strains on the soil ecosystem. In the 20th century the additions of fertiliser, pesticides and herbicides have stored up problems for the human race and all species.

When the industrialisation began people moved from working the soil to working in factories but unfortunately still applied the same processes to waste disposal thinking that when waste was deposited onto or into the soil then the problem had gone away. Over the last half century or so we have discovered that our forbears were wrong.

Pollution that is added to the soil may adhere to the soil matrix and become immobile but may still be available to plant roots and be taken up into the plant structure from where it can enter the food chain. Other pollutants can slowly permeate through the top soil structure to the sub strata and eventually into the aquifer below. An example of this is our problems with nitrate in drinking water which arise from the intensive use of nitrate fertilisers which have now been washed through the soil and found their way through to the aquifer. This may or may not be the case for heavy metals in soils as their mobility within the soil matrix is dependent upon several factors including the organic content of the soil, the soil type and soil pH.

In report number CSI 015 entitled Progress in Management of Contaminated Sites dated August 2007 the European Environment Agency (EEA) states that member countries had at the time identified 250,000 sites where soil contamination requires clean up. The EEA estimates that there are around 3,000,000 sites across Europe where potentially polluting activities have occurred and where investigation is needed to establish if soil remediation is required. The EEA also predicted that the number of sites requiring remediation is likely to increase by 50% by 2025. This compares to the number of sites where the remediation process has been completed which, over the last 30 years, is estimated to be in the order of 80,000 sites.

The EEA report also mentions briefly that the soil is not being considered as a valuable resource but as a disposable commodity. Where pollution of soil has occurred by whatever process within Europe the two main treatment processes are either excavation of the soil and removal to a waste disposal site or by containment of the area neither of which actually deals with the problem in the long term. There exist processes for the onsite remediation of soils by washing, bio remediation, direct chemical and most recently electro-chemical techniques by either in-situ or ex-situ (off site) systems and these are being taken up more often due in part to the increasingly high costs of landfill. In the UK the Environment Agency applies a fit for purpose approach to soil pollution applying the most stringent standard to agricultural use, lower for residential use and the lowest standard for industrial uses of the land involved (Gjoka 2011).

3. A hidden source of pollution

The sources of heavy metal pollution tend to be associated with heavy industry, both existing and historical. There is a legacy of pollution residing in the rivers of the developed world.

In a recent paper published by the UK Environment Agency Entitled ‘The Assessment of Metal Mining Contaminated River Sediments in England and Wales’ dated November 2008 states:

“Although metal discharges were greater during the peak period of active mining in the nineteenth century, significant inputs of dissolved and particulate metals still occur. Past discharges have left a substantial reservoir of highly contaminated sediments in lowland rivers many kilometres downstream of the mines, and these sediments are likely to be causing ecological damage. Re-suspension of these sediments during floods has the potential to cause additional harm to aquatic life, and to contaminate floodplain soils used for agriculture.”

With the predicted increase in the frequency of major flood events due to global warming, the likelihood of the mobilising of heavy metal containing sediments and their deposition onto the fertile land on the margins of rivers is greatly increased. So in the future there is a real risk of some areas of low lying land downstream of mining and industrial areas becoming so contaminated as to be of no use for agricultural purposes where originally these areas were considered the most fertile due to the same flooding events depositing nutrients (Gjoka 2011).

Heavy metals occur naturally in soils in trace amounts. However, some soils contain high levels of these elements derived from weathering of minerals or from human activity (Fergusson 1990). Accumulation of heavy metals in soils results in significant risks for living organisms and human health, and their negative effects depend on the metal concentrations and on the soilspecific properties (Lacatusu 1995). Therefore, investigation of soil pollution with these elements is becoming increasingly important.

Amounts of heavy metals that enter the soil as a result of human activity or soil metal pollution can be evaluated on the basis of background values of these elements in soil (LABO 1995). These values represent the contents found in nature reserves plus atmospheric depositions (Alloway 1995), which are measured in soil by chemical analysis and given as the 90th percentile. Moreover, to interpret the level of soil heavy metals, the reference values can be used (EWERS 1991, quoted by Lacatusu 1995). They are obtained from the data of soil attributes determined in the soil samples.

Urban soils are often enriched in metals (reviewed in Charlesworth et al. 2010) and are a major site for human exposure to metals (De Miguel et al. 2007 and references therein). Soil metal contamination has the potential to be a serious public health issue, especially if edible plants are grown in these soils. Elevated concentrations of Cd in soils used to grow rice (Oryza sativa L.) led to human renal tubular dysfunction among Asian families (Kasuya 2000). Pruvot et al. (2006) calculated that the consumption of homegrown vegetables and crops (lettuce, leeks, cereals) in Northern France was a major contributor to total Cd and Pb exposure.

The decline in agricultural soil productivity due to erosion, runoff, and loss of organic matter (OM) has stimulated interest in OM amendments including municipal organic waste, sewage sludge, agricultural waste, animal manure, and industrial byproducts (Stevenson 1994). As in agriculture, compost application is the most common input of OM to urban gardens. Soil OM improves soil structure through the formation of cationic bridges (Hernando et al. 1989). It also increases soil fertility and water retention, and influences chemical speciation (Soumare et al. 2003).

Risk assessment aims to characterize the potential adverse health effects of human exposures to environmental hazards (Markus and McBratney 2001). Quantitative guidelines for assessing risks associated with soil contamination are difficult to establish due to the complexity of the system. One solution is to establish maximum acceptable concentrations of metals in soils to be used for agriculture (cf., CCME 2006). Such guidelines are generally based on simplified assumptions about soil types and site conditions (Grasmuck and Scholz 2005) and thus may over- or underestimate potential risk. A second approach is to calculate a bioconcentration factor, which describes the concentration of a metal in the plant relative to the concentration in the soil (e.g., Antunes et al. 2006); the higher the bioconcentration factor, the more mobile the metal. A more direct, but labor-intensive, approach involves calculating a hazard quotient: the ratio of the amount of metal in a serving of food to the amount of metal known to be hazardous as determined through toxicity tests (Pierzynski et al. 2005).

The discharge of untreated or partly treated wastewater as well as the runoff from land surface results in contaminations of rivers and their floodplains with heavy metals and/or persistent organic pollutants (POPs; Baborowski et al. 2004; Antić et al. 2006; van der Veen et al. 2006; Nikarnorov et al. 2007; Moiseenko et al. 2008). In river water, the majority of the pollutants is associated with suspended particulate matter (Morales et al. 2007; Pepelnik et al. 2005), which can be aggregated to freshwater flocs (Droppo et al. 1997). Flocculation modifies the sorption of the pollutants and alters the transport and deposition of the suspended material including their sorbed pollutants (Droppo et al. 1997, 2001; Petticrew and McConnachie 2007). The settling rate of suspended and flocculated material depends on the flow velocity of the river and the critical shear stress of the aggregated material in the riverbed (Droppo et al. 2001; Böhme 2006; Büttner et al. 2006; Krüger et al. 2006; Schwartz 2006). During flood events, polluted river water as well as resuspended river sediments can be deposited in floodplain soils according to the ratio of flow velocity to sedimentation velocity (Büttner et al. 2006; Schwartz 2006; Baborowski et al. 2007). Therefore, the concentrations of pollutants vary horizontally and vertically, resulting in pronounced spatial distribution patterns in the floodplains (Witter et al. 2003; Gröngröft et al. 2005; Krüger et al. 2005; Büttner et al. 2006). The above processes explain why floodplains are sinks for pollutants. However, floodplains can also be sources for groundwater and downstream contamination or for the transfer of pollutants into the food chain (Gröngröft et al. 2005; Bethge-Steffens 2008).

Potentially toxic trace elements are considered as one of the main sources of contamination, mainly because of human activities such as industry and agriculture, which mobilize and redistribute these elements, often causing adverse effects. The study of the distribution of trace elements in soils is important because they are considered as the principal sinks for these elements and ideal means for monitoring contamination (Cui et al. 2005).

Concentration of heavy metals in sediments deposited during floods on floodplains is related to their content in transported and accumulated sediments in a river channel in the period, which immediately precedes a flood. As a result, changes of heavy metal concentrations in the vertical profiles of overbank sediments reflect changes of river pollution in the period of sediments deposition (Macklin and Klimek, 1992).

Heavy metals discharged from industries return to nature mainly through pathways of solid wastes, wastewater and waste gas. The entry of heavy metals into air, water and soil, and subsequent transfer from soil to plant can ultimately have significant adverse biological and ecological effects. Soil properties (pH, OM, E*h*, CEC, etc.), plant species at different growth stages, and pollution inputs all influence the mobility of heavy metals. Surveys of heavy metal levels in air-water-soil-plant systems have been made in many countries. Especially soil-plant systems have been surveyed by taking field samples of soils and plants as well as samples of selected vegetables and foodstuffs from main wholesale markets, for example, France (Sterckeman *et al*., 2000), Germany (Muller *et al*., 1996), Latvia (Klavins *et al*., 2000), Norway (Steinnes *et al*., 1997) and Poland (Chlopecka *et al*., 1996).

Excess heavy metal accumulation in soils is toxic to humans and other animals. Exposure to heavy metals is normally chronic (exposure over a longer period of time), due to food chain transfer. Acute (immediate) poisoning from heavy metals is rare through ingestion or dermal contact, but is possible. Chronic problems associated with long-term heavy metal exposures are:

- Lead – mental lapse.
- Cadmium – affects kidney, liver, and GI tract.
- Arsenic – skin poisoning, affects kidneys and central nervous system.

The most common problem causing *cationic* metals (metallic elements whose forms in soil are positively charged cations e.g., Pb2+) are mercury, cadmium, lead, nickel, copper, zinc, chromium, and manganese. The most common anionic compounds (elements whose forms in soil are combined with oxygen and are negatively charged e.g., $MoO4^{2-}$) are arsenic, molybdenum, selenium, and boron.

4. Floodplains

The history of local and regional contamination of European environments by Pb has been a subject of numerous studies (Farmer et al. 1997; Kober et al. 1999; Weiss et al. 1999; Middelkoop 2000; Novák et al. 2003). Much less attention has been paid to the history of regional pollution by Zn (Martínez-Cortizas et al. 1997; Middelkoop 2000), perhaps because of its lower toxicity. In the fluvial domain, most previous studies on historical contamination were focused on sites or regions strongly impacted by mining and metallurgy (Borůvka et al. 1996; Hudson-Edwards et al. 1998; Middelkoop 2000), but enhanced input of heavy metals can be identified also in topsoils of mainly agricultural regions (Martin 2000; Desenfant et al. 2004; Kadlec et al. 2009) and even at sites very remote from local sources (Renberg et al. 2001; Novák et al. 2003). Actually, in these “cleaner” regions, the major input of heavy metals, in particular Pb, has been a result of long atmospheric transport and hence it reflects larger-scale sources (Renberg et al. 2001), more geographically smoothed and attributed to broader societal processes, such as massive introduction of coal burning in Central and West Europe (Weiss et al. 1999; Renberg et al. 2001; Novák et al. 2008) as the main energy source in the nineteenth and early twentieth centuries.

Floodplains are stratigraphically less straightforward sedimentary archives of chemical contamination Two main reasons are a complex stratigraphy of the floodplain and mobility of heavy metals due to water level fluctuations. It has probably hindered the use of fluvial sediments to reconstruct the changes of regional contamination in the twentieth century. The spatial complexity of heavy metal distribution in floodplains has been observed since the beginning of their systematic study. To understand how the contaminant content depends on the distance from the river channel (Borůvka et al. 1996), relative position of dikes (Middelkoop 2000) and depth in the floodplain soil profile (Borůvka et al. 1996; Martin 2000), the depth profiles of the contaminants must be interpreted in accord with complex depositional environments of floodplains (Lewin and Macklin 2003). The effort of understanding these patterns is justified by the fact that floodplains of lowland rivers are the most widespread sedimentary environment in many countries including central European ones. The complexity of floodplains as a sedimentary archive can be dealt with by describing them to the more remote past than only to the target period of the environmental geochemical studies to get reliable lithogenic (pre-industrial) heavy metal contents. It is essential to separate the influence of the variable sorting of the suspended and bed-load sediment before its deposition in floodplain, which is facilitated by normalisation of the heavy metals to selected matrix elements (Nováková 2009; Grygar et al. 2010).

5. Relation plants and soil

One of the most important aspects of managing successfully biomass crop systems amended with composted biodegradable wastes on contaminated land is their impact on contaminant fate and transport including their potential for remediation and stabilisation. Partial in situ remediation of organic contaminants is anticipated while stabilisation and phytoremediation may have a long-term impact on available metals due to amendment, plant cover and the resulting modification of the biochemical and physical properties of the soil.

Plants that grow on sites with high metal concentrations use one of two known mechanisms to tolerate the toxic effects. One mechanism, known as hyper-accumulation, involves the active uptake of metals and subsequent detoxification in aboveground tissue. The other, known as exclusion, is whereby the plant maintains a certain concentration in the shoot regardless of soil loadings with detoxification taking place in the roots. It is also thought that the accumulation mechanism is one that has evolved in plants that are entirely confined to metalliferous soils while exclusion is a strategy expressed by plants that have evolved some species tolerant and some intolerant to elevated soil metal concentrations.

Plants have been shown to form intimate symbiotic relationships with soil microorganisms that result in their tolerance abilities. The release of root exudates, chemicals and elaborated metabolites, which supply a food source for this microbially-enriched zone, creates a zone of influence described as the rhizosphere. The rhizosphere processes occurring as a result of plant or soil microorganism metabolisms alter the soil biochemistry and have an influence on the availability of metals by either increasing the metal mobility through proton production and acidification of the soil or reducing mobility by catalysing the precipitation of metals as sulphides. This acidification has the consequence that plants which have adopted the accumulation mechanism can thus increase the amount of available metals in their rhizosphere and their phytoremediation potential becoming hyperaccumulators. On the other hand, precipitation results in a reduction in metals bioavailability and in a reduction in the risk they pose (stabilisation). Likewise, several studies have shown that the microbial degradation of organic contaminants in soils can be enhanced in the rhizosphere of plants through the increased numbers and metabolic activity of degrading microorganisms which are fuelled by the input of organic carbon from root exudates and from the sloughing of root cells. But some contaminants such as Polycyclic Aromatic Hydrocarbons (PAHs) may also interact with plants by accumulation in plant tissues or adsorption on root surfaces.

The stabilisation of the contaminated sites due to plants is likely to play a larger role in the mitigation of the risk posed by contaminants. Plants can assist in controlling and treating secondary non-point source pollution in air and water as well as in containing down gradient movement of contaminated ground water. Plants stabilise the soil, thus preventing erosion. Plants help improve soil quality and soil structure. The resulting increase in water infiltration and the reduction in runoff can help mitigate contaminant migration.

Some laboratory studies on the bioremediation of organic contaminants have shown that the addition of compost to contaminated soil can have a positive effect on soil microbial properties and increase the rate of degradation of some organic contaminants. However, hydrophobic organic contaminants such as PAHs are known to become sequestrated by partitioning into organic matte or diffusing into nano- and micropores and become less bioavailable with time which is believed to be responsible for a decrease in biodegradation rates in aged soils. Similarly heavy metals can sorb strongly onto organic matter. This reduction in contaminant bioavailability through interaction with organic matter also results in a reduction in contaminant mobility.

In poor soils one of the major limiting factors in the degradation process of organic contaminants is the supply of nutrients such as nitrogen and phosphorus to the soil microbial population and the plants. The imbalance in the ratio of carbon to nitrogen is compounded by the immobilisation of the available soil nitrogen by the growing microbial population and can lead to insufficient supplies to meet the needs of plants. Also, an important factor in improving phytoremediation is to maximise the root surface in order to optimise exchanges between soil and plant. Another important factor in the uptake rate is the evapotranspiration rate which should be optimised. Thus phytoremediation efficiency should be improved through healthy plant and root system growth. Hence it would appear that the necessity of improving the agronomic performance Contaminated Soils of plants for acceptable yields is also a condition for improving contaminant remediation.

Several studies have shown that, in an agricultural context, organic amendments improve soil physical and hydraulic properties. Increasing soil organic matter content improves soil structure leading to more stable aggregates, lower bulk density, better infiltration rates and higher water retention capacity.

It has been recognized for more than seventeen years that plant uptake could be exploited as a biological clean-up technique for various polluted rooting media including soils, composted materials, effluents and drainage waters. Before phytoextraction of soils is possible on a large scale, a number of important issues must be addressed. Firstly, metal hyperaccumulator plants are relatively rare, often occurring in remote areas geographically and being of very restricted distribution in areas often threatened by devastation from mining activities. Population sizes can be extremely small. There is thus an urgent need to collect these materials, bring them into cultivation and establish a germplasm facility for large-scale production for future research and development and trials work. Secondly, the potential exploitation of metal uptake into plant biomass as a means of soil decontamination is clearly limited by plant productivity. Many of the temperate hyperaccumulator plants are of small biomass, although considerable natural variation exists within populations. Selection trials are needed to identify the fastest growing (largest potential biomass and greatest nutrient responses) and most strongly metal-accumulating genotypes. However, such a combination may not be possible and a trade-off between extreme hyperaccumulation and lower biomass (or vice versa) may be acceptable. Selection could also identify the individuals with the deepest and most extensive and efficient root systems, and those of greatest resistance to disease. Breeding experiments are required to incorporate all these desirable properties into one plant.

Future work will involve genetic engineering to further improve metal-uptake characteristics, if the genes for metal accumulation can be identified and manipulated. The possibility then exists to transfer genes for metal hyperaccumulation into a very productive (but inedible), sterile host plant. Excellent opportunities also exist through protoplast fusion techniques. There are very few hyperaccumulator plants discovered to date that have a capacity for multiple metal accumulation. Some, whilst primarily accumulating a single metal, do also show enhanced uptake of others. However, there is some experimental evidence to indicate metal antagonisms may limit uptake from multiply metal-contaminated soils. Increasing systematic effort in screening plant materials for these characteristics will most certainly reveal new hyperaccumulator plants - and new potentials for phytoextraction, phytomining and biorecovery.

Many plant phyisological properties are of importance for phytoremediation such as low or high metal accumulation, low or high root-to-shoot translocation of metal, high biomass production and, often, high tolerance to the specific metal.

Hyperaccumulator plants accumulate inordinate amounts of one or more Trace Elements (TE) in their above ground biomass. Hyperaccmulators can have TE concentrations in their dry biomass that are 100 times higher than non-hyperaccumulators growing in the same soil. For most TEs a common threshold concentration for a plant to be considered a TE hyperaccumulator is 0.1%. For zinc and manganese, the threshold concentration is 1% and for cadmium, the threshold concentration is 0.01%. At present, there are over 400 species of known hyperaccumulators. A continual stream of new discoveries adds to this list. Hyperaccumulators species may accumulate one or more of a range of TEs that currently includes nickel, manganese, zinc, cadmium, thallium, copper, cobalt and arsenic.

The hyperaccumulation trait has evolved (or was, er, created) several times, as it is occurs in several families in the plant kingdom. Many hyperaccumulators belong in the Brassicaceae. One current mystery is what, if any, advantage does TE hyperaccumulation confer on the plant. Five theories are:

1 - tolerance to, or disposal of, the TE from the plant,
2 - a drought-resistance strategy,
3 - a means of avoiding competition from less TE-tolerant plants,
4 - inadvertent uptake of TEs,
5 - defence against herbivores or pathogens.

Despite the obvious appeal of the herbivore defence theory, studies have shown that, in many cases, TE accumulation does not protect the plant from herbivore attack.

6. How do plants take up and transport metal?

The process of metal accumulation involves several steps, outlined in Fig. 1, one or more of which are enhanced in hyperaccumulators.

6.1 Solubilization of the metal from the soil matrix

Many metals are found in soil-insoluble forms. Plants use two methods to desorb metals from the soil matrix: acidification of the rhizosphere through the action of plasma membrane proton pumps and secretion of ligands capable of chelating the metal. Plants have evolved these processes to liberate essential metals from the soil, but soils with high concentrations of toxic metals will release both essential and toxic metals to solution. To our knowledge, there are no reports of plants with the ability to solublize Pb from the soil matrix, where most of soil Pb exists in an insoluble form (Blaylock and Huang 2000). Experiments demonstrating Pb hyperaccumulation have used Pb(NO3)2, a soluble form of Pb, though it must be questioned whether this is the most appropriate form of Pb for analysis. Aside from Pb, the solubilization mechanisms for hyperaccumulators are similar for metals discussed, and therefore will not be addressed independently for each metal. While no hyperaccumulators have evolved to handle high concentrations of toxic metals if they are present in solution, phytoremediator plants could be modified to solubilize contaminants that are bound to the soil.

6.2 Uptake into roots

Soluble metals can enter into the root symplast by crossing the plasma membrane of the root endodermal cells or they can enter the root apoplast through the space between cells (Fig. 1). While it is possible for solutes to travel up through the plant by apoplastic flow, the more efficient method of moving up the plant is through the vasculature of the plant, called the xylem. To enter the xylem, solutes must cross the Casparian strip, a waxy coating, which is impermeable to solutes, unless they pass through the cells of the endodermis (Fig. 1). Therefore, to enter the xylem, metals must cross a membrane, probably through the action of a membrane pump or channel. Most toxic metals are thought to cross these membranes through pumps and channels intended to transport essential elements. Excluder plants survive by enhancing specificity for the essential element or pumping the toxic metal back out of the plant (Hall 2002; Meharg and Macnair 1992a, 1992b).

6.2 Transport to the leaves

Once loaded into the xylem, the flow of the xylem sap will transport the metal to the leaves, where it must be loaded into the cells of the leaf, again crossing a membrane (Fig. 1). The cell types where the metals are deposited vary between hyperaccumulator species. For example, T. caerulescens was found to have more Zn in its epidermis than in its mesophyll (Kupper et al. 1999), while A. halleri preferentially accumulates its Zn in its mesophyll cells instead of its epidermal cells (Kupper et al. 2000).

7. Common elemental contaminants

Elements naturally occur in the earth’s crust in a range of background levels that are generally below the critical load, i.e., the amount of the element above which there is a negative effect on biodiversity and ecosystem function. However, the concentrations of elements in localized, naturally occurring metalliferous soils or in depositions from anthropogenic activity (e.g. mining, waste disposal, etc.) are considerably higher. In the following section, plant mechanisms of tolerance and/or hyperaccumulation of common elemental contaminants are discussed.

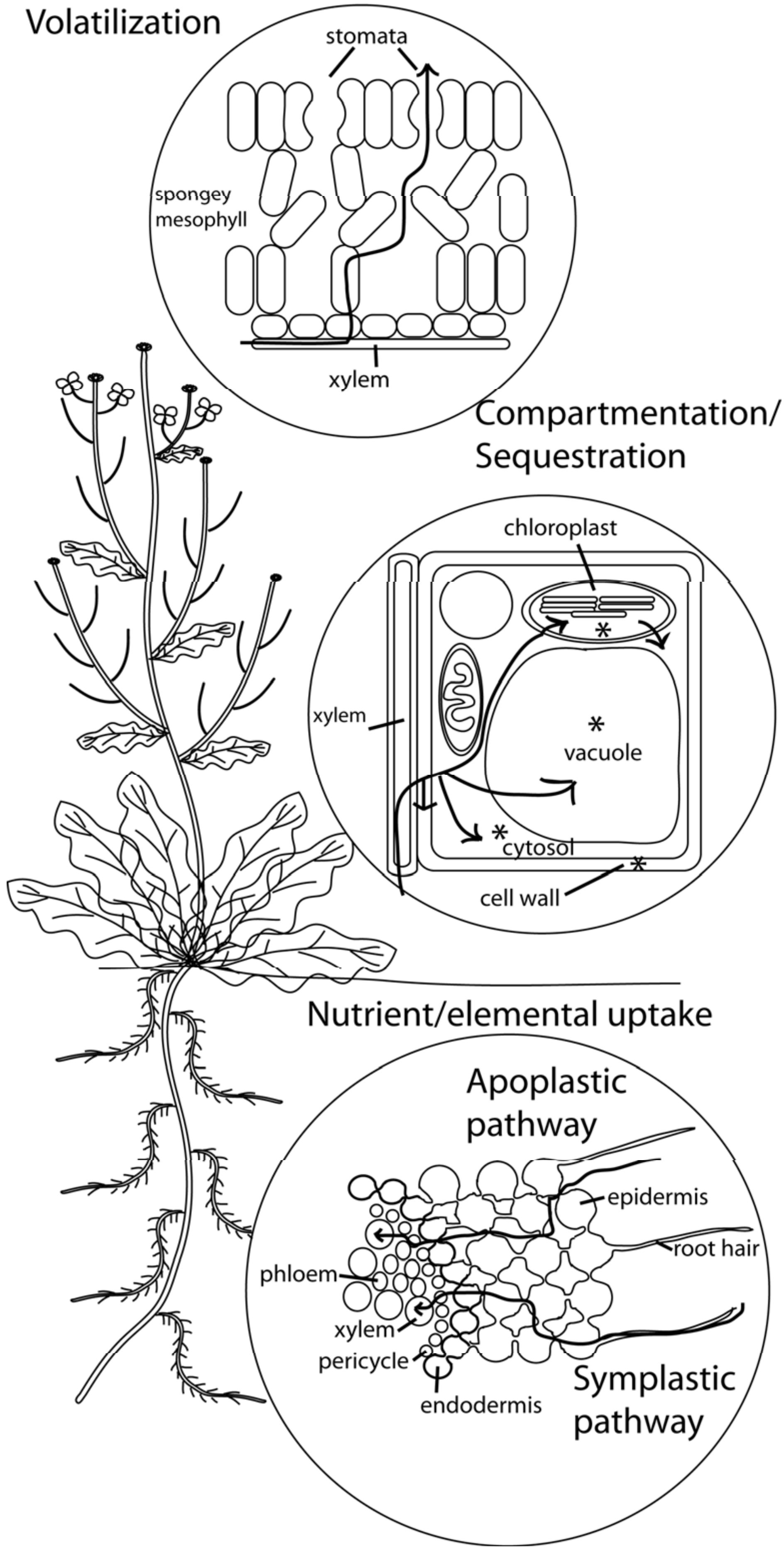

Volatilization
stomata
spongey mesophyll
xylem
Compartmentation/ Sequestration
chloroplast
*
xylem
*
vacuole
*
cytosol
cell wall
*
Nutrient/elemental uptake
Apoplastic pathway
epidermis
root hair
phloem
xylem
pericycle
endodermis
Symplastic pathway

Fig. 1. Pathway of metal/nutrient uptake in plants. Soluble metals can enter into the root symplast by crossing the plasma membrane of the root endodermal cells or they can enter the root apoplast through the space between cells. If the metal is translocated to aerial tissues, then it must enter the xylem. To enter the xylem, solutes must cross the Casparian strip, a waxy coating which is impermeable to solutes, unless they pass through the cells of the endodermis probably through the action of a membrane pump or channel. Once loaded into the xylem, the flow of the xylem sap will transport the metal to the leaves, where it must be loaded into the cells of the leaf, again crossing a membrane. Once in the shoot or leaf tissues, metals can be stored in various cell types, depending on the species and the form of the metal, since it can be converted into less toxic forms (to the plant) through chemical conversion or complexation. The metal can be sequestered in several subcellular compartments (cell wall, cytosol, vacuole) or volatilized through the stomata.

7.1 Cadmium (Cd)

Cd is a toxic metal and probable carcinogen associated with Zn mining and industrial operations where Cd has been used to prevent corrosion of machinery. Resulting air-borne Cd dust presents a significant health hazard. Ecotypes of T. caerulescens accumulate a wide range of Cd levels. The Ganges and Vivez ecotypes can accumulate up to 10,000 mg kg-1 Cd dry weight and 12, 500 mg kg-1 Cd dry weight, respectively, without showing signs of toxicity; however, the Puy de Wolf and Prayon ecotypes only accumulate 2,300 mg kg-1 Cd dry weight and 4,800 mg kg-1 Cd dry weight, respectively (Lombi et al. 2000; Peer et al. 2003). Hyperaccumulation of Cd in Arabidopsis hallerii has also been reported (Cosio et al. 2004; Kupper et al. 2000). However, reports of hyperaccumulation of Cd in B. juncea are questionable, although some Cd accumulation in this species is evident (Salt et al. 1997).

7.1.1 Uptake into the root

Cd uptake is likely mediated through transporters or channels for other divalent ions (Cosio et al. 2004). Several of the Zn and Fe transporting ZIP genes in plants have been shown to transport Cd, although with a wide range of affinities (Grotz et al. 1998; Pence et al. 2000; Ramesh et al. 2003). Excess divalent cations in the media, such as Zn, can reduce Cd uptake in many plant species, including T. caerulescens Prayon (Lombi et al. 2002a, 2001a). Significantly, divalent cations and Ca channel blockers had no effect on the Cd uptake of T. caerulescens Ganges, suggesting that this ecotype may have developed a novel Cd uptake system (Lombi et al. 2001a, 2002a).

7.1.2 Transport to the leaves

Piñeros and Kochian (2003) demonstrated that T. caerulescens and T. arvense mesophyl cells exhibit different plasma membrane ion transport properties, but the differences cannot be directly linked to the differences in Zn and Cd accumulation. Analysis of Cd/Zn transport capacity in leaf mesophyll protoplasts demonstrated that the constitutive transport capacity and affinity for these metals were indistinguishable in T. caerulescens Ganges, A. halleri, and T. caerulescens Prayon; however, Cd accumulation increased in Ganges protoplasts but decreased in A. halleri protoplasts in conjunction with Cd pre-exposure (Cosio et al. 2004). Therefore, there may be multiple Cd transport systems in leaves. This suggests that in ddition to its novel Cd root uptake pathway, Ganges has developed mechanisms in leaves to facilitate hyperaccumulation.

7.2 Copper (Cu)

Cu is an essential element and enzyme co-factor for oxidases (cytochrome c oxidase, superoxide dismutase) and tyrosinases; however, animals and plants can accumulate toxic levels of Cu. Cu contamination for soil and groundwater usually results from mine sites like Blackbird Creek, ID (Mebane 1997) or munitions research or disposal sites like the Picatinny Arsenal, in northern New Jersey (EPA 1989). The Deer Lodge Valley and Anaconda areas of the Beaverell pedon in Montana, a region affected by long-term Cu smelting, has soil polluted with high levels of Cu and lower concentrations of As, Zn, and Pb (Burt et al. 2000, 2003). Cu contamination is also problematic in European soils exposed to historical mining and smelting activity. During the 19th century, the Devon Great Consols Mine in the Tamar Valley of Devon, UK, was the world's largest producer of Cu. As a result, Cu in the form of chalcopyrite has permeated the soil in this region (Lombi et al. 2004). In France, due to the continuous treatment of vine downy mildew with Bordeaux mix since the end of the 19th century, extensive deposits of Cu have accumulated in many vineyard soils (Brun et al. 2001). In Kayseri, Anatolia, high Cu concentrations were found in Zn-producing industrial sites (Aksoy et al. 2000), while a study in Denizli found that high levels of Cu characterize urban roadsides associated with road traffic (Celik et al. 2005). Cyprus (Kupros) has an extensive history of Cu mining extending back over 2,000 years. Cu mining wastes from this prolonged activity have had a significant impact on the environment and biota of this island (Pyatt 2001). Greece is also afflicted by Cu contamination: lake ecosystems, such as Lake Pamvotis in north-western Greece, exhibithigh concentrations of this metal (Papagiannis et al. 2004), and Cu poisoning from high Cu content in local food has occurred in areas such as Veria county (Zantopoulos et al. 1999).

7.3 Nickel (Ni)

Ni is an essential element that can be toxic and possibly carcinogenic in high concentrations. Ni toxicity in humans usually results from repeated occupational exposure resulting in dermatitis, asthma or headaches (Davies 1986), but Ni contamination of soils is primarily restricted to regions surrounding Ni smelting operations such as Sudbury, Ontario, and Harare, Zimbabwe (Johnson and Hale 2004). As, Cd, and Pb are often present in Ni mining and smelting wastes, and soil and water heavy metal concentrations from Ni mining operations can exceed governmental safety limits (Lupankwa et al. 2004). Serpentine and ultramafic soils are naturally occurring regions of high Ni concentrations characterized by unique Ni-tolerant flora. The majority of Ni hyperaccumulators have been collected from these soils. Alyssum lesbiacum and Thlaspi goesingense are both Ni hyperaccumulating plants in the Brassicaceae family. In the genus Alyssum alone, 48 different speices have been discovered containing between 1,000 µg g-1 and 30,000 µg g-1 Ni in leaf dry biomass (Kerkeb and Kramer 2003). Thlaspi goesingense has been reported to accumulate 9,490 mg Ni g-1 dry weight. (Freeman et al. 2004). Ni phytoextraction using hyperaccumulators has been patented (Chaney et al. 1999).

7.3.1 Uptake into the root

Little is known about Ni uptake into roots. Evidence that histidine (His) chelates Ni suggests that His might assist root uptake of Ni. Alyssum lesbiacum has constitutively high free His levels, and when Salmonella typhimurium ATP phosphoribosyl transferase enzyme (StHisG) was expressed in A. thaliana, the His increased twofold and biomass increased 14-

40-fold when grown on Ni (Wycisk et al. 2004). But, a comparison of the uptake mechanisms of A. lesbiacum and B. juncea, a non-accumulator, indicated that Ni and His are taken up independently, as His uptake inhibitors had no effect on the Ni uptake, and Ni was taken up as a free cation (Kerkeb and Kramer 2003). Furthermore, Salt et al. (1999) found that, while root exudation of histidine and citrate may help reduce Ni uptake for the nonaccumulator T. arvense, these exudates did not appear to be involved in the hyperaccumulation of Ni by T. goesingense.

7.3.2 Transport to the leaves

Ni and his loading into the xylem appear to be correlated (Kerkeb and Kramer 2003), and nicotianamine-Ni complexes have been shown to be transported from the roots to the shoots and across plant membranes in a manner similar to nicotianamine-Fe complexes (Becher et al. 2004).

7.4 Lead (Pb)

Pb is an extremely toxic heavy metal which is a serious threat to the health of children and wildlife. The main sources of Pb poisoning include lead paint and old gasoline spills (PbBrCl, 2PbBrCl·NH4Cl) resulting in dust and soil contamination of food and water (Xintaras 1992). Other areas with high Pb concentrations include Pb mines and smelters (PbSO4, PbO·PbSO4, and PbS), such as the Leadington mine in Leadington, MO (Tom and Miles 1935), shooting ranges, and disposal sites for old batteries. A shooting range in Cortland, NY was estimated to have accumulated 500 tons of Pb after 30 years of use. The New York Department of Environmental Conservation found that the Pb concentrations at this site posed a health threat to people and wildlife, and the site received clean-up order from the EPA. College Grove, TN, has been identified as an area of concern due to Pb contamination from old battery cases on railroad property, with Pb levels ranging from 2,700 to 5,500 ppm (Chavez 1999). Elemental Pb is insoluble and the most water soluble forms of Pb compounds are lead acetate (2 mg ml-1), lead chloride (0.009 mg ml-1), and lead nitrate (5 mg ml-1) (Xintaras 1992).

Atmospheric Pb mostly exists as PbSO4 and PbCO3. Although many plants may have a strategy of Pb exclusion as Thlaspi praecox, which hyperaccumulates Cd and Zn but excludes Pb, several plant species can hyperaccumulate soluble Pb in the soil. It has been reported that Sesbania drummondii, a leguminous shrub, and several Brassica species can accumulate significant amounts of Pb in their roots (Blaylock et al. 1997) and Piptathertan miliacetall, a grass, accumulates Pb directly correlating to soil concentrations without symptoms of toxicity for 3 weeks (Garcia et al. 2004). Sahi et al. (2002) have noted that S. drummondii can tolerate Pb levels up to 1500mg L-1 and accumulate ~40g kg-1 shoot dry weight. Brassica juncea shows reduced growth at 645 ug g-1 Pb in the soil substrate, but can accumulate 34.5 g kg-1 shoot dry weight, although significant shoot accumulation is not observed until Pb reaches saturation levels in the roots. Most of the shoot accumulation was found in stems and not leaves suggesting that Pb is relatively insoluble (Kumar et al. 1995). Microanalysis spectra data through S. drummondii root sections show a decreasing gradient of Pb contents from the epidermis to the root central axis, and electron microscopy of S. drummondii roots revealed Pb deposition in the cell membrane and cell wall (Sahi et al. 2002).

7.5 Zinc (Zn)

Zn is an essential microelement, but is toxic to animals and plants at high concentrations
(Gupta and Gupta 1998). The first Zn hyperaccumualtor identified was T. caerulescens. This plant was reported to accumulate between 25,000 and 30,000 µg g-1 total Zn before exhibiting symptoms of toxicity, although T. caerulescens can accumulate a maximum dry weight of 40,000 ug g-1 Zn in its shoots (Pence et al. 2000). Arabidopsis halleri has also been found to increase in its shoot Zn concentration from 300 ug g-1 dry wt at 1 µM Zn to 32 000 ug g-1 at 1000 µM Zn without phytototoxicity (Zhao et al. 2000). Arabidopsis lyrata ssp. Friedensville accumulates high leaf concentrations of Zn in the field (Cannon 1960), but exhibits variable accumulation in the axenic culture.

7.5.1 Uptake into the root

The ZIP family of proteins (ZRT/IRT-like proteins) transport Zn into the plants (Grotz et al. 1998; Ramesh et al. 2003). ZNT1 from T. caerulescens mediates low affinity Zn uptake as expected for a plant that grows on high concentrations of Zn (Pence et al. 2000). ZNT1 expression is higher in the hyperaccumulator T. caerulescens than in the non-accumulator T. arvense, possibly leading to a higher density of Zn transporters in the root-cell plasma membrane (Pence et al. 2000). This difference in transporter concentration could account for the observation that the hyperaccumulator and the nonaccumulator have the same affinity for Zn, but the hyperaccumulator has a higher rate of uptake (Lasat et al. 1996).

7.5.2 Transport to the leaves

Despite lower rates of uptake, the roots of T. arvense were found to accumulate substantially more Zn than in T. caerulescens (Lasat et al. 1996). This difference is likely due to better transport to the leaves in the hyperaccumulator. T. caerulescens had five times more xylem sap Zn (Lasat et al. 1998) and ten times more Zn was translocated to the shoots in T. caerulescens than in T. arvense (Lasat et al. 1996). The leaf cells of the hyperaccumulator are able to accumulate more Zn when leaf sections are subjected to high Zn (1mM) conditions (Lasat et al. 1998). The molecular mechanisms of this increased uptake are unknown.

8. Articles

This thesis is based on three major articles. All study the potencial risk of heavy metals in fluvisols but each from a slightly different perspective.

8.1 State of contamination of fluvisols of the Morava river by potentially hazardeous elements

The content of potentially hazardous elements – heavy metals – in samples of fluvisols from flooded areas was evaluated. The following hazardous elements were selected for research: Cd, Cu, Ni, Pb and Zn. The soil samples were taken from six locations (flooded annually, irregularly and rarely) to the north and south of the city of Olomouc. Physical and chemical properties of fluvisols were selected. The evaluation was carried out according to legislative regulations and using the results of the research results, regarding the load of hazardous substances on soil. The aim of the evaluation was to compare the soil load in individual areas and characterise the risk of contamination of plant production by hazardous substances.

The article deals with the problems of the load on soil in agricultural areas near the city of Olomouc, its industrial areas and the areas of fluvial zones with a load resulting from flooding by contaminated water. These soils have very specific character in terms of contamination because the sediments are very often transported by water recipients as well as various organic and inorganic contaminants.

The soil-plant system is the fundamental constructive unit of the geosphere and biosphere. Therefore, heavy metal pollution of soil has an important influence not only on the yield and quality of crops, but also on the quality of atmospheric and aquatic environment, and even on the health of human beings via food chains (Lasheen et al. 2000).

If we focus on localities influenced by flooding, the contamination of fluvisols with hazardous substances is still present and the content of hazardous substances is decreasing slowly. The following hazardous elements are the most common contaminants of fluvisols: Zn>Cd>Ni>Cu>Pb. The exceeding of the proposed indicative limit has been observed in the following cases. On the basis of the results presented, it can be concluded that values of heavy metals in Morava river fluvisols exceed the limits for all elements except lead in all localities except one (loam soil). The heaviest burden on soils is from zinc, while lead is below the limit. The situation is complicated by the increased solubility of hazardous elements in fluvisols and also by the increased risk of their transfer to plants.

8.2 The Effect of Heavy Metal Content of Floodplain Soils on its Accumulation in Plants with the Example of Carrots

This article deals with the effect of different pollution levels of soils and the accumulation of heavy metals in carrots. Fluvisols from the Morava floodplain area were used for the research. The soil samples were taken from three different locations (yearly, irregularly and rarely flooded). Homogenized sludge from a municipal wastewater treatment plant was artificially introduced to the soil samples. Analysis showed high levels of heavy metals in fluvisols with almost all elements, with the exception of lead. The yield of carrots was influenced by the amount of applied sludge, having the same effect as a fertilizer. Total increases in heavy metals in the plants were monitored in, relation to their content in the soil and after the addition of sludge. During the experiment relations between heavy metals in soil and plants were described. The results were subsequently evaluated using soil – plant transfer factors. A higher sludge pH level and lower amount of heavy metals in applied sludge partly limited the introduction of most heavy metals into the plants. Transfer factors closely relate to the quality of the resulting product – carrots. The research results indicate the necessity to monitor hazardous elements in polluted fluvisols, presuming that unless the limit for common soils is exceeded, the monitored produce should not be polluted with contents of these elements in excess of limits.

The results of fluvisol studies show above-limit concentrations of some, potentially hazardous elements in sandy soils. The results of pollution in fluvisols are in accordance with the aforementioned studies. The results indicate the necessity to monitor hazardous elements in the soils of alluvial areas, presuming that unless the limit for normal soils is exceeded, the monitored produce should not be polluted with content of these elements in excess of limits.

8.3 The Effect of Different Pollution Levels in Fluvisols on the Accumulation of Heavy Metals in Carrots (Daucus carota L.) and Spring Barley (Hordeum vulgare L.)

This article deals with the effect of different pollution levels of soils with an accumulation of heavy metals by two plants. Fluvisols from the Morava floodplain area were used. The soil samples were taken from three different localities (yearly, irregularly and rarely flooded). Homogenized sludge from a municipal wastewater treatment plant was artificially introduced into the samples.

Two types of accumulative plants were used: the Rubina variety of Daucus carota and the Amulet variety of Hordeum vulgare. In the following analysis of changes in soil characteristics were observed, i.e., mainly the heavy metal concentrations of cadmium, lead, zinc, copper and nickel in soils before the experiment, after finishing each cycle and in the plants themselves.

Analysis showed exceedingly high levels of heavy metals in fluvisols with almost all elements, with the exception of lead. Zinc and nickel had the highest concentrations. The yield of both plants was influenced by the amount of applied sludge, with the same effect as a fertilizer. On the other hand the high sludge pH level partly limited introduction of most heavy metals into the plants.

Total increases of heavy metals in the plants were monitored and evaluated by using transfer factors. They were compared at all locations including the differences between the controls and soil with the sludge, and total transfer factors focused on various elements. The research showed almost the same accumulation abilities of heavy metals except cadmium by both plants. As expected there was a higher accumulation potential in the root of D. carota and in the straw in H. vulgare.

9. General results

- The following hazardous elements are the most common contaminants of fluvisols: Zn>Cd>Ni>Cu>Pb.

- Values of heavy metals in Morava river fluvisols exceed the limits for all elements except lead in all localities except one (with loam soil).

- The heaviest burden on soils is from zinc, while lead is below the limit.

- The results confirmed the stagnant tendency of hazardous substance content in fluvisols of the Morava river after flooding.

- The average values of the transfer factors for D.carota are: cadmium 0.33, lead 0.004, copper 0.03, zinc 0.12 and nickel 0.1.

- Higher TF were primarily detected in the root of D.carota, except for zinc, equal values were found for lead and copper. Nickel values were two times higher. The lowest TF was evaluated for Pb.

- The results showed that the uptake of Cd, Cu, Ni and Zn by plants (including carrot) correspond to the increasing level of soil contamination, while the uptake of Pb was low. Soil to plant transfer factor values decreased from Ni > Zn > Cd > Cu > Pb.

- Soil pH ranges were from moderately acidic to neutral, pHKCl was from 6.5 to 6.9 and these values should reduce the transport of heavy metals from soil to plants.

- For H.vulgare the values of transfer factors are: cadmium 0.04, lead 0.003, copper 0.02, zinc 0.16 and nickel 0.1.

- If we focus on both plants – D.carota and H.vulgare, the higher TF levels are almost same except for cadmium (almost seven times higher) and zinc (almost two times higher) for D.carota.

- The most remarkable differences for TF are for cadmium in D.carota - values almost two times higher in fluvisols from Nemilany location than in the other two and for copper in D.carota and H.vulgare were found a difference of almost three times higher at the Novy Dvur location.

- Higher TF were detected in the root of D.carota except for zinc, equal values were found for lead and copper. Nickel values were two times higher.

- H.vulgare has a better phytoextraction potential in the straw than in the seed. Equal values were detected for copper. Nickel values were more than two times higher.

10. Conclusions

In general this project proved the phytoextraction potential for both plants grown on fluvisols mixed with sludge as a donor of elements (contamination of heavy metals in fluvisols was purposely left at a natural value). Use of this method for cleaning polluted fluvisols, under real conditions, would be very difficult - most of all due to the time required. This fact results from the comparison of the transfer factors of plants with higher phytoextraction potential.

The values of TF are generally lower. One possible explanation could be the higher pH level of the sewage sludge. The pH level of the sludge increased the soil reaction to alkaline (pHKCl=7.2). It could limit the transfer of heavy metals into the plants.

Sewage sludge from municipal wastewater treatment plants contains high quantities of nutrients and organic matter needed for plant growth, but it also contains heavy metal concentrations. Heavy metals can accumulate in soil and in plants when sludge is applied as a fertilizer and eventually can produce harmful effects in animals and humans. Therefore, the study of effective methods for heavy metal removal from sludge is very important in order to minimize prospective health risks during application.

In any case toxic metals from chemical factories can be spread by water and other factors into wide areas that can cause contamination of agricultural soils and, consequently, the food chain.

ACKNOWLEDGEMENT

I would like to acknowledge and thank to my supervisor prof. Dr. Ing. Bořivoj Šarapatka, CSc. for his number of valuable ideas, guidance and material and methodological comments. Further, I would like to thank Ing. Michaela Smatanová, Ph.D. for her support and guidance.

REFERENCES

Aksoy A, Sahün U, Duman F (2000) Robinia pseudo-acacia L. as a posssible biomonitor of heavy metal pollution in Kayseri. Turk J Bot 24: pp. 279.

Alloway BJ (1995) Heavy metals in soils, 2nd Ed. (pp. 122). London, U.K.: Blackie Academic & Professional.

Antić A, Cvetković O, Jovančićević B, Blagojević S, & Nikolić-Mandić S (2006) Eco-chemical characterisation of fluvisol of Velika Morava River valley (Serbia) based on the physico-chemical parameters and distribution of heavy metals. Fresenius' Environmental Bulletin, 15(11), pp. 1434.

Antunes PMC, Berkelaar EJ, Boyle D, Hale B, Hendershot W (2006) The biotic ligand model for plants and metals: technical challenges for field application. Environ Toxicol Chem 25:pp. 875.

Bethge-Steffens D (2008) Der Bodenwasserhaushalt von zwei repräsentativen Flussauenstandorten der Mittelelbe–Untersuchungen mit wägbaren Grundwasserlysimetern: Helmholtz Centre for Environmental Research–UFZ, Dissertation no. 01.

Baborowski M, von Tümpling W, Jr, & Friese K (2004) Behaviour of suspended particulate matter (SPM) and selected trace metals during 2002 summer flood in the River Elbe (Germany) at Magdeburg monitoring station. Hydrology and Earth System Sciences, 8(2), pp. 135.

Baker AJM and Brooks RR (1989) Terrestrial higher plants which hyperaccumulate metalic elements. A review of their distribution, ecology and phytochemistry. Biorecovery 1, pp. 81-126.

Banuelos GS, Ajwa HA, Mackey B, Wu L, Cook C, Akohoue S and Zambruzuski S (1997) Evaluation of different plant species used for phytoremediation of high soil selenium. J. Environ. Qual. 26: pp. 639-646.

Blaylock M, Huang J (2000) Phytoextraction of Metals. In: Raskin I, Ensley B (eds) Phytoremediation of toxic metals: Using plants to clean up the environment. Wiley Interscience, New York. Pp. 53.

Blaylock MJ, Salt DE, Dushenkov S, Zakharova O, Gussman C, Kapulnik Y, Ensley BD and Raskin I (1997) Enhanced accumulation of Pb in Indian mustard by soil-applied chelating agents. Environ. Sci. Technol. 31: pp. 860-865.

Burt R, Wilson M, Mays MD, Keck TJ, Fillmore M, Rodman AW, Alexander EB, Hernandez L (2000) Trace and major elemental analysis applications in the US Cooperative Soil Survey program. Commun Soil Sci Plant Anal 31: pp. 1757-1771.

Borůvka L, Huan-Wei C, Kozák J & Krištoufková S (1996) Heavy contamination of soil with cadmium, lead and zinc in the alluvium of the Litavka river. Rostlinná výroba, 42, pp. 543–550.

Böhme M (2006) Distribution of water quality parameters in two cross-sections of the River Elbe measured with high local, temporal, and analytic resolution. Acta Hydrochimica et Hydrobiologica, 34, pp. 201.

Brun LA, Maillet J, Hinsinger P, Pepin M (2001) Evaluation of copper availability to plants in copper-contaminated vineyard soils. Environ Pollut 111: pp. 293.

Becher M, Talke I, Krall L, Kramer U (2004) Cross-species microarray transcript profiling reveals high constitutive expression of metal homeostasis genes in shoots of the zinc hyperaccumulator Arabidopsis halleri. Plant J 37: pp. 251-261.

Burt R, Wilson M, Mays MD, Lee CW (2003) Major and trace elements of selected pedons in the USA. J Environ Qual 32: pp. 2109.

Büttner O, Otte-Witte K, Krüger F, Meon G & Rode M (2006) Numerical modelling of floodplain hydraulics and suspended sediment transport and deposition at the event scale in the middle River Elbe, Germany. Acta Hydrochimica et Hydrobiologica, 34, pp. 265.

Cannon H (1960) Botanical prospecting for ore deposits. Science 132: pp. 591-598. http://www.capecodonline.com/base/bullets.htm

CCME (Canadian Council of Ministers of the Environment) (2006) Canadian environmental quality guidelines, Ottawa, Canada. http://ceqg-rcqe.ccme.ca/. Accessed 14 August 2010.

Celik A, Kartal AA, Akdogan A, Kaka Y (2005) Determining the heavy metal pollution in Denizli (Turkey) by using Robinio pseudo-acacia L. Environ Int 31: pp. 105.

Chaney RL, Angle JS Li YM, Baker AJM (1999) Method for phytomining of nickel, cobalt and other metals from soil. U.S. Patent No. 5944872 (continuation in-part of US Patent 5711784 issued Jan. 27, 1998).

Chaney RL, Malik M, Li YM, Brown SL, Brewer EP, Angle JS and Baker AJM (1997) Phytoremediation of soil metals. Curr. Opin. Biotechnol. 8: pp. 279-284.

Charlesworth S, De Miguel E, Ordonez A (2010) A review of the distribution of particulate trace elements in urban terrestrial environments and its application to considerations of risk. Environ Geochem Health 33(2):pp. 103.

Chavez K (1999) Lead contamination spurs public meeting. Tennessean. http://www.tennessean.com/sii/99/08/14/lead14.shtml

Chlopecka A, Bacon J R, Wilson M J and Kay J (1996) Forms of cadmium, lead and zinc in contaminated soils from Southwest Poland, J. Environ. Qual. 25, pp. 69.

Cosio C, Keller C (2004) Hyperaccumulation of cadmium and zinc in Thlaspi caerulescens and Arabidopsis halleri at the leaf cellular level. Plant Physiol 134: pp. 716.

Cui Y J, Zhai R H, Huang Y Z, Qiu Y & Liang J Z (2005) Exposure to metal mixtures and human health impacts in a contaminated area in Nanning, China. Environment International, 31, pp. 784.

Cunningham SD and Ow DW (1996) Promises and prospects of phytoremediation. Plant Physiol. 110, pp. 715-719.

Davies J (1986) Occupational asthma caused by nickel salts. J Soc Occ Med 36: pp. 29.
Desenfant, F., Petrovský, E., & Rochette, P. (2004) Magnetic signature of industrial pollution of stream sediments and correlation with heavy metals: Case study from South France. Water, Air, and Soil Pollution, 152, pp. 297–312.

De Miguel E, Iribarren I, Chacon E, Ordonez A, Charlesworth S (2007) Risk-based evaluation of the exposure of children to trace elements in playgrounds in Madrid (Spain). Chemosphere 66:pp. 505.

Droppo IG, Lau YL & Mitchell C (2001) The effect of depositional history on contaminated bed sediment stability. Science of the Total Environment, 266, pp. 7.

Droppo IG, Leppard GG & Flannigan DT (1997) The freshwater floc: A functional relationship of water and organic and inorganic floc constituents affecting suspended sediment properties. Water, Air, and Soil Pollution, 99, pp. 43.

EPA (1989) EPA Superfund Record of Decision: Picatinny Arsenal (US Army). Rockaway Township, NJ, U.S. Environmental Protection Agency Superfund. http://www.epa.gov/superfund/sites/rods/fulltext/r0289093.pdf

Farmer JG, Mackenzie AB, Sugden CL, Edgar P J & Eades LJ (1997) A comparison of the historical lead pollution records in peat and freshwater lake sediments from central Scotland. Water, Air, and Soil Pollution, 100, pp. 253-266.

Fergusson JE (1990) The heavy elements: Chemistry, environmental impact and health ef fects (pp. 329). Oxford, U.K.: Pergamon Press plc. Headington Hill Hall.

Freeman JL, Persans MW, Nieman K, Albrecht C, Peer WA, Pickering IJ, Salt DE (2004) Increased glutathione biosynthesis plays a role in nickel tolerance in Thlaspi nickel hyperaccumulators. Plant Cell 16: pp. 2176.

Garcia G, Faz A, Cunha M (2004) Performance of Piptatherum miliaceum (Smilo grass) in edaphic Pb and Zn phytoremediation over a short growth period. Int Biodeter & Biodeg 54: pp. 245-250.

Gjoka F, Felix-Henningsen P, Wegener HR, Salillari I, Beqiraj A (2011) Heavy metals in soils from Tirana (Albania). Environ Monit Assess (2011) 172: pp. 517–527.

Grasmuck D, Scholz RW (2005) Risk perception of heavy metal soil contamination by high-exposed and low-exposed inhabitants: the role of knowledge and emotional concerns. Risk Anal 25:pp. 611.

Grotz N, Fox T, Connolly E, Park W, Guerinot ML, Eide D (1998) Identification of a family of zinc transporter genes from Arabidopsis that respond to zinc deficiency. Proc Natl Acad Sci USA 95: pp. 7220-7224.

Gröngröft A, Krüger F, Grunewald K, Meißner R & Miehlich G. (2005) Plant and soil contaminationwith trace metals in the Elbe floodplains: A case study after the flood in August 2002. Acta Hydrochimica et Hydrobiologica, 33(5), pp. 466–468.

Grygar T, Světlík I, Lisá L, Koptíková L, Bajer A, Wray DS, et al. (2010) Geochemical tools for the stratigraphic correlation of floodplain deposits of the Morava River in Strážnické Pomoraví, Czech Republic from the last millennium. Catena, 80, pp. 106–121.

Gupta UC, Gupta SC (1998) Trace element toxicity relationships to crop production and livestock and human health: Implications for management. Commun Soil Sci Plant Anal 29: pp. 1491.

Hall JL (2002) Cellular mechanisms for heavy metal detoxification and tolerance. J Exp Bot 53: pp.1-3.

Hernando S, Lobo MC, Polo A (1989) Effect of the application of a municipal refuse compost on the physical and chemical properties of a soil. Sci Total Environ 81:pp. 589.

Huang JW and Cunningham SD (1996) Lead phytoextraction: Species variation in lead uptake and translocation. New Phytol. 134: pp. 78-81.

Hudson-Edwards KA, Macklin MG, Curtis CD & Vaughan DJ (1998) Chemical remobilization of contaminant metals within floodplain sediments in an incising river system: Implications for dating and chemostratigraphy. Earth Surface Processes and Landforms, 23, pp. 671–684.

Johnson D, Hale B (2004) White birch (Betula papyrifera Marshall) foliar litter decomposition in relation to trace metal atmospheric inputs at metal-contaminated and uncontaminated sites near Sudbury, Ontario and Rouyn-Noranda, Quebec, Canada. Environ Pollut 127: pp. 72.

Kadlec J, Grygar T, Světlík I, Ettler V, Mihaljevič M, Diehl JF, et al. (2009) Morava River floodplain development during the last millennium, Strážnické Pomoraví, Czech Republic. The Holocene, 19, pp. 499–509.

Kasuya M (2000) Recent epidemiological studies on itai-itai disease as a chronic cadmium poisoning in Japan.Water Sci Technol 42:pp. 147.

Kerkeb L, Kramer U (2003) The role of free histidine in xylem loading of nickel in Alyssum lesbiacum and Brassica juncea. Plant Physiol 131: pp. 716-24.

Klavins M, Briede A, Rodinov V, Kokorite I, Parele E and Klavina I (2000) Heavy metals in rivers of Latvia, Sci. Total Environ. 262, pp. 175.

Kober B, Wessels M, Bollhofer A & Mangini A (1999) Pb isotopes in sediments of Lake Constance, Central Europe constrain the heavy metal pathways and the pollution history of the catchment, the lake and the regional atmosphere. Geochimica et Cosmochimica Acta, 63, pp. 1293-1299.

Kramer U, Smith RD, Wenze WW, Raskin I and Salt DE (1997) The role of metal transport and tolerance in nickel hyperaccumulation by Thelapsi geosingense Halacsy. Plant Physiol. 115, pp. 1641-1650.

Krüger F, Schwartz R, Kunert M & Friese K (2006) Methods to calculate sedimentation rates of floodplain soils in the middle region of the Elbe River. Acta Hydrochimica et Hydrobiologica, 34, pp. 175.

Kumar P, Dushenkov V, Motto H, Raskin I (1995) Phytoextraction - the use of plants to remove heavy-metals from soils. Environ Sci Technol 29: pp. 1232-1238.
Kupper H, Lombi E, Zhao FJ, McGrath SP (2000) Cellular compartmentation of cadmium and zinc in relation to other elements in the hyperaccumulator Arabidopsis halleri. Planta 212: pp. 75-76.

Kupper H, Zhao F, McGrath SP (1999) Cellular compartmentation of zinc in leaves of the hyperaccumulator Thlaspi caerulescens. Plant Physiol 119: pp. 305.

LABO (1995) Soil background and reference values in Germany (pp. 48). Report.

Lacatusu R (1995) Appraising levels of soil contamination and pollution with heavy metals (no. 4, pp. 393). Research report, European Soil Bureau.

Lasat MM, Baker A, Kochian LV (1996) Physiological characterization of root Zn2+ absorption and translocation to shoots in Zn hyperaccumulator and nonaccumulator species of Thlaspi. Plant Physiol 112: pp. 1715-1722.

Lasat MM, Baker AJ, Kochian LV (1998) Altered Zn compartmentation in the root symplasm and stimulated Zn absorption into the leaf as mechanisms involved in Zn hyperaccumulation in Thlaspi caerulescens. Plant Physiol 118: pp. 875-883.

Lasheen MR, Ashmaway A, Ibrahim H (2000) Heavy metal removal from municipal and industrial sludge. In: Nriagu, J. (ed.): 11th Annual International Conference on Heavy metals in the Environment, Contribution 1431. University Of Michigan, School of Public Health, Ann Arbor, MI (CD-ROM).

Lewin J & Macklin MG (2003) Preservation potential for Late Quaternary river alluvium. Journal of Quaternary Science, 18, pp. 107–120.

Lombi E, Hamon RE, Wieshammer G, McLaughlin MJ, McGrath SP (2004) Assessment of the use of industrial by-products to remediate a copper- and arsenic-contaminated soil. J Environ Qual 33: pp. 902.

Lombi E, Zhao FJ, Dunham SJ, McGrath SP (2000) Cadmium accumulation in populations of Thlaspi caerulescens and Thlaspi goesingense. New Phytol 145: pp. 11-12.

Lombi E, Zhao FJ, Dunham SJ and McGrath SP (2000) Phytoremediation of Heavy Metal-Contaminated Soils: Natural Hyperaccumulation versus Chemically Enhanced Phytoextraction. Technical report - Bioremediation and Biodegradation.

Lombi E, Tearall KL, Howarth JR, Zhao FJ, Hawesford MJ, McGrath SP (2002a) Influence of iron status on cadmium and zinc uptake by different ecotypes of the hyperaccumulator Thlaspi caerulescens. Plant Phyiol 128: pp. 1359-1367.

Lombi E, Zhao F, Fuhrmann M, Ma LQ, McGrath SP (2002b) Arsenic distribution and speciation in the fronds of the hyperaccumulator Pteris vittata. New Phytol 156: pp. 195.

Lombi E, Zhao F, McGrath S, Young S, Sacchi G (2001a) Physiological evidence for a high-affinity cadmium transporter highly expressed in a Thlaspi caerulescens ecotype. New Phytol 149: pp. 53.

Lombi E, Zhao FJ, Dunham SJ, McGrath SP (2001b) Phytoremediation of heavy metalcontaminated soils: natural hyperaccumulation versus chemically enhanced phytoextraction. J Environ Qual 30: pp. 1919.

Lupankwa K, Love D, Mapani, BS, Mseka, S (2004) Impact of a base metal slimes dam on water systems, Madziwa Mine, Zimbabwe. Physics Chem Earth 29: pp. 1151.

Macklin MG and Klimek K (1992) Dispersal, storage and transformation of metal contaminated alluvium in the Upper Vistula basin, southwest Poland, Appl. Geogr. 12, pp. 30.

Markert B (1994) Plants as Biomonitors for Heavy Metal Pollution of the Terrestrial Environment. VCH, Weinheim.

Martin CW (2000) Heavy metal trends in floodplain sediments and valley fill, River Lahn, ermany. Catena, 39, pp. 53–68.

Martínez-Cortizas A, Pontevedra-Pombal X, Nóvoa Muñoz JC & García-Rodeja E (1997) Four thousand years of atmospheric Pb, Cd and Zn deposition recorded by the ombrotrophic peat bog of Penido Vello (northwestern Spain). Water, Air, and Soil Pollution, 100, pp. 387–403.

Mebane C (1997) Use Attainability Analysis Blackbird Creek Lemhi County Idaho, Idaho Division of Environmental Quality, Water Quality Assessment and Standards Bureau and U.S. Environmental Protection Agency Region 10.

Meharg A, Macnair M (1992a) Genetic correlation between arsenate tolerance and the rate of influx of arsenate and phosphate in Holcus lanatus L. Heredity 69:pp. 336.

Meharg A, Macnair M (1992b) Suppression of the high-affinity phosphate-uptake system - a mechanism of arsenate tolerance in Holcus lanatus L. J Exp Botany 43: pp. 519.

Middelkoop H (2000) Heavy-metal pollution of the river Rhine and Meuse floodplains in The Netherlands. Geologie en Mijnbouw-Netherlands Journal of Geosciences, 79, pp. 411–427.

Moiseenko TI, Gashkina NA., Sharova YN & Kudryavtseva LP (2008) Ecotoxicological assessment of water quality and ecosystem health: A case study of the Volga River. Ecotoxicology and Environmetal Safety, 71, pp. 837.

Morales CFL, Strathmann M & Flemming HC (2007) Role of biofilms on sediment transport—Investigations with artificial sediment columns. In B. Westrich & U. Förstner (Eds.), Sediment dynamics and pollutant mobility in rivers (pp. 358–368). Berlin: Springer.

Markus J, McBratney AB (2001) A review of the contamination of soil with lead II, spatial distribution and risk assessment of soil lead. Environ Int 27:pp. 399.

Muller M, Anke M, Hartmann E and Illing-Gunther H (1996) Oral cadmium exposure of adults in Germany. 1. Cadmium content of foodstuffs and beverages, Food Additives and Contaminants 13, pp. 359.

Nikarnorov AM, Bryzgalo VA & Chernogaeva GM (2007) Anthropogenically modified natural background and its formation in the Russian freshwater ecosystems. Russian Meteorology and Hydrology, 32(11), pp. 698.

Novák M, Emmanuel S, Vile MA, Erel Y, Véron A, Pačes T, et al. (2003) Origin of lead in eight Central European peat bogs determined from isotope ratios, strengths, and operation times of regional pollution sources. Environmental Science and Technology, 37, pp. 437–445.

Nováková T (2009) Geochemické a izotopové datování recentních povodňových sedimentů řeky Moravy (Geochemical and isotopical dating of recent flood sediments of Morava River), MSc Thesis, Faculty of Science, Charles University, Prague, Czech Republic.

Papagiannis I, Kagalou I, Leonardos, J, Petridis, D, Kalfakakou, V (2004) Copper and zinc in four freshwater fish species from Lake Pamvotis (Greece). Environ Int 30: pp. 357.

Peer WA, Mamoudian M, Lahner B, Reeves, RD, Murphy AS, Salt DE (2003) Identifying model metal hyperaccumulating plants: germplasm analysis of 20 Brassicaceae accessions from a wide geographic area. New Phytol 159: pp.421.

Pence NS, Larsen PB, Ebbs SD, Letham DL, Lasat MM, Garvin DF, Eide D, Kochian LV (2000) The molecular physiology of heavy metal transport in the Zn/Cd hyperaccumulator Thlaspi caerulescens. Proc Natl Acad Sci USA 97: pp. 4956.

Pepelnik R, Karrasch B, Niedergesäß R, Erbslöh B, Mehrens M, Link U, et al. (2005) Influence of the flooding in 2002 on the plankton and the quality of water and sediment of the River Elbe over its longitudinal profile. Acta Hydrochimica et Hydrobiologica, 33(5), pp. 430.

Petticrew EL & McConnachie JL (2007) Sediment–water interactions. In B. Westrich & U. Förstner (Eds.), Sediment dynamics and pollutant mobility in rivers (pp. 217). Berlin: Springer.

Pierzynski GM, Sims JT, Vance GF (2005) Soils and environmental quality. Taylor and Francis, Boca Raton.

Piñeros M, Kochian L (2003) Differences in whole-cell and single-channel ion currents across the plasma membrane of mesophyll cells from two closely related Thlaspi species. Plant Physiol 131: pp. 583.

Pruvot C, Douay F, Hervé F, Waterlot C (2006) Heavy metals in soil, crops and grass as a source of human exposure in the former mining areas. J Soils Sediments 6:pp. 215.

Pyatt FB (2001) Copper and lead bioaccumulation by Acacia retinoides and Eucalyptus torquata in sites contaminated as a consequence of extensive ancient mining activities in Cyprus. Ecotoxicol Environ Saf 50: pp. 60.

Ramesh SA, Shin R, Eide DJ, Schachtman DP (2003) Differential metal selectivity and gene expression of two zinc transporters from rice. Plant Physiol 133: pp. 126.

Ramesh SA, Shin R, Eide DJ, Schachtman DP (2003) Differential metal selectivity and gene expression of two zinc transporters from rice. Plant Physiol 133: pp. 126.

Renberg I, Bindler R & Brannvall ML (2001) Using the historical atmospheric lead-deposition record as a chronological marker in sediment deposits in Europe. The Holocene, 11, pp. 511–516.

Salt DE, Pickering IJ, Prince RC, Gleba D, Dushenkov S, Smith RD, Raskin I (1997) Metal accumulation by aquacultured seedlings of Indian mustard. Environ Sci Technol 31: pp. 1635.

Sahi SV, Bryant NL, Sharma NC, Singh SR (2002) Characterization of a lead hyperaccumulator shrub, Sesbania drummondii. Environ Sci Technol 36: pp. 4676.

Schwartz R (2006) Geochemical characterisation and erosion stability of fine-grained groyne field sediments of the middle Elbe River. Acta Hydrochimica et Hydrobiologica, 34, pp. 223.

Shen ZG, Zhao FJ and McGrath SP (1997) Uptake and transport of zinc in the hyperaccumulator Thlaspi caerulescens and the non-hyperaccumulator Thlaspi ochroleucum. Plant Cell Environ. 20: pp. 898.

Soumare M, Tack FMG, Verloo MG (2003) Characterisation of Malian and Belgian solid waste composts with respect to fertility and suitability for land application. Waste Manag 23:pp. 517.

Steinnes E, Allen RO, Petersen HM, Rambaek JP and Varskog P (1997) Evidence of large scale heavy-metal contamination of natural surface soils in Norway from long-range atmospheric transport, Sci. Total Environ. 205, pp. 255.

Sterckeman T, Douay F, Priox N and Fourrier H (2000) Vertical distribution of Cd, Pb and Zn in soils near smelters in the North of France, Environ. Pollut. 107, pp. 377.

Stevenson FJ (1994) Humus chemistry: genesis, composition, reactions, 2nd edn. John Wiley and Sons, New York.

Van der Veen A, Ahlers C, Zachmann DW & Friese K (2006) Spatial distribution and bonding forms of heavy metals in sediment along the middle course of the River Elbe (km 287–390). Acta Hydrochimica et Hydrobiologica, 34, pp. 214.

Watanabe ME (1997) Phytoremediation on the brink of commercialization. Environ. Sci. Technol. 31: pp. 182-186.

Weiss D, Shotyk W, Appleby PG, Kramers ID & Cheburkin AK (1999) Atmospheric Pb deposition since the industrial revolution recorded by five Swiss peat profiles: Enrichment factors, fluxes, isotopic composition, and sources. Environmental Science and Technology, 33, pp. 1340–1352.

Witter B, Winkler M & Friese K (2003) Depth distribution of chlorinated and polycyclic aromatic hydrocarbons in floodplain soils of the River Elbe. Acta Hydrochimica et Hydrobiologica, 31(4–5), pp. 411.

Wycisk K, Kim EJ, Schroeder JL, Kramer U (2004) Enhancing the first enzymatic step in the histidine biosynthesis pathway increases the free histidine pool and nickel tolerance in Arabidopsis thaliana. FEBS Lett 578: pp. 128-134.

Xintaras C (1992) Analysis paper: Impact of lead-contaminated soil on public health. U.S. Department of Health and Human Services, Public Health Service, Agency for Toxic Substances and Disease Registry. http://www.atsdr.cdc.gov/cxlead.html.

Zantopoulos N, Antoniou V, Nikolaidis E (1999) Copper, zinc, cadmium, and lead in sheep grazing in North Greece. Bull Environ Cont Tox 62: pp. 691.

Zhao F, Lombi E, Breedon T, McGrath SP (2000) Zinc hyperaccumulation and cellular distribution in Arabidopsis halleri. Plant Cell Envirion 23: pp. 507-514.

APPENDIX

JAN KOČAŘ, BOŘIVOJ ŠARAPATKA

Department of Ecology and Environmental Science, Palacký University, tr. Svobody 26, 771 46 Olomouc, Czech Republic, e-mail: borivoj.sarapatka@upol.cz

STATE OF CONTAMINATION OF FLUVISOLS OF THE MORAVA RIVER BY POTENTIALLY HAZARDEOUS ELEMENTS

Abstract

Kočař, J., Šarapatka, B., 2007: State of contamination of fluvisols of the Morava river. Phytopedon (Bratislava), Vol. 6, 2007/2: p. xx–xx.

The content of potentially hazardous elements – heavy metals – in samples of fluvisols from flooded areas was evaluated. The following hazardous elements were selected for research: Cd, Cu, Ni, Pb and Zn. The soil samples were taken from six locations (flooded annually, irregularly and rarely) to the north and south of the city of Olomouc. Physical and chemical properties of fluvisols were selected. The evaluation was carried out according to legislative regulations and using the results of the research results, regarding the load of hazardous substances on soil. The aim of the evaluation was to compare the soil load in individual areas and characterise the risk of contamination of plant production by hazardous substances.

Key words: contamination, floods, fluvisols, heavy metals

INTRODUCTION

The article deals with the problems of the load on soil in agricultural areas near the city of Olomouc, its industrial areas and the areas of fluvial zones with a load resulting from flooding by contaminated water.

These soils have very specific character in terms of contamination because the sediments are very often transported by water recipients as well as various organic and inorganic contaminants. Their accumulation in the soil profile differs to a greater or lesser extent. In ever-increasing flooding, mainly caused by changes in climate, contaminants are transported on the soil's surface, and percolate into groundwater, or the flow of contaminated water influences the quality of groundwater. If we look at this soil in terms of contamination it can be classified as the most hazardous type of soil. The accumulation of contaminants in the soil profile usually comes from a wide-spread area, and also the transport of contaminants into other parts of the environment is stronger (KOBZA A MATÚŠKOVÁ 2003).

The soil-plant system is the fundamental constructive unit of the geosphere and biosphere. Therefore, heavy metal pollution of soil has an important influence not only on the yield and quality of crops, but also on the quality of atmospheric and aquatic environment, and even on the health of human beings via food chains (LASHEEN et al. 2000).

This type of soil is situated on a flat plain of predominantly arable land with crop-plants. For this reason fluvisols deserve more attention in terms of protection and hygiene (KOBZA A MATÚŠKOVÁ 2003).

MATERIALS AND METHODS

Research was carried out on samples from fluvisols of the Morava river floodplain. The samples were collected from six different locations contaminated with heavy metals from flooding in July 1997. The first group of soils is situated to the south of Olomouc with specific localities Novy Svet (flooded every year), Nemilany (flooded irregularly) and Novy Dvur (rarely flooded). The second group comes from localities situated to the north of Olomouc – Cernovir (flooded every year), Chomoutov (flooded irregularly) and Horka nad Moravou (rarely flooded). The samples were taken from a depth of 0–20 cm. For our research the following elements were selected: Cd, Cu, Ni, Pb and Zn.

Methods of analysis

The samples were air-dried and ground prior to analysis. Total heavy metal content was determined by atomic absorption spectroscopy (AAS) using flame (WEI 1992).

Soil samples were analyzed for: value of CEC, granular composition, content of humus, content of N_t, $pH_{(KCl)}$, content of nutrients by Mehlich III. method (ZBIRAL 1995). Only some of the results of this analysis were used for this article.

RESULTS

Heavy metal content of fluvisols

The heavy metal content of fluvisols from the Morava river is given in Tab 1. As expected, the total metal content of fluvisols including Cd, Cu, Ni, Pb and Zn differed in all six locations. The first group of soils taken from the Novy Svet locality is exceptionally high in total amount of Cd, Ni and Zn. The high level of metals from flooding (the biggest flood in recent years occurred in July 1997) coming from Olomouc industrial region led to anomalous large concentrations of these metals in the soil. The concentrations were higher than limits set in government regulations – Cd by about 23%, Ni by 38% and Zn by about 54% in the Novy Svet locality. The concentration of metals from Nemilany soil exceeded the limits in Cd by about 11%, Cu by 29%, Ni by 50% and Zn by 48%. Only soil from Novy Dvur did not exceed the limits in any heavy metal levels.

The second group of soil samples taken from the Cernovir locality exceeded the limits only in Cd by about 15% and markedly in Zn by about 118.7%. The situation was very similar in the Chomoutov locality where limits in Zn were exceeded by about 93.9% and in the Horka nad Moravou locality by about 75.6%.

Figure 1 shows normal values and excess values of heavy metal concentration in fluvisols of the Morava river.

Soil pH ranges from acid level to almost neutral, pH_{KCl} is from 6.6 to 6.9. This fact should reduce the transfer of heavy metals from soil to plants (HERMS 1989, COTTENIE, KIEKENS 1981, SAUERBECK 1989).

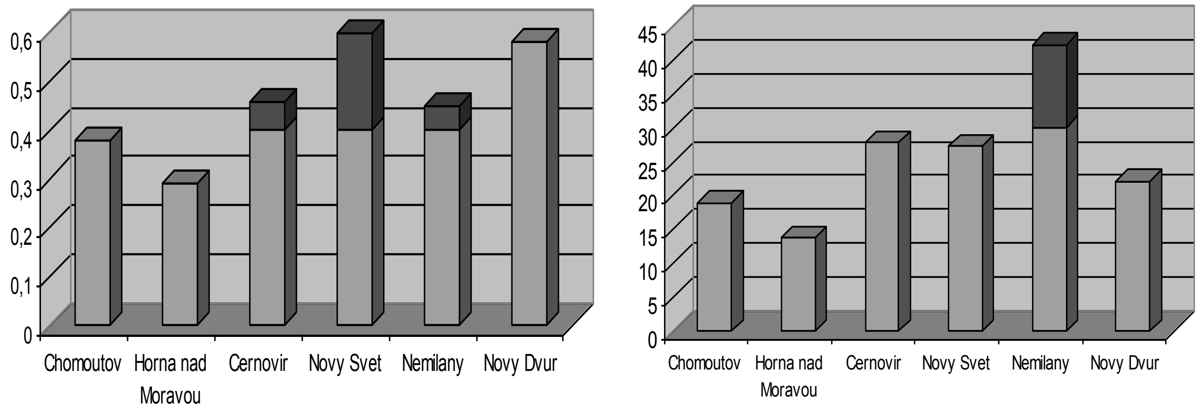
Chosen physical and chemical properties of fluvisols are given in Tab 2. There are two types of soil, light loamy-sand soil from Novy Svet, Nemilany, Cernovir, Chomoutov and Horka nad Moravou and loam soil from the Novy Dvur locality. These differences influence the division of soils into two groups regarding heavy metal concentration and its limits.

Table 1. Concentration of heavy metals (in mg kg^{-1}) in fluvisols of the Morava river (2M HNO_3). Values tagged with asterisk exceed the limits (different limits for two types of soil – light loamy-sand and loam).

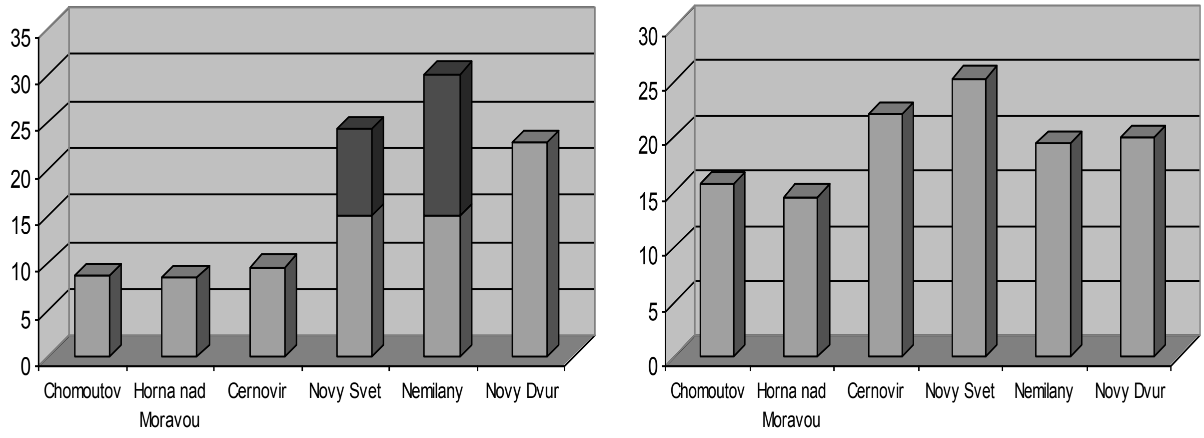
Locality	*Cd*	*Cu*	*Ni*	*Pb*	*Zn*
Novy Svet	0.6*	27.24	24.31*	25.3	108.5*
Nemilany	0.45*	42.24*	30.12*	19.4	95.57*
Cernovir	0.46*	27.78	9.6	21.98	109.36*
Chomoutov	0.38	18.97	8.72	15.68	96.96*
Horka nad Moravou	0.29	13.74	8.5	14.54	87.79*
Limits	0.4	30	15	50	50
Novy Dvur	0.58	22.14	22.88	19.9	87.13
Limits	1	50	25	70	100

Table 2. Physical and chemical properties of soils.

Parameters	*Unit*	*Novy Svet*	*Nemilany*	*Novy Dvur*	*Cernovir*	*Chomoutov*	*Horka nad Moravou*
<0.01mm	%	12.7	10.7	33	14	11	9.2
<0.001mm	%	1.6	2	9.2	1.2	3.1	3.9
0.01–0.05mm	%	26.3	25.5	35.8	28.3	22.8	21.6
0.05–0.25mm	%	48.8	43.3	28.2	45	32.9	47
0.25–2.0mm	%	12.2	20.5	3	16	19.3	15.1
Soil	%	light loamy-sand	light loamy-sand	loam	light loamy-sand	light loamy-sand	light loamy-sand
N_{tot}	$mg.kg^{-1}$	2664.87	3420.65	2377.08	2213.77	3332.67	3967.76
C_{org}	%	2.71	2.36	2.59	3.5	2.66	2.82
Humus	%	4.68	4.07	4.47	4.89	3.84	4.34


(a) Cadmium

(b) Copper


(c) Nickel

(d) Lead

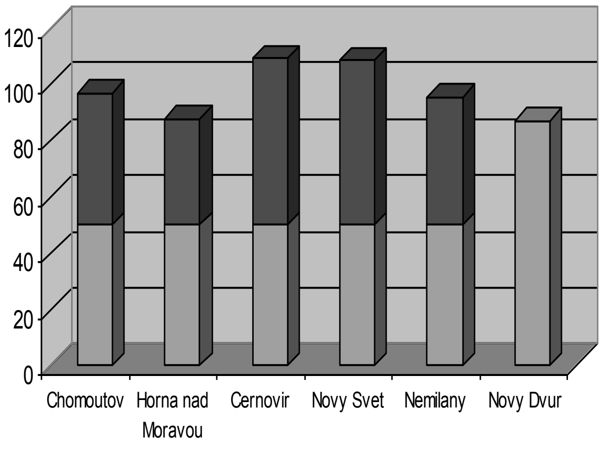

(e) Zinc

Figure 1. Concentration of heavy metals (mg kg^{-1}) in fluvisols of the Morava river (2M HNO_3) – normal values (in grey) and excess values (over the limit, dark grey) for Chomoutov, Horka nad Moravou, Cernovir, Novy Svet and Nemilany (light loamy-sand soil), different limits for Novy Dvur (loam soil).

DISCUSSION AND CONCLUSIONS

If we focus on localities influenced by flooding, the contamination of fluvisols with hazardous substances is still present and the content of hazardous substances is decreasing slowly. The following hazardous elements are the most common contaminants of fluvisols: Zn>Cd>Ni>Cu>Pb. The exceeding of the proposed indicative limit has been observed in the following cases. On the basis of the results presented, it can be concluded that values of heavy metals in Morava river fluvisols exceed the limits for all elements except lead in all localities except Novy Dvur (loam soil). The heaviest burden on soils is from zinc, while lead is below the limit.

The situation is complicated by the increased solubility of hazardous elements in fluvisols and also by the increased risk of their transfer to plants.

In Fig 2 values of heavy metals in all localities are compared with average values for rivers in Slovakia. The results of this study confirmed the stagnant tendency of hazardous substance content in fluvisols of the Morava river after flooding.

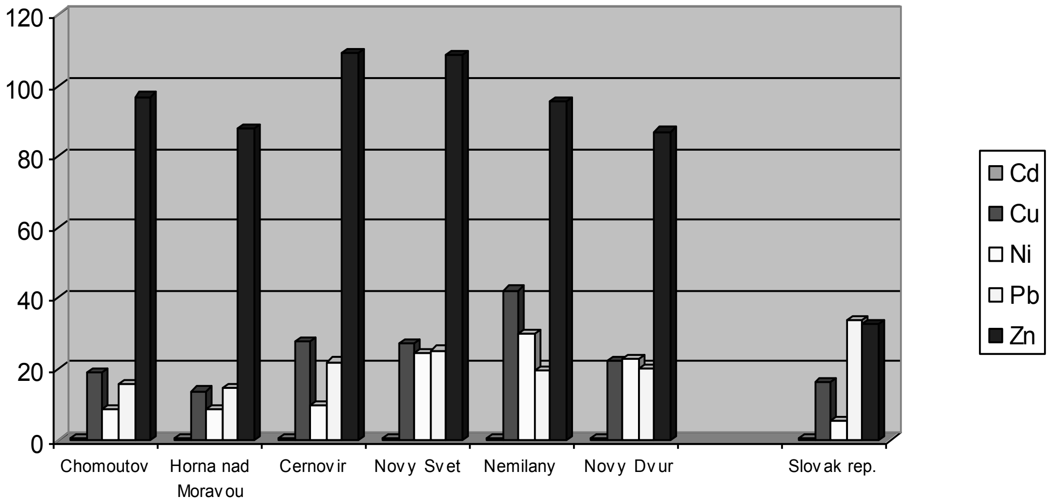


Figure 2. Comparison of heavy metal concentration in fluvisols of the Morava river (average values in Olomouc region) vs. fluvisols from Slovakia (average values for the country) (KOBZA 2003).

On the basis of the findings, the theme of fluvisol contamination is still topical due to its high potential in crop production (VÁCHA ET AL. 2006).

The use of fluvisols in the fluvial zones for plant production must follow the legislative standards set for the protection of the food chain. Monitoring the level of hazardous substances in individual locations used for plant production is highly recommended (VÁCHA ET AL. 2006).

Translated by the authors.

ACKNOWLEDGEMENT

The authors wish to thank the Czech Ministry of Education, Youth and Sports for supporting their research through Grant No 2B06101 “The optimization of both agricultural and river landscape in the Czech Republic with an emphasis on the biodiversity process” from the National Research Program II.

REFERENCES

KOBZA, J., MATÚŠKOVÁ, L., 2003: Kontaminácia pôd na fluviálnych sedimentoch v podmienkach Slovenska. Pedologické dny 2003, Sborník príspevku z konference “Ochrana a využití půdy v nivních oblastech”, Velké Bílovice, Czech republik: p. 33.

LASHEEN, M. R., ASHMAWAY, A.,IBRAHIM, H., 2000: Heavy metal removal from municipal and industrial sludge. In: Nriagu, J. (ed.): 11th Annual International Conference on Heavy metals in the Environment, Contribution 1431. University Of Michigan, School of Public Health, Ann Arbor, MI (CD-ROM).

VÁCHA, R., VYSLOUŽILOVÁ, M., HORVATHOVÁ, V., ČECHMÁNKOVÁ, J., 2006: Risks following from Husbandry on Agricultural Soils in loaded areas of the Czech republic. Soil and Water Res. 1, 3: p.108–116.

WEI, F.S., 1992: Modern Analytical Methods of Soil Elements. Chinese Environ. Sci. Press, Beijing, 64 pp.

ZBÍRAL, J., 1995: Analýza půd I. - jednotné pracovní postupy. ÚKZÚZ Brno, p. 87-114.

The Effect of Heavy Metal Content of Floodplain Soils on its Accumulation in Plants with the Example of Carrots

B. ŠARAPATKA,[1] J. KOČAŘ[1], M. SMATANOVÁ[2]

[1] Department of Ecology and Environmental Sciences, Palacký University,
tř. Svobody 26, 771 46 Olomouc, Czech Republic
[2] Central Institute for Supervising and Testing in Agriculture, Hroznová 2,
656 06 Brno, Czech Republic

ABSTRACT: This article deals with the effect of different pollution levels of soils and the accumulation of heavy metals in carrots. Fluvisols from the Morava floodplain area were used for our research. The soil samples were taken from three different locations (yearly, irregularly and rarely flooded). Homogenized sludge from a municipal wastewater treatment plant was artificially introduced to the soil samples. Analysis showed high levels of heavy metals in fluvisols with almost all elements, with the exception of lead. The yield of carrots was influenced by the amount of applied sludge, having the same effect as a fertilizer. Total increases in heavy metals in the plants were monitored in, relation to their content in the soil and after the addition of sludge. During our experiment relations between heavy metals in soil and plants were described. The results were subsequently evaluated using soil – plant transfer factors. A higher sludge pH level and lower amount of heavy metals in applied sludge partly limited the introduction of most heavy metals into the plants. Transfer factors closely relate to the quality of the resulting product – carrots in our case. The research results indicate the necessity to monitor hazardous elements in polluted fluvisols, presuming that unless the limit for common soils is exceeded, the monitored produce should not be polluted with contents of these elements in excess of limits.

Keywords: accumulation, fluvisols, sludge, heavy metals, carrot

INTRODUCTION

Heavy metal pollution of soils is increasingly becoming a global problem with the development of industry, mining activity, waste water irrigation and the application of sewage sludge, even if it is relatively localized at present. The soil-plant system is the fundamental building block of the geosphere and biosphere. Therefore, heavy metal pollution of the soil has an important influence not only on the yield and quality of crops, but also on the quality of the atmospheric and, aquatic environment, and even on the health of human beings via the food chains (LASHEEN et al. 2000; YAMAGATA, SHIGEMATSU 1970).

The problems of heavy metal pollution are also present in fluvisol type soils because of contaminated solid, deposits. As a result of current landscape management the floodplains are very intensive agricultural areas with low ecological status (ŠARAPATKA, ŠTĚRBA 1998) and most of the floodplains in central Europe are also contaminated by numerous pollutants (SCHWARTZ et al. 2006). In research conducted in the Spittelwasser floodplain SCHWARTZ et al. also described how the alluvial soil profile is severely contaminated by numerous inorganic and organic pollutants. MIDDELKOOP (2000) studied the accumulation of heavy metals in the floodplain of the lower Rhine river. The largest metal accumulations occur in low-lying floodplain sections, the lowest was found in the distal part of the floodplain area. Similar results are published by KRÜGER et al. (2005) for Elbe river floodplain soils in which high amounts of heavy metals accumulated over at least the last century. The floodplain section downstream of the river Mulde inflow is much more contaminated than the upstream section. The degree of contamination of the soil in floodplain and depths depending on, particular conditions was published by SIERRA et al. (2000). PETROVICH-GEGIC et al. (2007) studied heavy metal accumulation in the alluvial banks of the Tisa River and the residual concentration of metals available to the plants, including carrots. EBBS et al. (2006) grew plants incl. carrots in pots containing sediment from the Illinois River. Some these plant species showed significantly greater biomass and yield compared to plants from the reference soil and elemental analysis of the tissue revealed that Zn and Mo were the only elements that were significantly greater in sediment-grown plants on a consistent basis.

An important source of heavy metal soil contamination can become industrial compost, which is primarily produced from sewage sludge. At present we do not have enough knowledge of the negative effects of repeated compost application (with different levels of heavy metals) on soil and plants (VÁŇA 2003). PEREZ et al. (2007) described the effect on plants caused by increasing rates of application of composted municipal waste. Nevertheles, the levels of accumulation in both soil and plants were within permissible limits. A linear relationship between total soil and crop metal concentration in a long-term field experiment with carrots and other vegetables and crops after sludge application was also described by MCGRATH et al. (2000).

The aim of this research is to find out the accumulation of heavy metals in carrots (*Daucus carota* L.) from fluvisols with different concentrations of these metals after flooding and the application of sewage sludge and to describe the limits in these regions with the aim of producing good quality vegetables.

MATERIALS AND METHODS

Pot Experiment

In our experiment we grew carrots (*Daucus carota* L. var. Rubina) in soils mixed with various amounts of sewage sludge. A two-year experiment was carried out on samples from the fluvisols of the Morava River floodplain area. The samples were collected from three different locations (yearly, irregularly and rarely flooded, at depths of 0 - 20 cm) and the samples were used for pot experiments. In total we used in this experiment 36 pots per year (9 control pots) with 8kg portions of the soil in two periods. A total of 72 pots were used. All combinations were replicated three times.

The soil was air-dried, crushed, mixed thoroughly and passed through a 1 cm sieve. 8 kg portions of the soil were transferred to plastic pots and treated with the necessary amounts of fertilizer at a rate of 0.6 g super phosphate, 0.6 g potassium chloride and 0.53 g urea and then mixed well.

Stabilized sludge, which was obtained from a municipal wastewater treatment plant in Olomouc, was then added to the pots. The soils were mixed separately with sewage sludge at application rates of 0, 2.5, 5 and 10 tonnes/ha (according to public notice Nr.382/2001). A total of 700 carrots were grown in the pots and, the same as the soils, subsequently underwent detailed analysis.

Analytical Methods

Soil samples were taken from the pots. The samples were air-dried and ground prior to analysis. Total heavy metal contents were determined with atomic absorption spectroscopy (AAS) using flame (WEI 1992). Soil samples were analyzed for: value of STV, granular composition, content of mould, content of N_{org}, $pH_{(KCl)}$, content of nutrients (Mehlich III.

method) (Zbíral 1994). Plant materials for heavy metal analysis were harvested after maturation. The harvested plants were first dried, crushed and acids were added for mineralization by a Plazmatronica apparatus. Plant materials were analyzed for concentrations of Cd, Pb, Zn, Cu and Ni using AAS.

RESULTS

Heavy metal content of fluvisols

The heavy metal content in fluvisols from the Morava River is given in Table 1. As expected, the total of the expected metal contents of fluvisols including Cd, Cu, Ni, Pb and Zn were different at all three locations. The first group of samples taken from the Nový Svět location is high in total amount of Cd, Ni and Zn. Levels of metal from floods (the largest and most recent was in July 1997) coming from the Olomouc industrial area led to large concentrations of these metals in the soil. The concentrations were higher than the limits determined by public notice: for Cd about 23%, for Ni 38% and for Zn about 54% at the Nový Svět location. The concentrations of metals from the Nemilany soil broke the limits for Cd by about 11%, Cu 29%, Ni 50% and Zn 48%. Only soil from Nový Dvůr did not exceed any heavy metals limits.

Soil pH ranges were from moderately acidic to neutral, pH_{KCl} was from 6.5 to 6.9 (see Table 2) and these values should reduce the transport of heavy metals from soil to plants.

The selected physical and chemical properties of fluvisols mixed with sewage sludge are given in Table 3. There are two types of soil, sandy loam from Nový Svět and Nemilany and silt loam from the Nový Dvůr site. These differences influenced the splitting of soils into two groups for heavy metal concentration and their limits.

Location	**Cd**	**Cu**	**Ni**	**Pb**	**Zn**
Nový Svět	**0.6**	27.24	**24.31**	25.3	**108.5**
Nemilany	**0.45**	**42.24**	**30.12**	19.4	**95.57**
Limits	**0.4**	**30**	**15**	**50**	**50**
Nový Dvůr	0.58	22.14	22.88	19.9	87.13
Limits (different due to soil texture)	**1**	**50**	**25**	**70**	**100**

Table 1. Concentration of heavy metals (mg/kg) in Morava floodplain fluvisols (2 M HNO_3). Bold values are above limits (different limits for two soil types)

Location	**pH_{KCl}**	**% $CaCO_3$**
Nový Svět	6.5	0.2
Nemilany	6.7	0.3
Nový Dvůr	6.9	0.3

Table 2. pH ranges and carbonate content in the soil

Parameters	**unit**	**Nový Svět**	**Nemilany**	**Nový Dvůr**
<0,01mm	%	12.7	10.7	33
<0,001mm	%	1.6	2	9.2
0.01-0.05mm	%	26.3	25.5	35.8
0.05-0.25mm	%	48.8	43.3	28.2
0.25-2.0mm	%	12.2	20.5	3
Soil		Light loamy-sandy	light loamy-sandy	medium loamy
$N_{org.}$	$mg.kg^{-1}$	2664.87	3420.65	2377.08
C_{org}	%	2.71	2.36	2.59
Humus	%	4.68	4.07	4.47

Table 3. Chosen physical and chemical soil properties

Samples of soil for pot trials were taken from individual locations. Content of heavy metals – Cd and Pb on which we focused – in the samples for pot trials were within the limits according to the NOTICE OF THE CZECH MINISTRY OF ENVIRONMENT No. 13/1994 Sb. Sewage sludge from a municipal wastewater treatment plant was then added to these samples.

Heavy Metal Content of Sewage Sludge

Stabilized sewage sludge came from a municipal wastewater treatment plant in Olomouc. The sludge was used in three prescribed amounts – 2.5, 5 and 10 tonnes/ha converted to allow proper ratios for 8kg pots. The total amounts used were 10.37g, 20.75g and 41.5g per pot. These amounts corresponded to the limits published in notice Nr.382/2001. Table 4 shows the total concentration of heavy metals in the sludge, which was very low.

The neutral pH of sludge (pH_{KCl}=7.2) increases the soil's pH and the sludge is also used as a source of available nutrients which are given in Table 5.

Sludge dose	**Cd**	**Cu**	**Ni**	**Pb**	Zn
10.37g	0.01348	1.969	0.415	0.865	11.61
20.75g	0.02698	3.64	0.83	1.43	23.24
41.50g	0.05395	6.981	1.66	2.561	46.48
Limits	5	500	100	200	2500

Table 4. Concentration of heavy metals (mg/kg) in sewage sludge (2 M HNO_3)

	P	K	Ca	Mg	Na	pH_{KCl}
Sludge	20200	2900	45300	5110	730	7,2

Table 5. Nutrients (mg/kg) and pH in sewage sludge

The Plant Growth Experiment

Dry Weight Yields

The current soil conditions and nutrient contents of fluvisols led to yields shown in Figure 1.

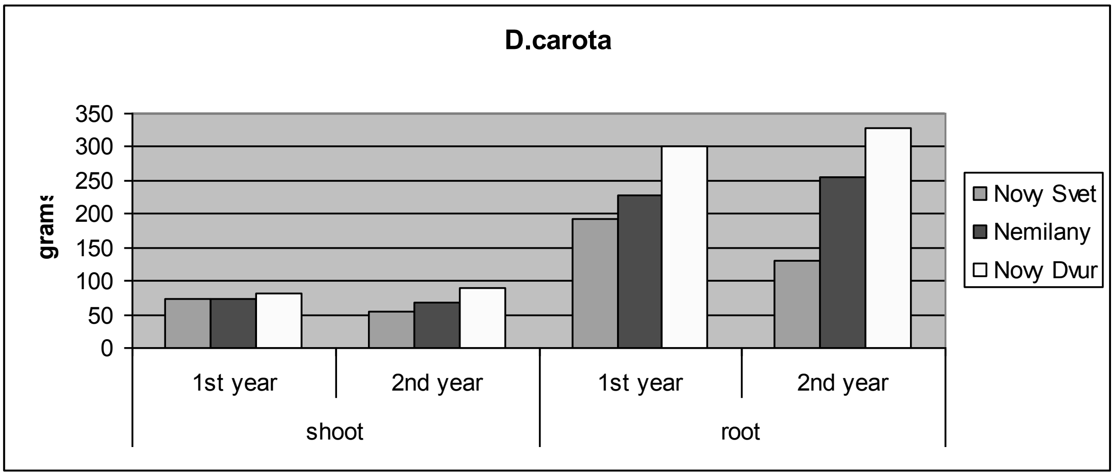


Fig. 1. The average yield (in grams of dry weight) of shoot and root in *D.carota* on fluvisols from three locations

In general the yield was higher in the first year, on average about 2%. This was caused by lower nutrient content in the pots in the second year, every year the same amount of nutrients was used by plants and the same amount of fertilizer was added but for both years only one sample of soil was used.

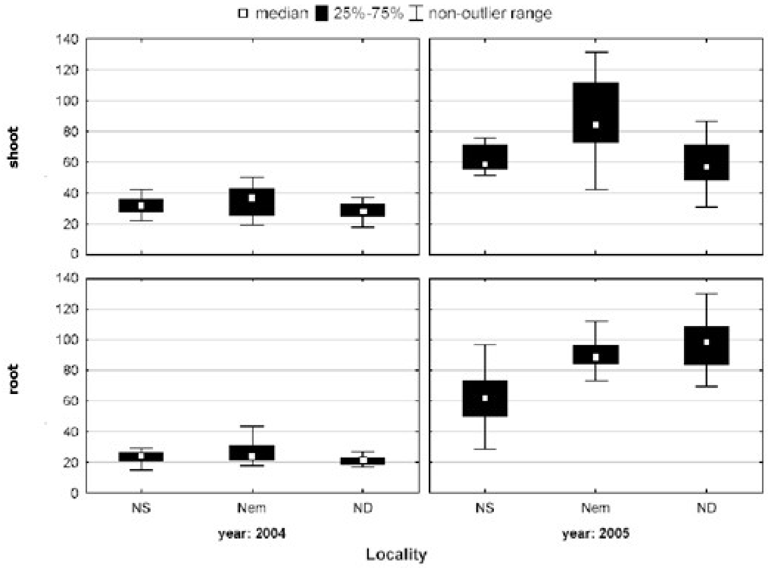
This difference was evident in control pots: for the roots of *D. carota* 161.29 g and 18.47 g in Nový Svět, 201.66 g and 183.6 g in Nemilany, 261.17g and 367 g in Nový Dvůr. Sewage sludge modification demonstrated a positive effect on plant growth at each sludge modification level. Most agricultural crops grow well when the soil pH is between 6.0 and 7.0, because nutrients are more available at a pH of about 6,5 (McConnel et al. 1993) and the yield of crops increases with increasing soil pH to an optimal pH value of between 6.5 – 7.0 (Smith 1993).

Heavy Metal Content in Plant Tissues

The heavy metal content was at different levels depending on the part of the plant, the heavy metals monitored and the experimental year (in the tables the average amount is used for both years). The transfer factor was introduced as an important factor (TF = rate of heavy metals in plant part and in ground).

Cadmium

The amounts of cadmium ranged from 0.13 to 0.28 mg/kg in the shoot and from 0.12 to 0.36 mg/kg in the roots of *D.carota*. The average transfer factor for *D. carota*: control (fluvisol) – 0.31 and for sludge (and fluvisol) – 0.34. Focusing on different locations, in Nový Svět the TF for soil with sludge was 0.32, in Nemilany 0.38 and in Nový Dvůr 0.33. Transfer factors and correlation between Cd in soil and in carrots root are in Figures 2 and 3.



Key for all figures - locations: NS – Nový Svět, Nem – Nemilany, ND – Nový Dvůr

Fig. 2. Cd transfer factors for both carrot root and shoot

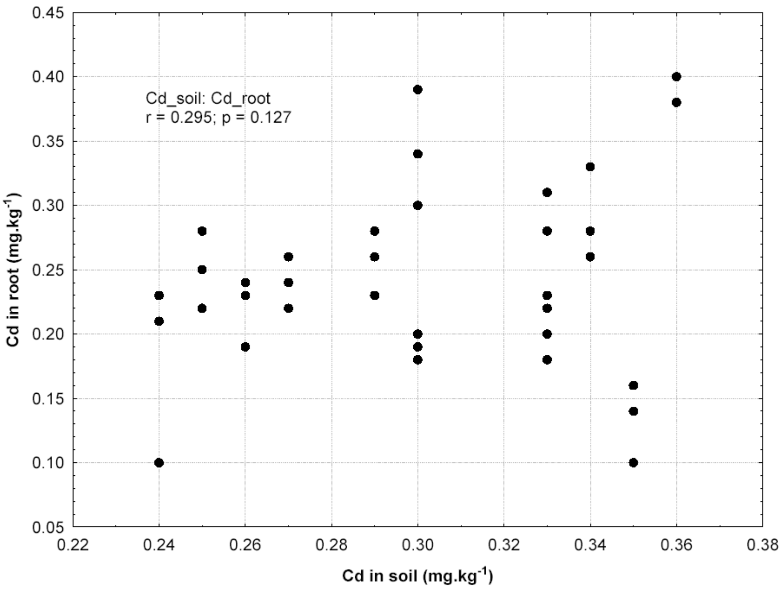


Fig. 3. Relation between Cd in soil and carrot root

Lead

The amount of lead fluctuated from 0.05 to 0.12 mg/kg in the shoot and from 0.05 to 0.12 mg/kg in the root of *D.carota*. The average transfer factor level for *D. carota*: control (fluvisol) – 0.003 and for sludge (and fluvisol) – 0.004 and for *H.vulgare*: control (fluvisol) – 0.003 and for sludge (and fluvisol) - 0.004. Focusing on different locations, in Nový Svět and Nemilany the TF for soil with sludge was 0.004 and in Nový Dvůr 0.005. Transfer factors and correlation between Pb in soil and in the carrot root are in Figures 4 and 5.

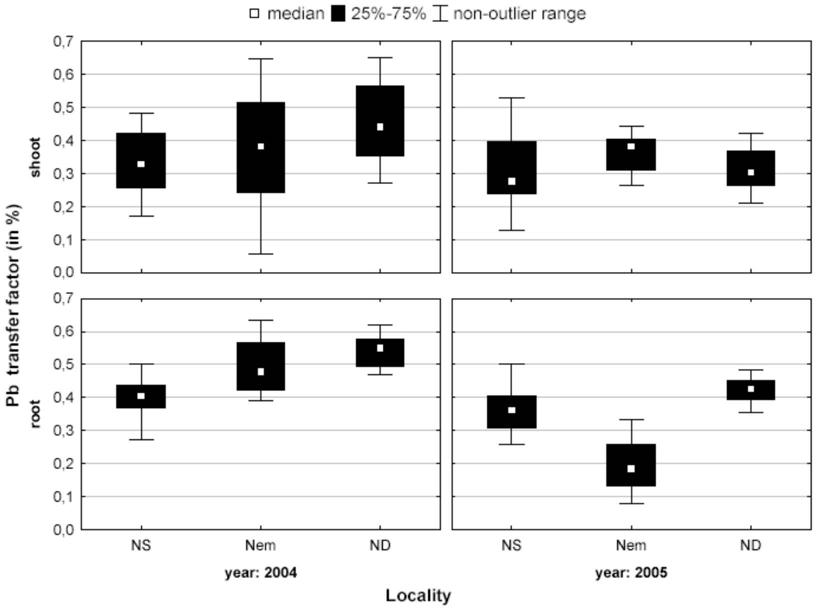


Fig. 4. Pb transfer factors for both carrot root and shoot

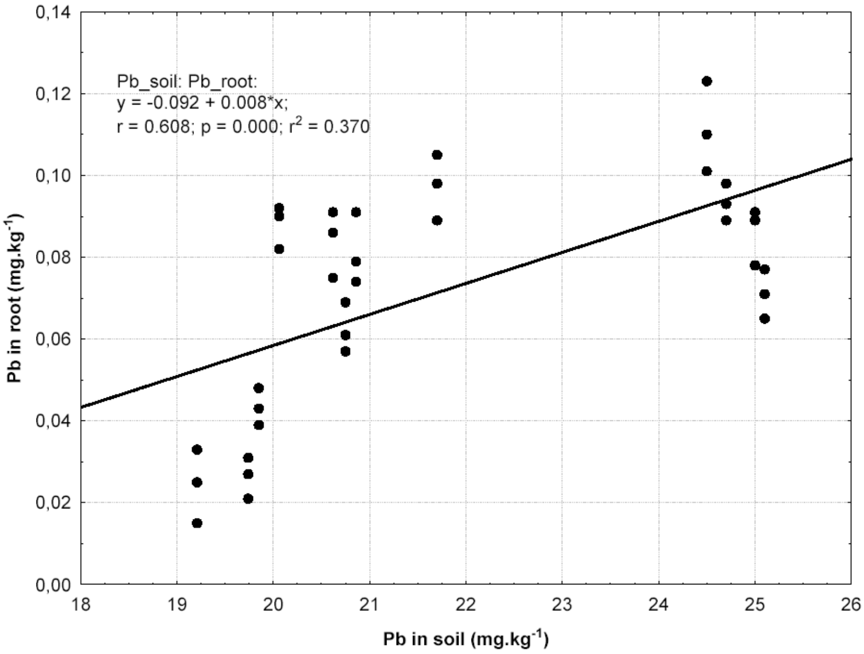


Fig. 5.Correlation between Pb in soil and carrot root

Copper

The amount of copper ranged from 0.84 to 1.13 mg/kg in the shoot and from 0.61 to 0.91 mg/kg in the root of *D.carota*. Average transfer factor level for: control (fluvisol) – 0.03 and for sludge (and fluvisol) – 0.03. Focusing on different locations, in Nový Svět the TF for soil with sludge was 0.03, in Nemilany 0.02 and in Nový Dvůr 0.04.

Zinc

The amount of zinc fluctuated from 25.28 to 42.53 mg/kg in the shoot and from 20.7 to 33.88 mg/kg in the root of *D.carota*. Average transfer factor level for: control (fluvisol) – 0.1 and for sludge (and fluvisol) – 0.12. Focusing on different locations, in Nový Svět the TF for soil with sludge was 0.12, in Nemilany and Nový Dvůr 0.13. Transfer factors and correlation between Pb in soil and in the carrot root are in Figures 6 and 7.

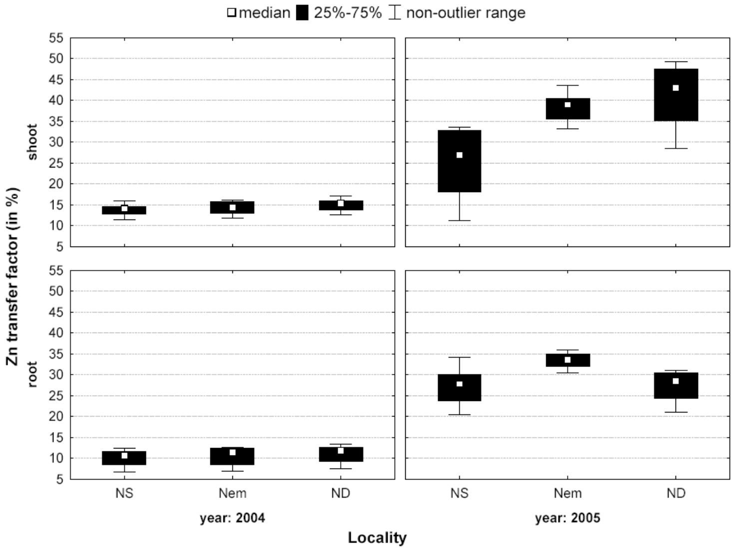


Fig. 6. Zn transfer factors for both carrot root and shoot

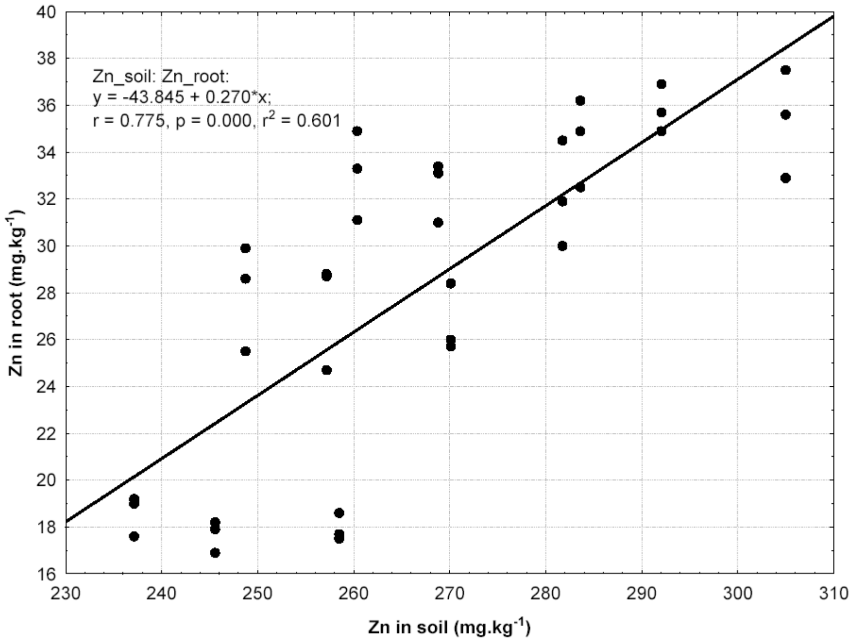


Fig. 7. Correlation between Zn in soil and carrot root

Nickel

The amount of nickel ranged from 1.23 to 2.24 mg/kg in the shoot and from 1.93 to 5.83 mg/kg in the root. Average transfer factor level for: control (fluvisol) – 0.09 and for sludge (and fluvisol) – 0.12. Focusing on different locations, in Nový Svět the TF for soil with sludge was for 0.12, in Nemilany 0.11 and in Nový Dvůr 0.14.

DISCUSSION AND CONCLUSION

The average values of the transfer factors (TF) for *D.carota* were: cadmium 0.33, lead 0.004, copper 0.03, zinc 0.12 and nickel 0.1.

In the final analysis we concentrated on the differences in TF in various parts of the plants – the shoot and root. The results are shown in Table 6.

Element	***D. carota***	
	Shoot	**Roots**
Cd	0.32	0.35
Pb	0.004	0.004
Cu	0.03	0.03
Zn	0.13	0.11
Ni	0.07	0.15

Table 6. Average values of transfer factors.

As expected, higher TF were primarily detected in the root of *D.carota,* except for zinc, equal values were found for lead and copper. Nickel values were two times higher. The lowest TF was evaluated for Pb which is consistent with data published by INTAWONGSE and DEAN (2006). The results showed that the uptake of Cd, Cu, Ni and Zn by plants (including carrot) correspond to the increasing level of soil contamination, while the uptake of Pb was low. Soil to plant transfer factor values decreased from Ni > Zn > Cd > Cu > Pb.

Transfer to plants depends on the bioavailability of the compound in soil. This depends on several factors including pH, soil temperature, and organic matter content or soil texture. The pH is of particular importance (EUROPEAN ENVIRONMENT AGENCY 2006). Table 7 presents heavy metal mobility according to the pH factor. A value is attributed to each metal between 1 (the least available) and 5 (the most available) to estimate its mobility (ADEME 2001).

	alkaline, neutral	**acid**
Cd	2	4
Pb	1	3
Cu	1	3
Zn	2	4
Ni	1	4

Table 7. Mobility of metals in soil

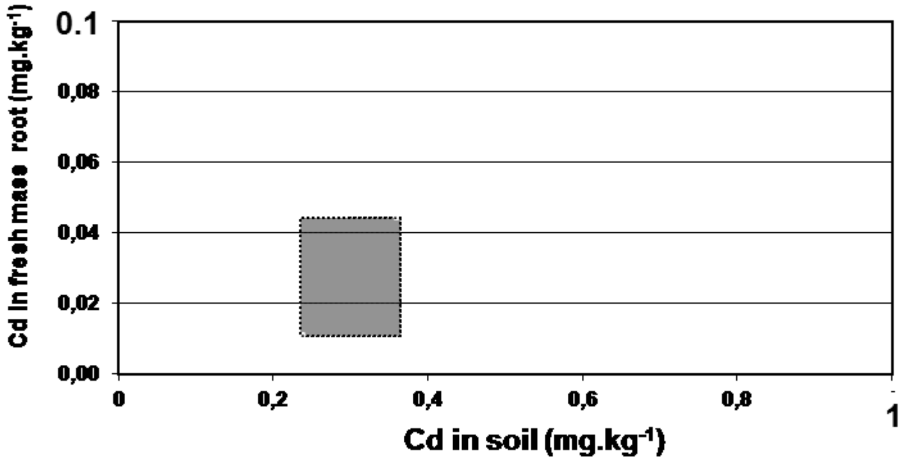
When compared with values found by DIEZ (1995), our results of transfer factors were generally lower. One possible explanation could be the higher pH level of the sewage sludge which increased the soil reaction. It could limit the transfer of heavy metals into the plants (BADALIKOVA et al. 2005). Soil pH ranges were from moderately acidic to neutral, pH_{KCl} was from 6.5 to 6.9 and these values should reduce the transport of heavy metals from soil to plants (HERMS 1989; COTTENIE, KIEKENS 1981; SAUERBECK 1989).

From the research of BALÍK et al. (1999) it is evident that liming resulted in significantly lower Cd mobility after application of limed sludge than in sludge alone. Results of GRAY et al. (1999) show that in general, increasing soil pH from 5.5 to 7.0 significantly reduced Cd concentration in carrots and other crops, although the magnitude of the reduction varied between plant species and soil types. Also analysis made by HOODA et al. (1997) shows that liming soils to pH 7 effectively reduces the metal content in carrots.

Sewage sludge from municipal wastewater treatment plants contains high quantities of nutrients and organic matter needed for plant growth, but it also contains a high concentration of heavy metals. Heavy metals can accumulate in soil and in plants when sludge is applied as a fertilizer and eventually can produce harmful effects in animals and humans. The biological role of heavy metals in plants depends on their concentration in the plant tissues. Different stages corresponding to deficiency, normal or toxic levels may be observed according to the metal level in the plant (EUROPEAN ENVIRONMENT AGENCY 2006). Because of possible harmful effects of heavy metals, the study of effective methods for heavy metal removal from sludge is very important in order to minimize prospective health risks during application (LASHEEN et al. 2000).

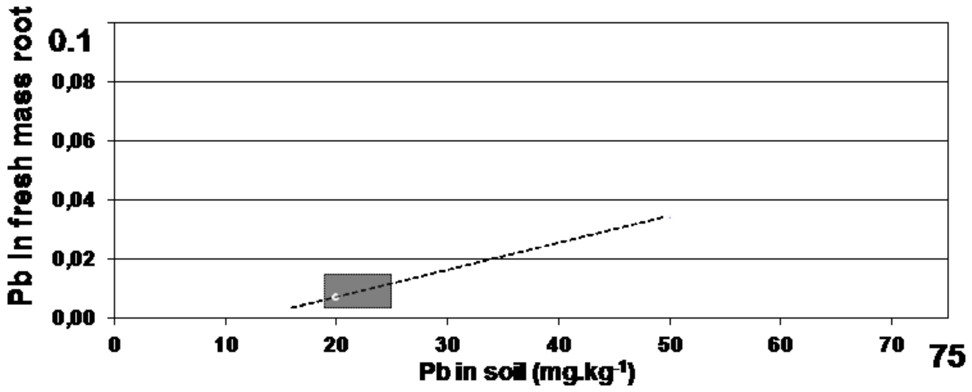
The concentration of heavy metals during our experiment in both sludge and fluvisol from the Morava river floodplain was not extremely high and this is why no differences were found in comparing the control plants and the total value of TF for all locations together.

Transfer factors closely relate to the quality of the resulting product – carrots in our case. According to COMMISSION REGULATION No. 1881/2006, the limit in biomass is only defined for Cd and Pb – to an amount of 0.1mg/kg of fresh mass. After transferring our results to moist mass we obtain results described in figures 8 and 9. The diagram shows the range of results with the limits for this element in soil and plant biomass given in bold. These show that if we monitor the limits of heavy metals in soil within the NOTICE OF THE CZECH MINISTRY OF THE ENVIRONMENT No. 13/1994 Sb., the limit of heavy metal content in the product is not exceeded. If, on the basis of our results, we calculate the maximum acceptable content of Cd in in the soil (1mg Cd /kg), then theoretical content of this element in the carrot root could be up to 0.06 mg/kg and for Pb content of 75 mg/kg in the soil the root content could also be up to 0.06 mg/kg and would not exceed the maximum value stated in the COMMISSION REGULATION.



Note: the rectangle shows the range of measured values in the pot trials, bold values 1 and 0.1 mg.kg^{-1} show limit of Cd in soil and plant biomass.

Fig. 8. Obtained correlation between content of Cd in soil and in carrot root



Note: the rectangle at the beginning of the line shows the range of measured values in the pot trials, bold values 75 and 0,1 mg.kg^{-1} show limit of Pb in soil and plant biomass.

Fig. 9. Obtained correlation between content of Pb in soil and in carrot root

The results of fluvisol studies show above-limit concentrations of some, potentially hazardous elements in sandy soils. The results of pollution in fluvisols are in accordance with the aforementioned studies. The results indicate the necessity to monitor hazardous elements in the soils of alluvial areas, presuming that unless the limit for normal soils is exceeded, the monitored produce should not be polluted with content of these elements in excess of limits.

Acknowledgement

The authors thank the Czech Ministry of Education, Youth and Sports for supporting their research through grant No. 2B06101 from the National Research Program II.

References

ADEME A., RECYVAL P., 2001. Compost de boues de stations d´épuration municipales: qualité performance agronomiques et utilisations. http://ec.europa.eu/environment/waste/sludge/sludge_disposal3a.pdf

BADALÍKOVÁ B., HRUBÝ J., HARTMAN I., 2005. Effect of humus content in soil on the uptake of heavy metals by plants. In: CD from ISTRO Conference "Soil – Agriculture, Environment, Landscape", Brno, Czech Republic: 135-141.

BALÍK J., TLUSTOŠ P., SVÁKOVÁ J., BLAHNÍK R., KAEWRAHUN S., 1999. Sorption of cadmium in soil treated by limed sewage sludges. Rostlinná výroba, *45/11*: 511-518.

COMMISSION REGULATION setting maximum levels for certain contaminants in food stuff, No. 1881/2006, European Commission.

COTTENIE A., KIEKENS L., 1981. Beweglichkeit von Schwermetallen in mit Schlamm angereicherten Böden. Korrespondenz Abwasser, *28*: 206 - 211.

DIEZ T., KRAUSS M., 1995. Odčerpávání těžkých kovů z kontaminovaných ploch pomocí rostlin. In: sborník Těžké kovy v zemědělské pudě a rostlinách, Praha: 41-43 (in Czech).

EBBS S., TALBOTT J., SANKARAN R., 2006. Cultivation of garden vegetables in Peoria Pool sediments from the Illinois River: A case study in trace element accumulation and dietary exposures. Environment International, *32*: 766-744.

EUROPEAN ENVIRONMENT AGENCY, 2006. Disposal and recycling routes for sewage sludge - http://ec.europa.eu/environment/waste/sludge/ sludge_disposal.htm

GRAY C.W., MCLAREN R.G., ROBERTS A.H.C., CONDRON L.M., 1999. Effect of soil pH on cadmium phytoavailability in some New Zealand soils. New Zealand Journal of Crop and Horticultural Science, *27*: 169-179.

HERMS U., 1989. Löslichkeit von Schwermetallen in Böden unter variierenden Milieubedingungen: In: Dechema (Hrsg. von D. Behrens u J. Wiesner, 1989). Beurleilung von SK-Kontamination im Boden. Frankfurt/Main: 189-199.

KRÜGER F., MEISSNER R., GRÖNGRÖFT A., GRUNEWALD K., 2005. Flood induced heavy metal and arsenic contamination of Elbe river floodplain soils. Acta Hydrochim. Hydrobiol., *33 (5*): 455-465.

LASHEEN M. R., ASHMAWAY A., IBRAHIM, H., 2000. Heavy metal removal from municipal and industrial sludges. In: 11th Annual International Conference on Heavy metals in the Environment (J. Nriagu, Editor), Contribution 1431. University of Michigan, School of Public Health, Ann Arbor, MI.

McGRATH S.P., ZHAO F.J., DUNHAM S.J., CROSLAND A.R., COLEMAN K., 2000. Long-term changes in the extractability and bioavailability of zinc and cadmium after sludge application. Journal of Environmental Quality, *29*: 875-883.

MIDDELKOOP H., 2000. Heavy metal pollution in the river Rhine and Meuse floodplains in the Netherlands. Geologie en Mijnbouw – Netherlands Journal of Geosciences, *79*: 411-427.

NOTICE OF THE CZECH MINISTRY OF ENVIRONMENT for the management of soil protection, No. 13/1994 Sb. (in Czech).

PEREZ D.V., ALCANTARA S., RIBEIRO C.C., PEREIRA R.E., FONTES G.C., WASSERMAN M.A., VENEZUELA T.C., MENEGUELLI N.A., DE NACEDÍ J.R., BARRADAS C.A.A., 2007. Composted municipal waste effects on chemical properties of a Brazilian soil. Bioresource Technology, *98*: 525-533.

PETROVICH-GEGIC A.D., VOJINOVICH-MILORADOV M.B., CEHMAJSTER K.J.S.,ILES, F.I., 2007. Adsorption of essential and toxic elements in various fruit and vegetable sorts in the flooded area of the Tisa river. Hemijska Industrija, *61, 5A*: 321-325.

ŠARAPATKA B., ŠTĚRBA O., 1998. Optimization of agriculture in relation to the multifunctional role of the landscape. Landscape and Urban Planning, 41 (2): 145-148.

SAUERBECK D., 1989. Der Transfer von Schwermetallen in die Pflanze. In: Beurteilungen von Schwermetallkontaminationene im Boden, Dechema. Frankfurt (Main): 281-316.

SCHWARTZ R., GERTH J., NEUMANN-HENSEL H., BLEY S., FÖRSTNER U., 2006. Assessment of highly polluted fluvisols in the Spittelwasser floodplain. J. Soils Sediments, *6 (3):* 145-155.

SIERRA J., MONTSERRAT G., MARTI E., GARAU E.A., CRUANAS R., 2000. Contamination levels in the soils affected by the food from Aznalcollar. Soils and Sediment Contamination, *9*: 311-329.

SMITH S.R., 1993. Environ. Pollut., *85*, 387.

VANA J., 2003. Obsahy těžkých kovů v pudě a v rostlinách po dlouhodobém hnojení průmyslovými komposty. VÚRV Praha-Ruzyně (in Czech).

WEI F.S., 1992. Modern Analytical Methods of Soil Elements. Chinese Environ. Sci. Press, Beijing: 64.

YAMAGATA N., SHIGEMATSU I., 1970. Cadmium pollution in perspective. Bulletin of the Institute of Public Health, *19*: 1-27.

ZBÍRAL J., 1994. Analýza rostlinného materiálu, ÚKZÚZ OAPUR. (in Czech).

The Effect of Different Pollution Levels in Fluvisols on the Accumulation of Heavy Metals in Carrots (*Daucus carota L.)* and Spring Barley (*Hordeum vulgare L.)*

Jan Kočař,[1] Bořivoj Šarapatka[1] and Michaela Smatanová[2]

[1] Department of Ecology and Environmental Sciences, Palacký University Olomouc, tř.
Svobody 26, 771 46 Olomouc, Czech Republic
[2] Central Institute for Supervising and Testing in Agriculture, Hroznová 2,
656 06 Brno, Czech Republic

SUMMARY

This article deals with the effect of different pollution levels of soils with an accumulation of heavy metals by two plants. Fluvisols from the Morava floodplain area were used. The soil samples were taken from three different localities (yearly, irregularly and rarely flooded). Homogenized sludge from a municipal wastewater treatment plant was artificially introduced into the samples.

Two types of accumulative plants were used: the Rubina variety of *Daucus carota* and the Amulet variety of *Hordeum vulgare*. In the following analysis of changes in soil characteristics were observed, i.e., mainly the heavy metal concentrations of cadmium, lead, zinc, copper and nickel in soils before the experiment, after finishing each cycle and in the plants themselves.

Analysis showed exceedingly high levels of heavy metals in fluvisols with almost all elements, with the exception of lead. Zinc and nickel had the highest concentrations. The yield of both plants was influenced by the amount of applied sludge, with the same effect as a fertilizer. On the other hand the high sludge pH level partly limited introduction of most heavy metals into the plants.

Total increases of heavy metals in the plants were monitored and evaluated by using transfer factors. They were compared at all locations including the differences between the controls and soil with the sludge, and total transfer factors focused on various elements. The research showed almost the same accumulation abilities of heavy metals except cadmium by both plants. As expected there was a higher accumulation potential in the root of *D. carota* and in the straw in *H. vulgare*.

KEY WORDS

accumulation, fluvisols, heavy metals, carrot, barley

INTRODUCTION

Heavy metal pollution of soils is increasingly becoming a global problem with the development of industry, mining activity, waste water irrigation and the application of sewage sludge, even if it is relatively localized at present. The soil-plant system is the fundamental building block of the geosphere and biosphere. Therefore, heavy metal pollution of the soil has an important influence not only on the yield and quality of crops, but also on the quality of the atmospheric and the aquatic environment, and even on the health of human beings via the food chains [1, 2].

An Important source of heavy metal soil contamination can become industrial compost, which is primarily produced from sewage sludge. At present we do not have enough knowledge of the negative effects of repeated compost application (with different levels of heavy metals) on soil and plants [3].

Traditional solutions such as disposal of contaminated soil in landfills account for a large proportion of the remediation operations at the present. However, some of the remediation techniques currently in use will probably lose economic favour and public acceptance in the near future [4].

Heavy metals can be translocated to plant parts aboveground. The metal-rich plant material may be safely harvested and removed from the site without extensive excavation, disposal costs, and loss of topsoil associated with traditional remediation practices [5].

In recent years this phytoaccumulation means the use of plants to cleanup soils contaminated with non-volatile hydrocarbons and immobile inorganics and it is showing promises as a new method for *in situ* cleanup of large volumes of low to moderately contaminated soils. Plants can be used to remove, transfer, stabilize and/or degrade heavy metal soil contaminants [6, 7, 8, 9, 10, 11].

Contaminated sites often support characteristic plant species, some of which are able to accumulate high concentrations of heavy metals in their tissue [12, 13, 14, 15].

Most plants that survive in polluted soils do so by either, avoiding heavy metals, or, hyperaccumulating them in their tissues. Such plants are uncommon [16] and, to date, approximately 400 hyperaccumulator species have been identified, according to an analysis of field-collected speciments [17].

Besides the limited distribution of hyperaccumulators in the wild, such plants also tend to be contaminant specific. No plant species has yet been found that will demonstrate a wide spectrum of hyperaccumulation [18].

The success of the phytoremediation process is dependent on adequate plant yield and high metal concentrations in plant shoots. Plants must produce sufficient biomass while accumulating high concentrations of heavy metals. Hyperaccumulator plants possess an ability to take up abnormally high amounts of heavy metals in their shoots [19, 20]. However, most hyperaccumulators species are not suitable for phytoremediation application in the field due to their small biomass and slow growth. As an alternative, it has been suggested to use high biomass species, such as maize (*Zea mays L.*), pea (*Pisum sativum L.*), oat (*Avena sativa L.*), canola (*Brassica napus L.*), barley (*Hordeum vulgare L.*) and Indian mustard [*Brassica juncea (L.) Czern.*], with improved plant husbandry and soil management practices to enhance metal uptake by these high biomass species [21, 22, 5, 23, 24, 25].

The aim of this research is to find out the possibilities of accumulation of heavy metals from fluvisols by two plants – *H.vulgare* and *D.carota*.

MATERIALS AND METHODS

1) Pot Experiment

In our experiment we used two different accumulative plants: *Daucus carota* L. Rubina and *Hordeum vulgare* L. Amulet in soils mixed with various amounts of sewage sludge from an Olomouc wastewater treatment plant. The soil was first fertilized with super phosphate, potassium chloride and urea. Then the soils were mixed separately with sewage sludge at application rates of 0; 2.5; 5 and 10 tonnes ha^{-1} (according to public notice Nr.382/2001) that are equivalent to 0; 10.37; 20.75 and 41.5 tonnes ha^{-1} (dry weight basis).

During the two-year experiment, the investigations were carried out on samples from the fluvisols of the Morava River floodplain area. The samples were collected from three different localities (at depths of 0-20 cm) contaminated with heavy metals from a flood in July 1997.

The soil was air-dried, crushed, mixed thoroughly and passed through a 1 cm sieve. 8 kg portions of the soil were transferred to plastic pots and treated with the necessary amounts of fertilizer at a rate of 0.6 g super phosphate, 0.8 g potassium chloride and 0.66 g urea for *H.vulgare* and 0.6 g super phosphate, 0.6 g potassium chloride and 0.53 g urea for *D.carota*, and then mixed well.

A stabilized sludge sample was obtained from a municipal wastewater treatment plant in Olomouc. Sewage sludge was added at a rate shown in Table 1. The three amount types, converted to the proper proportions, were used per 8 kg pot according to public notice Nr.382/2001 (2.5 tonnes ha^{-1}, 5 tonnes ha^{-1} and 10 tonnes ha^{-1}). All combinations were replicated three times. Deionized water was used throughout.

TABLE 1
THREE AMOUNT TYPES OF SEWAGE SLUDGE AT THREE DIFFERENT LOCALITIES USED IN THE POT EXPERIMENT.

Localities	**Fertilization (g/pot)**
A. Novy Svet	1. control
	2. sludge 10.37
	3. sludge 20.75
	4. sludge 41.50
B. Nemilany	5. control
	6. sludge 10.37
	7. sludge 20.75
	8. sludge 41.50
C. Novy Dvur	9. control
	10. sludge 10.37
	11. sludge 20.75
	12. sludge 41.50

2) Analytical Methods

Soil samples were taken from the pots. The samples were air-dried and ground prior to analysis. Total heavy metal contents were determined with atomic absorption spectroscopy (AAS) using flame [26]. Soil samples were analyzed for: value of STV, granular composition, content of mould, content of N_t, $pH_{(KCl)}$, content of nutrients (Mehlich III. method) [27]. Plant materials for heavy metal analysis were harvested after maturation. The harvested plants were first dried, crushed and acids were added for mineralization by a Plazmatronica apparatus. Plant materials were analyzed for concentrations of Cd, Pb, Zn, Cu and Ni using AAS.

RESULTS

1) Heavy metal content of fluvisols

The heavy metal content in fluvisols from the Morava River is given in Table 2. As expected, the total of the expected metal contents of fluvisols including Cd, Cu, Ni, Pb and Zn were different at all three locations. The first group of soil taken from the Novy Svet location is exceptionally high in total amount of Cd, Ni and Zn. High levels of metal from floods (the largest and most recent was in July 1997) coming from the Olomouc industrial area led to large concentrations of these metals in the soil. The concentrations were higher than the limits determined by public notice: for Cd about 23%, for Ni 38% and for Zn about 54% at the Novy Svet location. The concentrations of metals from the Nemilany soil broke the limits for Cd by about 11%, Cu 29%, Ni 50% and Zn 48%. Only soil from Novy Dvur did not exceed any heavy metals limits.

Soil pH ranges were from acidic to almost neutral, pH_{KCl} was from 6.7 to 6.9 (see Table 3). These values should decrease the transport of heavy metals from soil to plants [28, 29, 30].

The selected physical and chemical properties of fluvisols mixed with sewage sludge are given in Table 4. There are two types of soils, sandy loam from Novy Svet and Nemilany and silt loam from the Novy Dvur site. These differences influenced the splitting of soils to two groups for heavy metal concentration and their limits.

TABLE 2
CONCENTRATION OF HEAVY METALS (MG KG^{-1}) IN MORAVA RIVER FLUVISOLS (2M HNO_3). BOLD VALUES ARE ABOVE LIMITS (DIFFERENT LIMITS FOR TWO SOIL TYPES)

Locality	Cd	Cu	Ni	Pb	Zn
Novy Svet	**0.6**	27.24	**24.31**	25.3	**108.5**
Nemilany	**0.45**	**42.24**	**30.12**	19.4	**95.57**
Limits	**0.4**	**30**	**15**	**50**	**50**
Novy Dvur	0.58	22.14	22.88	19.9	87.13
Limits (different due to soil texture)	**1**	**50**	**25**	**70**	**100**

TABLE 3
pH RANGES AND LIME IN THE SOIL

Locality	pH_{KCl}	% $CaCO_3$
Novy Svet	6.7	0.2
Nemilany	6.8	0.3
Novy Dvur	6.9	0.3

TABLE 4
PHYSICAL AND CHEMICAL SOIL PROPERTIES

parameters	unit	Novy Svet	Nemilany	Novy Dvur
<0,01mm	%	12.7	10.7	33
<0,001mm	%	1.6	2	9.2
0.01-0.05mm	%	26.3	25.5	35.8
0.05-0.25mm	%	48.8	43.3	28.2
0.25-2.0mm	%	12.2	20.5	3
Soil		light loamy-sandy	light loamy-sandy	medium loamy
N_{tot}	mg.kg^{-1}	2664.87	3420.65	2377.08
C_{org}	%	2.71	2.36	2.59
Humus	%	4.68	4.07	4.47

2) Heavy Metal Content of Sewage Sludge

Stabilized sewage sludge came from municipal wastewater treatment plant in Olomouc. The sludge was used in three prescribed amounts – 2.5, 5 and 10 tonnes ha^{-1} converted to allow proper ratios per 8kg pots. The total amounts used were 10.37g, 20.75g and 41.5g per pot. These amounts corresponded to the limits published in notice Nr.382/2001. Table 5 shows the total concentration of heavy metals in the sludge, which is very low, at times one hundred times lower than the limited value that relates to its stabilization.

The pH of sludge increases the soil's pH to alkaline (pH_{KCl}=7.2). In this project the sludge is used mainly as a fertilizer. The amount of available nutrients is given in Table 6.

TABLE 5
CONCENTRATION OF HEAVY METALS (MG KG^{-1}) IN SEWAGE SLUDGE (2M HNO_3)

Sludge dose	Cd	Cu	Ni	Pb	Zn
10.37g	0.01348	1.969	0.415	0.865	11.61
20.75g	0.02698	3.64	0.83	1.43	23.24
41.50g	0.05395	6.981	1.66	2.561	46.48
Limits	**5**	**500**	**100**	**200**	**2500**

TABLE 6
NUTRIENTS (MG KG^{-1}) AND pH IN SEWAGE SLUDGE

	P	K	Ca	Mg	Na	pH_{KCl}
Sludge	20200	2900	45300	5110	730	7,2

3) The Plant Growth Experiment

Dry Weight Yields

The current soil conditions and nutrient contents of fluvisols led to yields shown in Figure 1 and Figure 2.

FIGURE 1
THE AVERAGE YIELD (IN GRAMS OF DRY WEIGHT) OF SHOOT AND ROOT IN *D.CAROTA* ON FLUVISOLS FROM THREE LOCATIONS

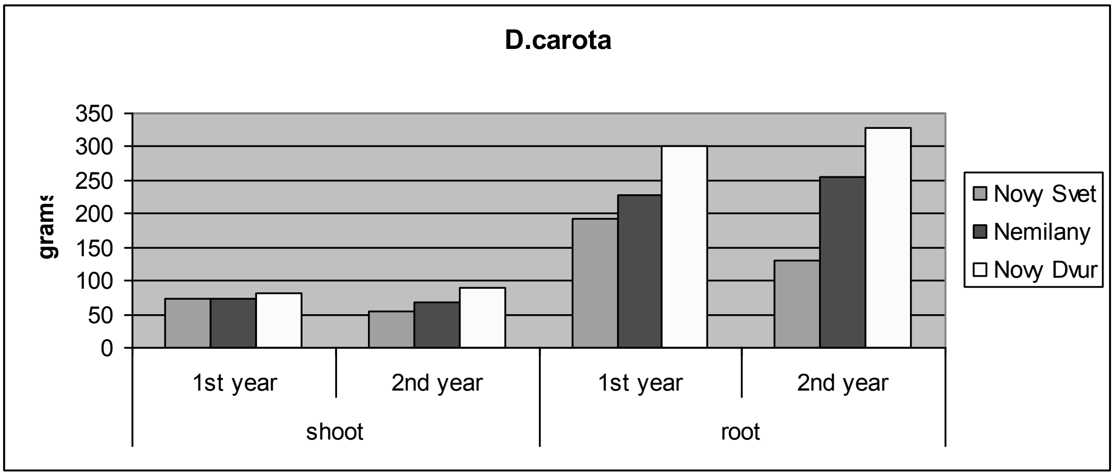
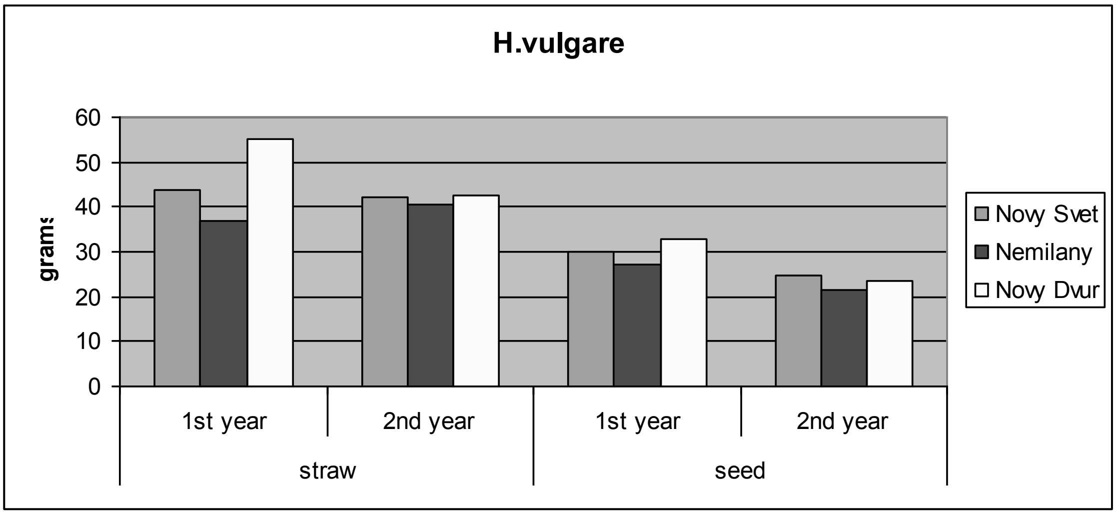


FIGURE 2
THE AVERAGE YIELD (IN GRAMS OF DRY WEIGHT) OF STRAW AND SEED OF *H.VULGARE* ON FLUVISOLS FROM THREE LOCALITIES



In general the yield was higher in the first year, on average about 13% for barley and about 2% for carrots. This was caused by lower nutrient contents in the pots in the second year, every year the same amount of nutrients were used by plants and the same amounts of fertilizers were added but for both years used only one sample of soil.

This difference was evident in control pots: for the shoot of *D. carota* 61.54 g and 12.47 g in Novy Svet (L1), 58.13 g and 60 g in Nemilany (L2), 62.4 g and 78.6 g in Novy Dvur (L3); for the root of *D. carota* 161.29 g and 18.47 g in L1, 201.66 g and 183.6 g in L2, 261.17g and 367 g in L3; for the seed of *H. vulgare* the result was 23.97g and 20.97g in L1, 23.84 g and 21.45 g in L2, 33.7 g and 24.47 g in L3; for the straw of *H. vulgare:* 26.87 g and 25.59 g in L1, 34.66 g and 40.55 g in L2, 42.77 g and 47.43 g in L3. Large differences were only observed in the yield of *D. carota* in the control pots of shoot and in pots of root in the Novy Svet locality.

Sewage sludge modification demonstrated a positive effect on plant growth at each sludge modification level. Most agricultural crops grow well when the soil pH is between 6.0 and 7.0, because nutrients are more available at a pH of about 6,5 [31] and the yield of crops increases with increasing soil pH to an optimal pH value of between 6.5 – 7.0 [32].

Heavy Metal Contents in Plant Tissues

The heavy metal content was at different levels depending on the part of plant, the heavy metals monitored and the experimental year (in the tables the average amount is used for both years). The transfer factor was introduced as an important factor (TF = rate of heavy metals in plant part and in ground). Average transfer factors in different parts of plants at the different locations are presented in Figure 3, in Figure 4 and Figure 5.

Cadmium

The amounts of cadmium ranged from 0.13 to 0.28 mg kg^{-1} in the shoot and from 0.12 to 0.36 mg kg^{-1} in the roots of *D.carota*, from 0.02 to 0.06 mg kg^{-1} in the straw and 0.01 – 0.06 mg kg^{-1} in the seed of *H.vulgare*. Average transfer factor level for *D. carota*: control (fluvisol) – 0.31 and for sludge (and fluvisol) – 0.34 and for *H.vulgare*: control (fluvisol) – 0.03 and for sludge (and fluvisol) – 0.06. Focusing on different localities, in Novy Svet the TF for soil with sludge was 0.32 for *D.carota* and 0.06 for *H.vulgare*, in Nemilany 0.38 and 0.07, in Novy Dvur 0.33 and 0.06.

Lead

The amounts of lead fluctuated from 0.05 to 0.12 mg kg^{-1} in the shoot and from 0.05 to 0.12 mg kg^{-1} in the roots of *D.carota*, from 0.06 to 0.12 mg kg^{-1} in the straw and 0.04 – 0.11 mg kg^{-1} in the seed of *H.vulgare*. The average transfer factor level for *D. carota*: control (fluvisol) – 0.003 and for sludge (and fluvisol) – 0.004 and for *H.vulgare*: control (fluvisol) – 0.003 and for sludge (and fluvisol)- 0.004. Focusing on different localities, in Novy Svet the TF for soil with sludge was 0.004 for *D.carota* and 0.003 for *H.vulgare*, in Nemilany 0.004 and 0.004, in Novy Dvur 0.005 and 0.004.

Copper

The amounts of copper ranged from 0.84 to 1.13 mg kg^{-1} in the shoot and from 0.61 to 0.91 mg kg^{-1} in the roots of *D.carota*, from 0.52 to 0.84 mg kg^{-1} in the straw and 0.5 – 0.78 mg kg^{-1} in the seed of *H.vulgare*. Average transfer factor level for *D. carota*: control (fluvisol) – 0.03 and for sludge (and fluvisol) – 0.03 and for *H.vulgare*: control (fluvisol) – 0.02 and for sludge (and fluvisol) – 0.02. Focusing on different localities, in Novy Svet the TF for soil with sludge for was 0.03 *D.carota* and for 0.02 *H.vulgare*, in Nemilany 0.02 and 0.01, in Novy Dvur 0.04 and 0.03.

Zinc

The amounts of zinc fluctuated from 25.28 to 42.53 mg kg^{-1} in the shoot and from 20.7 to 33.88 mg kg^{-1} in the roots of *D.carota*, from 37.98 to 54.15 mg kg^{-1} in the straw and 34.05 – 48.42 mg kg^{-1} in the seed of *H.vulgare*. Average transfer factor level for *D. carota*: control (fluvisol) – 0.1 and for sludge (and fluvisol) – 0.12 and for *H.vulgare*: control (fluvisol) – 0.15 and for sludge (and fluvisol) – 0.16. Focusing on different localities, in Novy Svet the TF for soil with sludge was for 0.12 *D.carota* and 0.16 for *H.vulgare*, in Nemilany 0.13 and 0.17, in Novy Dvur 0.13 and 0.18.

Nickel

The amounts of nickel ranged from 1.23 to 2.24 mg kg^{-1} in the shoot and from 1.93 to 5.83 mg kg^{-1} in the roots of *D.carota*, from 2.08 to 6.08 mg kg^{-1} in the straw and 0.47 – 3.4 mg kg^{-1} in the seed of *H.vulgare*. Average transfer factor level for *D. carota*: control (fluvisol) – 0.09 and for sludge (and fluvisol) – 0.12 and for *H.vulgare*: control (fluvisol) – 0.06 and for sludge (and fluvisol) – 0.11. Focusing on different localities, in Novy Svet the TF for soil with sludge was for 0.12 *D.carota* and 0.13 for *H.vulgare*, in Nemilany 0.11 and 0.11, in Novy Dvur 0.14 and 0.14.

FIGURE 3
AVERAGE TRANSFER FACTORS IN DIFFERENT PARTS OF PLANTS

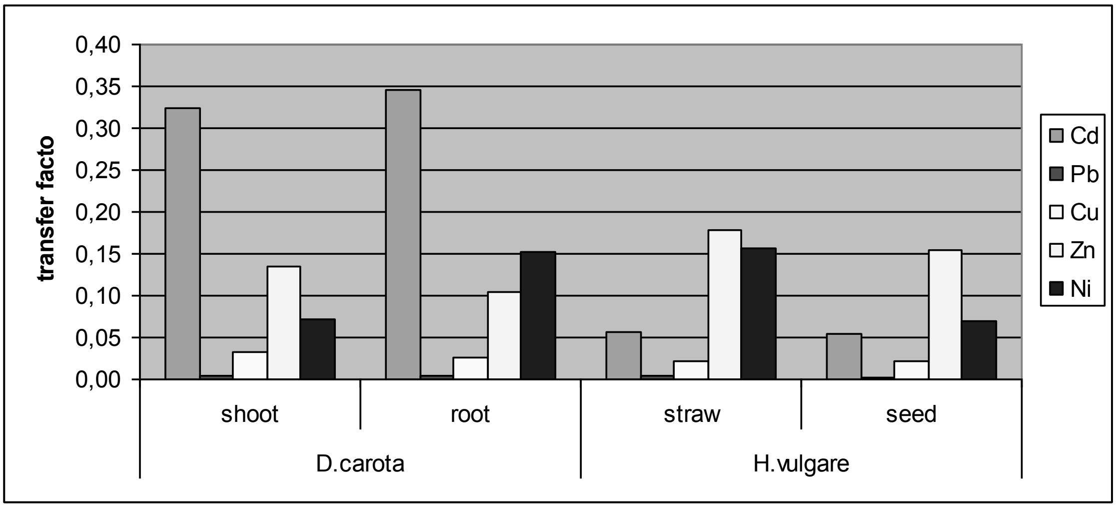


FIGURE 4
AVERAGE TRANSFER FACTORS FOR *D.CAROTA* AT DIFFERENT LOCALITIES

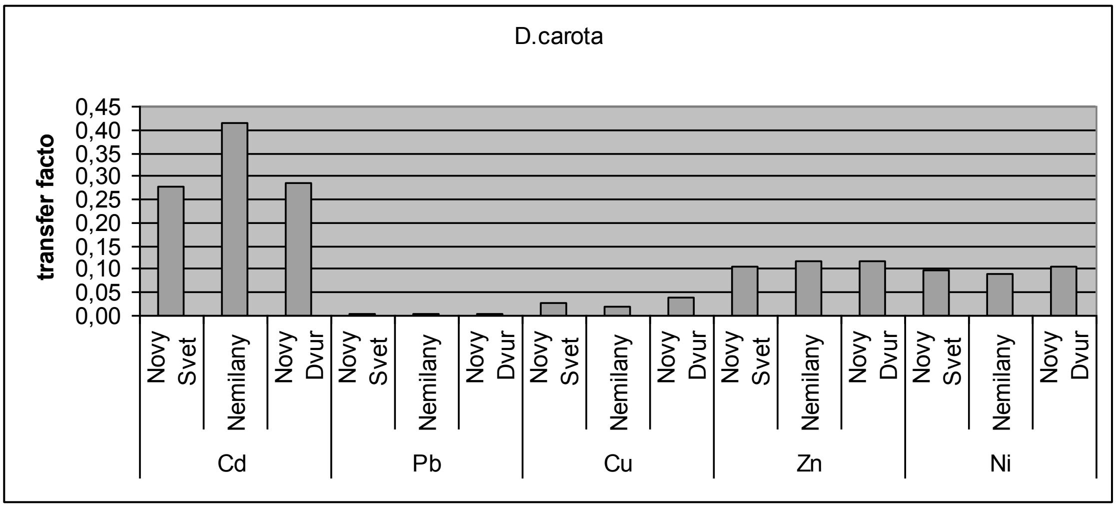


FIGURE 5
AVERAGE TRANSFER FACTORS FOR *H.VULGARE* AT DIFFERENT LOCALITIES

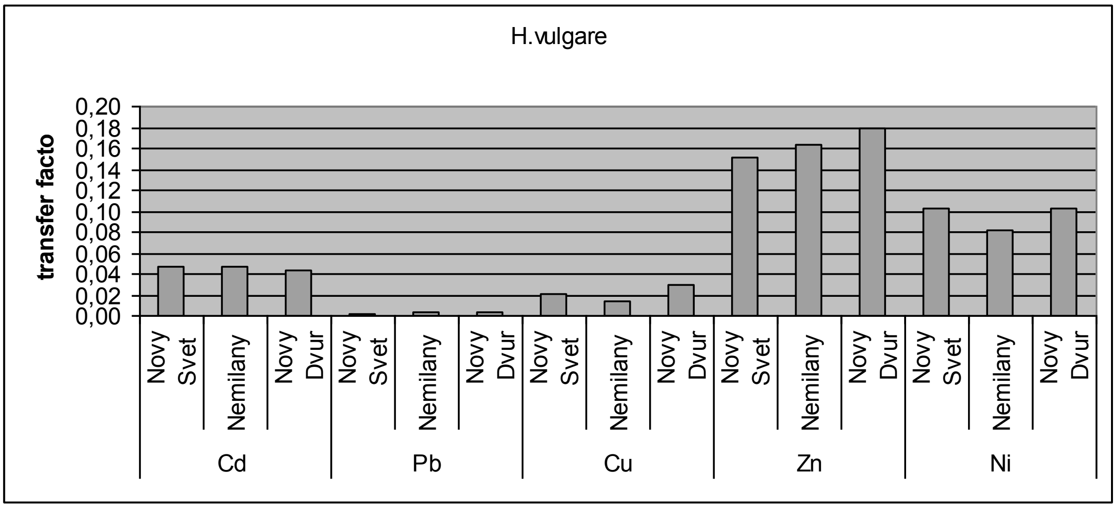


FIGURE 6
AVERAGE TRANSFER FACTORS FOR Pb FOR *D.CAROTA* ON DIFFERENT LOCALITIES

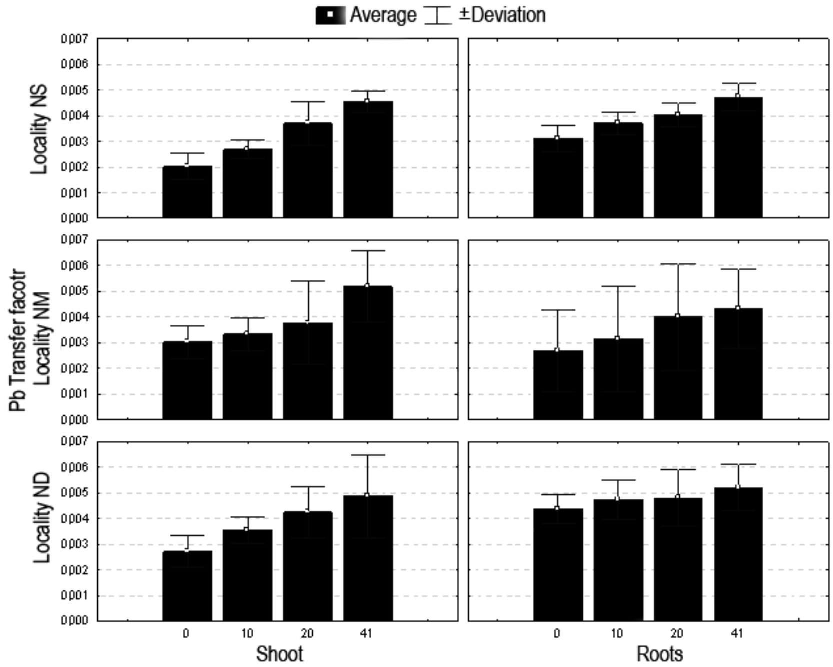
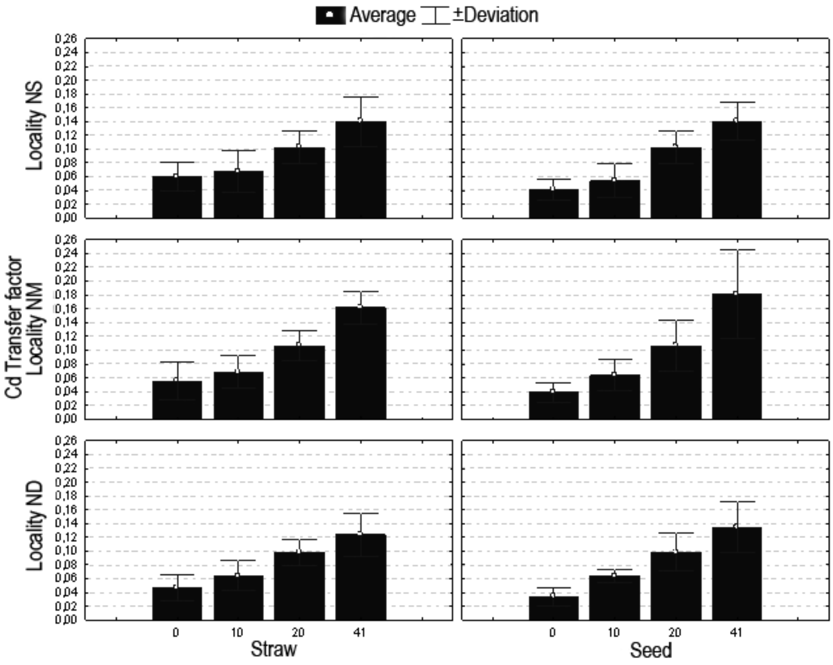


FIGURE 7
AVERAGE TRANSFER FACTORS FOR Pb FOR *H.VULGARE* AT DIFFERENT LOCALITIES



DISCUSSION AND CONCLUSIONS

The total values of the transfer factors (TF) for *D.carota* were: cadmium 0.33, lead 0.003, copper 0.03, zinc 0.11 and nickel 0.1. For *H.vulgare* the values were: cadmium 0.04, lead 0.003, copper 0.02, zinc 0.16 and nickel 0.1. If we focus on both plants, the higher TF levels are almost same except for cadmium (almost seven times higher) and zinc (almost two times higher) for *D.carota*.

Compared with values found by [33], our values of TF were generally lower. One possible explanation could be the higher pH level of the sewage sludge. The pH level of the sludge increased the soil reaction to alkaline (pH_{KCl}=7.2). It could limit the transfer of heavy metals into the plants [34]. This fact is consistent with the findings of other authors [28, 29, 30].

Transfer depends on the bioavailability of the compound in soil. This depends on several factors including pH, soil temperature, organic matter content or soil texture. The pH is of particular importance [35]. Table 7 presents heavy metals mobility according to the pH factor. A value is attributed to each metal between 1 (the least available) and 5 (the most available) to estimate its mobility [36].

TABLE 7
MOBILITY OF METALS IN SOIL

	alkaline, neutral	**acid**
Cd	2	4
Pb	1	3
Cu	1	3
Zn	2	4
Ni	1	4

The biological role of heavy metals in plants depends on their concentration in the plant tissues. Different stages corresponding to deficiency, normal or toxicity levels may be observed according to the metal level in the plant [35].

The concentration of heavy metals in sludge and soil was very low and this is why no differences were found in comparing the control plants and the total value of TF for all localities together. The only exceptions were cadmium and nickel; these values were two times higher for *H.vulgare*.

A comparison of the three locations gave us these results: the most remarkable differences for TF were for cadmium in *D.carota*--values almost two times higher in fluvisols from our Nemilany location than in the other two, and for copper in *D.carota* and *H.vulgare* we found a difference of almost three times higher at the Novy Dvur location. Other elements and localities have almost no discernable differences.

In the final analysis we concentrated on the differences in TF in various parts of the plants - shoot, root, seed and straw. The results are shown in Table 8.

TABLE 8
AVERAGE VALUES OF TRANSFER FACTORS. BOLD VALUES SHOW DIFFERENCES

Element	***D. carota***		***H. vulgare***	
	shoot	**roots**	**straw**	**seed**
Cd	0.32	**0.35**	**0.06**	0.05
Pb	0.004	0.004	**0.004**	0.003
Cu	0.03	0.03	0.02	0.02
Zn	**0.13**	0.11	**0.18**	0.15
Ni	0.07	**0.15**	**0.16**	0.07

As expected, higher TF were primarily detected in the root of *D.carota* except for zinc, equal values were found for lead and copper. Nickel values were two times higher. *H.vulgare* has a better phytoextraction potential in the straw than in the seed. Equal values were detected for copper. Nickel values were more than two times higher.

In general this project proved the phytoextraction potential for both plants grown on fluvisols mixed with sludge as a donor of elements (contamination of heavy metals in fluvisols was purposely left at a natural value). Use of this method for cleaning polluted fluvisols, under real conditions, would be very difficult--most of all due to the time required. This fact results from the comparison of the transfer factors of plants with higher phytoextraction potential [37, 38].

Sewage sludge from municipal wastewater treatment plants contains high quantities of nutrients and organic matter needed for plant growth, but it also contains high heavy metal concentrations. Heavy metals can accumulate in soil and in plants when sludge is applied as a fertilizer and eventually can produce harmful effects in animals and humans. Therefore, the study of effective methods for heavy metal removal from sludge is very important in order to minimize prospective health risks during application [1].

In any case toxic metals from chemical factories can be spread by water and other factors into wide areas that can cause contamination of agricultural soils and, consequently, the food chain [39].

ACKNOWLEDGEMENT

The authors thank the Czech Ministry of Education, Youth and Sports for supporting their research through grant No. 2B06101 from the National Research Program II.

REFERENCES

[1] Lasheen, M. R., Ashmaway, A. and Ibrahim, H. (2000) Heavy metal removal from municipal and industrial sludges. In: 11th Annual International Conference on Heavy metals in the Invironment (J. Nriagu, Editor), Contribution 1431. University of Michigan, School of Public Health, Ann Arbor, MI (CD-ROM)

[2] Yamagata, N. and Shigematsu, I. (1970) Cadmium pollution in perspective. Bulletin of the Institute of Public Health 19, 1-27

[3] Vana, J. (2003) Obsahy tezkych kovu v pude a v rostlinach po dlouhodobem hnojeni prumyslovymi komposty. VURV Praha-Ruzyne (in Czech).
http://stary.biom.cz/sborniky/sb95vana/vana.html

[4] Lombi, E., Zhao, F.J., Dunham, S.J. and McGrath, S.P. (2000) Phytoremediation of Heavy Metal-Contaminated Soils: Natural Hyperaccumulation versus Chemically Enhanced Phytoextraction. Technical report - Bioremediation and Biodegradation.
Markert, B. (1994) Plants as Biomonitors for Heavy Metal Pollution of the Terrestrial Environment. VCH, Weinheim

[5] Blaylock, M.J., Salt, D.E., Dushenkov, S., Zakharova, O., Gussman, C., Kapulnik, Y., Ensley, B.D. and Raskin, I. (1997) Enhanced accumulation of Pb in Indian mustard by soil-applied chelating agents. Environ. Sci. Technol. 31: 860-865

[6] Aderson, T.A., Coats, J.R. (1994) Bioremediation Through Rhizospere Technology. ACS Symposium Series No. 563. American Chemical Society, Washington, DC, USA

[7] Baker, A.J.M., McGrath, S.P., Sidoli, C.D.M. and Reeves, R.D. (1994) The possibility of in site heavy metal decontamination of polluted soils using crops of metal-accumulation plants. Resour. Conserv. Recyc. 11, 41-49

[8] Raskin, I., Kumar, P.B.A.N., Dushenkov, V. and Salt, D.E. (1994) Bioconcentration of heavy metals by plants. Current Opin. Biotech. 5, 285-290

[9] Salt, D.E., Blaylock, M., Kumar, P.B.A.N., Dushenkov, V., Ensley, B.D., Chet, I. et Raskin, I. (1995) Phytoremediation: a novel strategy for the removal of toxic metal from the environment using plants. Bio/Technology 13, 468-478

[10] Kumar, P.B.A.N., Dushenkov, V, Motto, H. and Raskin, I. (1995) Phytoextraction - the use of plants to remove heavy metals from soils. Environ. Sci. Technol. 29: 1232-1238

[11] Kling, J. (1997) Phytoremediation of organics moving rapidly into field trials. Environ. Sci. Technol. 31, A129

[12] Baker, A.J.M. and Brooks, R.R. (1989) Terrestrial higher plants which hyperaccumulate metalic elements. A review of their distribution, ecology and phytochemistry. Biorecovery 1, 81-126

[13] Hegde, R.S. and Fletcher, J.S. (1996) Influence of plant growth stage and season on the release of root phenolics by mulberry as related to development of phytoremediation technology. Chemosphere 32: 2471-2479

[14] Chaudhry, T.M., Hayes, W.J., Khan, A.G. and Khoo, C.S. (1998) Phytoremediation-focusing on accumulator plants that remediate metal contaminated soils. Australasian J. Ecotoxicol. 4, 37-51

[15] Khan, A.G., Chaudhry, T.M., Hayes, W.J., Khoo, C.S., Hill, L., Fernandez, R. and Gallardo, P. (1998) Physical, chemical and biological characterization of a steelworks waste site at Port Kembla, NSW, Australia. Water, Air Soil Pollut. 104, 389-402

[16] Cunningham, S.D. and Ow, D.W. (1996) Promises and prospects of phytoremediation. Plant Physiol. 110, 715-719

[17] Kramer, U., Smith, R.D., Wenzel, W.W., Raskin, I. and Salt, D.E. (1997) The role of metal transport and tolerance in nickel hyperaccumulation by Thelapsi geosingense Halacsy. Plant Physiol. 115, 1641-1650

[18] Watanabe, M.E. (1997) Phytoremediation on the brink of commercialization. Environ. Sci. Technol. 31:182-186

[19] Chaney, R.L., M. Malik, Y.M. Li, S.L. Brown, E.P. Brewer, J.S. Angle and Baker, A.J.M. (1997) Phytoremediation of soil metals. Curr. Opin. Biotechnol. 8: 279-284

[20] Shen, Z.G., Zhao, F.J. and McGrath, S.P. (1997) Uptake and transport of zinc in the hyperaccumulator Thlaspi caerulescens and the non-hyperaccumulator Thlaspi ochroleucum. Plant Cell Environ. 20: 898-906

[21] Huang, J.W. and Cunningham, S.D. (1996) Lead phytoextraction: Species variation in lead uptake and translocation. New Phytol. 134: 75-84

[22] Banuelos, G.S., Ajwa, H.A., Mackey, B., Wu, L., Cook, C., Akohoue, S. and Zambruzuski, S. (1997) Evaluation of different plant species used for phytoremediation of high soil selenium. J. Environ. Qual. 26: 639-646

[23] Huang, J.W., Chen, J.J., Berti, W.R. and Cunningham, S.D. (1997) Phytoremediation of lead-contaminated soils: Role of synthetic chelates in lead phytoextraction. Environ. Sci. Technol. 31: 800-805

[24] Ajwa, H.A., Banuelos, G.S. and Mayland, H.F. (1998) Selenium uptake by plants from soils amended with inorganic and organic materials. J. Environ. Qual. 27: 1218-1227

[25] Ebbs, S.D. and Kochian, L.V. (1998) Phytoextraction of zinc by oat (Avena sativa), barley (Hordeum vulgare) and Indian mustard (Brassica juncea). Environ. Sci. Technol. 32: 802-806

[26] Wei, F.S. (1992) Modern Analytical Methods of Soil Elements. Chinese Environ. Sci. Press, Beijing, p. 64

[27] Zbiral, J. (1994) Analyza rostlinneho materialu, UKZUZ OAPUR. (in Czech)

[28] Herms, U. (1989) Löslichkeit von Schwermetallen in Böden unter variierenden Milieubedingungen: In: Dechema (Hrsg. von D. Behrens u J. Wiesner, 1989). Beurleilung von SK-Kontamination im Boden. Frankfurt/Main: 189-199

[29] Cottenie, A. and Kiekens, L. (1981) Beweglichkeit von Schwermetallen in mit Schlamm angereicherten Böden. Korrespondenz Abwasser 28: 206-211

[30] Sauerbeck, D. (1989) Der Transfer von Schwermetallen in die Pflanze.
In: Beurteilungen von Schwermetallkontaminationene im Boden, Dechema. Frankfurt (Main): 281 - 316

[31] McConnell, D.B., Shiralipour, A. and Smith, W.H. (1993) J. Environ. Qual. 26, 534
Negri, M.C. and Hinchman, R.R. (1996) Plants that remove contaminants from the environment. Lab. Med. 27, 36-40

[32] Smith, S.R. (1993) Environ. Pollut. 85, 387

[33] Diez, T. and Krauss, M. (1995) Odcerpavani tezkych kovu z kontaminovanych ploch pomoci rostlin. In: sbornik Tezke kovy v zemedelske pudy a rostlinach. Praha: 41 – 43 (in Czech).

[34] Badalikova, B., Hruby, J. and Hartman, I. (2005) Effect of humus content in soil on the uptake of heavy metals by plants. In: CD from ISTRO Conference "Soil - Agriculture, Environment, Landscape", Brno, Czech Republic 135-141

[35] European Environment Agency (2006) Disposal and recycling routes for sewage sludge
http://ec.europa.eu/environment/waste/sludge/sludge_disposal.htm

[36] Ademe, A., Recyval, P. (2001) Kompost de boues de stations d´épuration municipales: qualité performance agronomiques et utilisations.
http://ec.europa.eu/environment/waste/sludge/sludge_disposal3a.pdf

[37] Diez, T. and Krauss, M. (1995) Prijem tezkych kovu plodinami z ruzne kontaminovanych pud - Bayerische Landesanstalt für Bodenkultur und Pflanzenbau Freising, BRD. http://stary.biom.cz/sborniky/sb95vana/diez3.html

[38] Vana, J. (1995) Dekontaminace tezkych kovu z pudy rostlinami. Vyzkumny ustav rostlinne vyroby Praha-Ruzyne. (in Czech)
http://stary.biom.cz/sborniky/sb97PrVana/sb97PrVana_vana.html

[39] Jizba, J. (2003) Metoda odstranovani tezkych kovu z pudy. Envicho s.r.o., Chomutov (in Czech)
http://stary.biom.cz/sborniky/sb95vana/jizba.html