Developments in Environmental Science, Volume 5
D. Sarkar, R. Datta and R. Hannigan (Editors)

ISSN: 1474-8177/DOI:10.1016/S1474-8177(07)05005-X

Chapter 5

Phytoremediation of some heavy metals by agronomic crops

Honey Aggarwal and Dinesh Goyal

Abstract

Remediation of metal contaminated soil faces challenges, as unlike organic contaminants metals cannot be degraded. Phytoremediation is an emerging technology that employs the use of higher plants for the cleanup of contaminated environments. Plant-based approach for remediation is cost effective and takes advantage of their remarkable ability to grow and uptake heavy metals from severely polluted sites. Among several crops, *Brassica juncea*, *Cucurbita pepo*, *Amaranthus sp.*, *Raphanus sativus oleiformis*, *Zea mays*, etc., has been demonstrated for remediation of cadmium (Cd), nickel (Ni), lead (Pb), zinc (Zn), selenium (Se), chromium (Cr), copper (Cu) and uranium (U). This review compiles the relevant information on possible uses of agronomic crops for metal extraction as an alternative for the removal of heavy metals excess from soil.

5.1. Introduction

A large portion of biosphere is contaminated by heavy metals as a result of human activities. Conventional solutions such as disposal of contaminated soil in landfills, which relies heavily on 'dig and dump' or encapsulation, neither of which takes into consideration the issue of decontamination of the soil, account for a large proportion of the remediation operations at present (Pulford and Watson, 2003). Remediation techniques like immobilization or extraction by physico-chemical techniques are expensive and are often appropriate only for small areas. On the other hand, for the decontamination of polluted sites phytoremediation seems attractive as it offers site restoration, partial decontamination, maintenance of biological activity and biorecovery of metals (Baker et al., 1991, 1994; Gardea-Torresdey et al., 2005a). It has attracted attention for its low cost of implementation and environmental benefits.

Phytoremediation is the use of green plants as well as associated rhizospheric microbes to remove pollutants from the environment or to render them harmless (Raskin et al., 1994; Salt et al., 1998). It is an innovative biological technique, which can be applied for the cleanup of severely polluted soil (Salt et al., 1995, 1998; Chaney et al., 1997; Chaudhry et al., 1998; Meagher, 2000; Lasat, 2002; Prasad and Freitas, 2003; Pulford and Watson, 2003; Alkorta et al., 2004; Gardea-Torresdey et al., 2005a).

Phytoremediation of metals occurs due to following activities:

- Phytoextraction: Pollutant accumulating plants remove metals from the soil and concentrate them in the harvestable part of plants (Kumar et al., 1995a).
- Rhizofiltration: Removal of contaminants from aqueous waste streams by absorption onto plant roots (Dushenkov et al., 1995).
- Phytostabilization: Immobilization or prevention of migration of contaminants in the environment by plant exudates, leading to the reduction in the mobility and bioavailability of the contaminants (Vangronsveld et al., 1995).
- Phytovolatilization: Volatilization of pollutants into the atmosphere via plants (Burken and Schnoor, 1997; Banuelos et al., 1997a).
- Phytomining: Capability of plants to extract large amount of metals from soils that can be exploited to recover metals of high economic value from ore deposits and other soils (Glass, 2000; Gardea-Torresdey et al., 2005a).

Governments worldwide are establishing research and decontamination programs to use this potential. Environment Canada has developed a database (PHYTOREM) of 775 plants with capabilities to accumulate or hyperaccumulate one or several of 19 key metallic elements (McIntyre, 2003). Phytoremediation is easier to manage because it is an autotrophic system of large biomass that requires little nutrient input (Evans and Furlong, 2003). Moreover, plants offer protection against water and wind erosion and in preventing spreading of contaminants (Pulford and Watson, 2003).

The success of phytoremediation as an environmental-cleanup technology depends upon number of factors including the extent of soil contamination, metal availability for uptake into roots and plant ability to intercept, absorb and accumulate metals in shoots (Ernst, 1996). This review aims to give a broad overview of the various phytoremediation technologies and their potential role in clean up of pollutants especially heavy metals.

5.2. Phytoextraction

The concept of using plants to cleanup contaminated environment is very old and cannot be traced to any particular source (Blaylock and Huang, 2000). About 300 years ago, plants were proposed for use in the treatment of wastewater (Hartman, 1975). At the end of the 19th century, *Thlaspi caerulescens* and *Viola calaminaria* were the first plant species documented to accumulate high levels of metals in leaves (Baumann, 1885). Plants able to accumulate upto 1% Ni in shoots were identified (Minguzzi and Vergnano, 1948). The idea of using plants to extract metals from contaminated soil was reintroduced and developed by Utsunamyia (1980) and Chaney (1983). At present, there are two basic strategies of metal phytoextraction; natural phytoextraction and induced or chemically assisted phytoextraction (Salt et al., 1998).

Phytoextraction is based on the use of pollutant accumulating plants for the removal of metals and organics from soil by concentrating them in the harvestable parts (Brooks, 1977; Salt et al., 1998; Reeves and Baker, 2000; Vassilev et al., 2004). Metal phytoextraction is not as extreme as conventional metal removal methods but still involves considerable alterations in the environment, which includes elimination of the existing vegetation cover and application of fertilizers and various soil amendments to increase metal availability to plants (Tichy et al., 1997; Ebbs et al., 1998; Huang et al., 1998; Pawlowska et al., 2000). Plants for phytoextraction, i.e., metal removal from soil, should have the following characteristics: (i) tolerant to high levels of the metal, (ii) accumulate reasonably high levels of the metal, (iii) rapid growth rate, (iv) produce reasonably high biomass in the field and (v) profuse root system (Garbisu et al., 2002).

The roots of the established hyperaccumulators absorb metal elements from the soil and translocate them to the above ground shoots, where they get accumulated in high concentration (Prasad and Freitas, 2003). It is also based on high biomass-producing plants used together with chemical agents enhancing both metal solubility and uptake by plants (Blaylock et al., 1997; Huang et al., 1997). After sufficient plant growth and metal accumulation, the above ground portions of the plants are harvested and removed, which results in the permanent removal of the metals from site. After removal of heavy metals from the soil, the disposal of the contaminated material is an environmental concern. Some researchers suggested incineration (Kumar et al., 1995a), while others suggested about the extraction of valuable metals from the metal-rich ash (Comis, 1996; Cunningham and Ow, 1996).

In general, the reports assessing metal phytoextraction potential are based on pot experiments, when compared to field experiments higher metal-extracting values have been observed, which might be due to higher solubility of metals and the effect of amendments aiming at mobilizing the metals etc. (Vassilev et al., 2004). The selection of heavy metal tolerant species is a reliable tool to achieve success in phytoremediation. One hundred and sixty three plant taxa belonging to 45 families have been found to be metal tolerant and are capable of growing on elevated concentration of toxic metals (Prasad and Freitas, 2003). The use of metal tolerant species and their metal indication and accumulation is a function of immense use for biogeochemical prospecting (Brooks, 1983; Badri and Springuel, 1994; Mcinnes et al., 1996). Chemically assisted phytoextraction is based on the use of non-accumulator plants with metal accumulation levels far below those of hyperaccumulators but with high biomass potential (Vassilev et al., 2004). This is aimed to overcome the main limitations of natural phytoextraction, i.e., a limited number of suitable hyperaccumulators for some important metal pollutants such as Pb (Huang et al., 1997; Lasat, 2000), as well as their low biomass production. Lopez et al. (2005) has shown the combined effects of ethylenediaminetetraacetic acid (EDTA) and phytohormone indole-3-acetic acid (IAA) on Pb uptake by *Medicago sativa.* After 10 days of treatment with 0.2 mM Pb and different combinations of EDTA and IAA, the quantification of Pb content in plant tissues using an inductively coupled plasma optical emission spectrometer (ICP/OES) showed that Pb accumulation in leaves was increased by about 2800% in plants cultivated with Pb/EDTA as compared to plants exposed to Pb alone respectively, where it was increased by only 600%.

The first field trial on natural phytoextraction was conducted in 1991–1992 in sewage sludge treated plot at Woburn, England (McGrath et al., 1993). The maximum Zn uptake was found in *Thlaspi caerulescens* accumulating 2000–8000 mg Zn/kg. It was also shown to accumulate 1000–4000 mg kg^{-1} Cd (Brooks, 1998). *Sedum alfredii* Hance has been identified as a new Zn and Cd hyperaccumulating plant species (Yang et al., 2004). Zn concentration in its shoot can reach over 20 g kg^{-1} when grown at 80 mg Zn/l in nutrient solution without showing any toxic symptoms (Yang et al., 2002). Cd concentration in leaves and stem of *S. alfredii* increased with increasing Cd supply levels, and reached a maximum upto 9000—65,000 mg kg^{-1} dry weight (Yang et al., 2004). Seedlings of *Sesbania drummondii* can hyperaccumulate Pb in a controlled hydroponic environment (Sahi et al., 2002). Green bean, beetroot, green cabbage, lettuce, onion, pea, radish, spinach, tomato, turnip, watercress and Iberis were grown on a thallium-contaminated soil and *Iberis intermedia* was found to hyperaccumulate thallium (LaCoste et al., 2001).

Maximum thallium levels ranged from nearly 400 mg kg^{-1} in *Iberis intermedia* down to just over 1 mg kg^{-1} in green bean. High Cu concentration has been found in *Betula* roots (Kozlov et al., 1995; Maurice and Lagerkvist, 2000) as well as in *Salix* roots (Punshon and Dickinson, 1997). The Pb, Zn and Cd phytoextraction potential of 14 different plants was assessed in a chelate induced phytoextraction experiment. EDTA and EDDS (ethylenediamine disuccinic acid) were used as chelates. The addition of these chelates increased the proportion of phytoavailable Pb, Zn and Cd in the soil and also their uptake by tested plants upto 48 times by *Sinapsis alba*, 4.6 times by *Raphanus sativus oleiformis* and 3.3 times by *Amaranthus sp*., respectively. *Cannabis sativa* hyperaccumulated 105 times Pb, 2.3 times Zn and 31.7 times Cd higher than control (Kos et al., 2003). Blaylock et al. (1997) and Huang et al. (1997) found that application of EDTA at 2 g kg^{-1}soil resulted in a concentration of more than 1.5% Pb in the shoots of *Brassica juncea* and 1% in maize and pea plants. It was also shown that other chelators such as EGTA (ethylene–bis [oxy ethyletrinitrilo] tetracetic acid) had high affinity to Cd, while DTPA (diethylene triamine pentacetic acid) showed high affinity to Zn (Blaylock et al., 1997). Turgut et al. (2004) used two cultivars of *Helianthus annuus* in conjunction with EDTA and CA (citric acid) as chelators for phytoremediation of heavy metal contaminated soil. Results showed that EDTA at a concentration of 0.1 g kg^{-1} yielded the best results for both cultivars achieving a total metal (Cd, Cr and Ni) uptake of approximately 0.73 mg.

Restrictions apply, however, to both use of complexing agents and artificial soil acidification. It was found that EDTA and EDTA-heavy metal complexes are toxic for some plants and high doses of EDTA inhibited the development of arbuscular mycorrhiza (Dirilgen, 1998; Creman et al., 2001; Geebelen et al., 2002). *In-situ* application of chelating agents can cause groundwater pollution by uncontrolled metal dissolution and leaching (Creman et al., 2001; Sun et al., 2001).

The ideal plant species for metal phytoextraction has to be highly productive in biomass and to uptake and translocate a significant part of metals to its shoots (Vassilev et al., 2004). Some tree species mainly *Salix* sps. and *Populus* sps. exhibit these traits and are already used in phytoremediation programmes and for Cd phytoextraction from lightly polluted agricultural soils (Landberg and Greger, 1994). In fact, *Salix* sps. are not metal hyperaccumulators, but it was shown that among different clones there are some species which are hyperaccumulators of Cd and Zn. About 150 clones of *Salix* sps. have been screened for uptake, transport of metals to shoots and tolerance to Cd, Zn and Cu (Landberg and

Greger, 1994; Landberg and Greger, 1996). Some reports by Grant and Bailey (1997), Yankov et al. (2000) and Yankov and Tashin (2001), Griga et al. (2003) showed that crops for fibre or oil production could be used for profitable crop production accompanied by phytoextraction of metal from polluted soils. Brake fern (*Pteris vittata*) was reported to tolerate soils contaminated with arsenic as much as 1500 ppm and its fronds were found to concentrate up to 22,630 ppm in six weeks (Ma et al., 2001). Wang et al. (2002) showed that among maize, wheat, rapeseed, field pea and fodder vetch grown on a multiple metal contaminated site, maize had the highest concentrations of Mn, Zn and Cd, rapeseed had the highest concentration of Cr, Cu concentration was highest in fodder vetch and Pb was highest in wheat, but heavy metal accumulation was there in grain of wheat. The results suggested that on sites with multiple metal contaminations, growing maize and rapeseed would be safer than growing wheat or legumes and could be used for phytoremediation of lightly contaminated soils. A study by Peralta-Videa and Gardea-Torresdey (2002) showed that Alfalfa (*Medicago sativa*) plants were able to take up metals from a mixture of Cd(II), Cu(II), Ni(II) and Zn(II) in soils. The maximum relative uptakes in comparison to control were found to be 36 times for Ni, 23 times for Cd, 12 times for Zn and 6 times for Cu. In another field trial the possibility of using *Beta vulgaris*, *Cichorium intybus*, *Cucurbita pepo*, *Phaseolus vulgaris*, *Hordeum vulgare*, *Brassica oleracea*, *Zea mays*, *Medicago sativa* and *Pastinaca sativa* for removing heavy metals from soil was investigated. Results showed that the most effective crop for phytoextraction of Cd, Cu, Ni, Pb and Zn was *Cucurbita pepo* and for Cr – *Zea mays* (Ciura et al., 2005).

The main advantage of this technology is its lower cost as compared to other known remediation technologies (EPA, 2000; Glass, 2000). The possible metal recycling should provide further economic advantage as the ash of some hyperaccumulators consists of significant amount of metals (20–40% Zn for *T. caerulescens*) and there is no need to pay for safe disposal (Chaney et al., 1997), it can work without further disturbing the site, which is of great importance for its public acceptance (Vassilev et al., 2004). Besides all its advantages, it has certain limitations also, the major limitation is that it can only be used for low to moderately contaminated soils and it is applicable only to surface soils with few exceptions and is a time consuming process (Robinson et al., 1998; Blaylock and Huang, 2000; Vassilev et al., 2004). It is still at developmental stage, small companies and universities are driving much of its innovation and research, whereas, environmental engineering firms are involved in application projects.

5.3. Rhizofiltration

Rhizofiltration refers to the use of plant roots to sorb, concentrate and precipitate metal contaminants from surface or groundwater (Dushenkov et al., 1995). It is effective in cases where wetlands can be created and all of the contaminated water is allowed to come into contact with roots. Contaminants should be those that sorb strongly to roots such as hydroponic organics, Pb, Cr(III), etc. Rhizofiltration is primarily used to remediate extracted groundwater, surface water and wastewater with low concentration of contaminant and it can be very cost effective (Salt et al., 1995). It can be used for Pb, Cd, Cu, Ni, Zn and Cr, which are primarily retained within the roots (USEPA, 2000). An ideal plant for rhizofiltration should have rapidly growing roots with the ability to remove toxic metals from solution over extended periods of time. Dushenkov et al. (1995) demonstrated that many 'large root' species have the ability to absorb and precipitate heavy metals from solution, such as sunflower (*Helianthus sp.*), rye (*Elymus sp.*), corn (*Zea mays*) and Indian mustard (*Brassica juncea*). The mechanisms of toxic metal removal by plant roots are not necessarily similar for different metals.

In rhizofiltration, plants used have the ability to remove upto 60% of their dry weights as toxic metals (Salt et al., 1995). The process involves raising plants hydroponically and transplanting them into metal polluted waters where plants absorb and concentrate the metals in their roots and shoots (Dushenkov et al., 1995; Salt et al., 1995; Flathman and Lanza, 1998; Zhu et al., 1999). Root exudates and changes in the pH of rhizosphere soil may also cause metals to precipitate onto root surfaces. Plants for rhizofiltration should be able to accumulate and tolerate significant amounts of the target metals alongwith easy handling, low maintenance cost and a minimum of secondary waste requiring disposal (Dushenkov and Kapulnik, 2000). Several aquatic species have the ability to remove heavy metals from water, including water hyacinth (*Eichhornia crassipes*; Kay et al., 1984; Zhu et al., 1999), pennywort (*Hydrocotyle umbellate* L.; Dierberg et al., 1987) and duckweed (*Lemna minor* L.; Mo et al., 1989). As a result of their small, slow growing roots these plants are not much efficient at metal removal and have limited potential for rhizofiltration (Dushenkov et al., 1995). Sunflower (*Helianthus sp.*), Indian mustard (*Brassica juncea*), tobacco (*Nicotiana tabacum*), rye (*Elymus sp.*), spinach (*Spinacea oleracea*) and corn (*Zea mays*) have been studied for their ability to remove Pb from water, with sunflower having the greatest ability. In a study, after only 1 h of treatment sunflower reduced Pb concentration significantly (Raskin and Ensley, 2000). Indian mustard had bioaccumulation coefficient of 563 for Pb and had proven to be effective in removing a wide concentration range of Pb (4–500 mg l^{-1}) (Raskin and Ensley, 2000; USEPA, 2000). The hairy root cultures of *Brassica napus* were used to study the removal of 2,4-dichlorophenol (2,4-DCP), a common contaminant in industrial effluents that is highly toxic for human and aquatic life. High removal efficiencies (93–95%) were observed in a broad pH range (pH 3–9), reaching 98–99% in the pH range 4–8 (Agostini et al., 2003). The hairy root systems of *Brassica napus* and *Chenopodium amaranticolor* were used for removal of uranium from the solution of concentration up to 5000 μM. The results indicated that the hairy roots could remove uranium from the aqueous solution within a short period of incubation. *Brassica juncea* could take up 20–23% of uranium on dry weight basis and *Chenopodium amaranticolor* showed a slow and steady uptake of uranium upto 13% (Eapen et al., 2003). In pot culture experiment, palak (*Beta vulgaris* L.), coriander (*Coriandrum sativum*) and fenugreek (*Trigonella foenum-graecum*) grown in soil contaminated with heavy metals (Fe, Zn, Cu, Pb, Cr, Ni, Cd) showed elevated levels of heavy metals in roots as compared to shoot in the order of $Fe > ZnNi > Cr > Cd > CuPb$ (Jaj, 2005).

The advantages associated with the rhizofiltration are the ability to use both terrestrial and aquatic plants for either *in-situ* or *ex-situ* applications, applicability to many problem metals, ability to treat high volumes, lesser need for toxic chemicals, reduced volume of secondary waste, possibility of recycling and likelihood of public acceptance (Dushenkov et al., 1995; Kumar et al., 1995b; Raskin and Ensley, 2000). The disadvantages include the constant need to maintain pH, plants may first need to be grown in a nursery and then transplantation and maintenance of successful hydroponic systems in the field would require expertise of qualified personnel, periodic harvesting and plant disposal and a good understanding of the chemical speciation (USEPA, 2000).

5.4. Phytostabilization

Phytostabilization is defined as immobilization of a contaminant in soil through absorption and accumulation by roots, adsorption onto roots or precipitation within the root zone of plants. It is also known as phytorestoration. It is a plant-based remediation technology that stabilizes wastes and prevents exposure pathways via wind and water erosion; provides hydraulic control, which suppresses the vertical migration of contaminants into groundwater and physically and chemically immobilizes contaminants by root sorption and by chemical fixation with various soil amendments (Cunningham et al., 1995; Salt et al., 1995; Flathman and Lanza, 1998; Berti and Cunningham, 2000; Schnoor, 2000). Phytostabilization of organic pollutants that are foreign to living organisms is based on sequestration processes, such as humification (McCutcheon and Schnoor, 2003). Phyostabilization involves root zone microbial and chemical processes. It can change metal solubility and mobility or impact the dissociation of organic compounds. The plant affected soil environment can convert metals from a soluble to an insoluble oxidation state (Salt et al., 1995). Phytostabilization can occur through sorption, precipitation, complexation or metal valence reduction (EPA, 1997). In a vegetative cap for phytostabilization, a combination of trees and grasses may be used. Fast transpiring trees such as poplar maintain an upward flow to prevent downward leaching, while grasses prevent wind erosion and lateral runoff with their dense root systems (Bennet et al., 2003; McCutcheon and Schnoor, 2003).

Sometimes there is no immediate effort to clean metal polluted sites, either because the responsible companies no longer exist or because the sites are of no high priority on a remediation agenda (Berti and Cunningham, 2000). Metal tolerant plants are required for heavy metal contaminated soils. Plants chosen for phytostabilization should be poor translocaters of metal contaminants to above ground plant tissues that could be consumed by humans and animals. The plants selected should be easy to establish and care for, grow quickly, have dense canopies and root systems and be tolerant to metal contaminants and other site conditions that may limit plant growth. *Brassica juncea* has been shown to reduce leaching of metals from soil by over 98% (Raskin et al., 1994). Arsenic might be taken up by plants because it is similar to the plant nutrient phosphate, although poplar leaves in a field study did not accumulate amounts of As. Poplars were grown in soil containing an average of $1250\,\mathrm{mg\,kg^{-1}}$ As (Pierzynski et al., 2002). Grasses were used to stabilize mine wastes containing Cu (Salt et al., 1995). The research of Smith and Bradshaw (1992) led to the development of two cultivars of *Agrostis tenuis* and one *Festuca rubra*, which are now commercially available for phytostabilization of Pb, Zn and Cu contaminated soils. Soil amendments can also be used to stabilize metals in soils. Amendments should be selected that will maximize the growth of vegetation, which then also helps to phytostabilize the soil (Berti and Cunningham, 2000).

It has advantages over other soil remediation practices in that it has a lower cost and is less disruptive than other more vigorous soil remedial technologies (EPA, 2000), easy to implement (Schnoor, 2000). Revegetation offers aesthetic value and enhances ecosystem restoration. The lack of appreciable metals in shoot tissue also eliminates the necessity of treating harvested shoot residue as hazardous waste (Flathman and

Lanza, 1998). The main disadvantage is that the contaminants remain in place. The vegetation and soil may require long-term maintenance to prevent release of the contaminants and future leaching (EPA, 2000). Highly contaminated sites are not suitable for phytostabilization, because plant growth and survival is not possible (Berti and Cunningham, 2000).

5.5. Phytovolatilization

Phytovolatilization is a multimedia transfer of contaminants from water or soil to the atmosphere. Volatile organic compounds are taken up and transpired with water vapour or diffused out of the leaves, stems and roots (McCutcheon and Schnoor, 2003). Plants normally transpire water as vapour, but volatile compounds can be transpired as well. It occurs via diffusion from tree's xylem through its bark or leaves. In recent years, researchers have searched for plants that are capable of absorbing elemental forms of these metals from the soil, biologically converting them to gaseous species within the plant and releasing them into the atmosphere (Prasad and Freitas, 2003). Phytovolatilization has mainly been applied to groundwater, but it can be applied to soil, sediments and sludge. Plants may serve as effective pump and treat systems for mobile contaminants. Selenium was taken up and transpired from groundwater with concentrations of 100–500 $\mu g\, l^{-1}$ Se (Banuelos et al., 1997a) and at soil concentrations of 40 $mg\, l^{-1}$ (Banuelos et al., 1997b). Indian mustard (*Brassica juncea*) and canola (*Brassica napus*) have been used in the phytovolatilization of Se (Banuelos et al., 1997b). Lewis et al. (1966) first showed that both selenium non-accumulator and accumulator species volatilize selenium. Selenium (as selenate) was converted to less toxic dimethyl selenite gas and released to the atmosphere (Adler, 1996). Kenaf (*Hibiscus cannabinus* L. cv. Indian) and tall fescue (*Festuca arundinacea Schreb* cv. Alta) have also been used to take up Se, but to a lesser degree than canola (Banuelos et al., 1997b). Some aquatic plants, such as cattail (*Typha latifolia* L.) are also good for Se phytoremediation (Pilon-Smits et al., 1999). Plants that volatilize Hg were genetically modified, for example, *Arabidopsis thaliana* L. and *Nicotiana tabaccum* L. with bacterial organomercurial lyase (*MerB*) and mercuric reductase (*MerA*) genes (Heaton et al., 1998; Rugh et al., 1998). These plants absorb elemental Hg(II) and methylmercury (MeHg) from the soil and release volatile Hg(0) from the leaves into the atmosphere (Heaton et al., 1998). Heaton et al. (1998) suggested that the addition of Hg(0) into the atmosphere would not contribute significantly to the atmospheric pool.

Advantage of phytovolatilization is that the contaminants could be transformed to less-toxic forms, such as elemental mercury and dimethyl selenite gas, without need for plant harvesting and disposal. Contaminants or metabolites released to the atmosphere might be subjected to more effective or rapid natural degradation processes such as photodegradation. The only disadvantage is that the contaminant or a hazardous metabolite might be released into the atmosphere or get accumulated in vegetation (Newman et al., 1997).

5.6. Phytomining

Phytomining is defined as the production of crop of metal by growing high biomass plants that accumulate high metal concentration. Some of these plants are natural metal hyperaccumulators, while in others the property can be induced (Brooks, 1998). Plant ability to extract large amount of metals from soils can be exploited to recover metals of economic value from ore deposits and other soils (Glass, 2000). Studies have shown that using certain plants to extract metals from soil is commercially feasible. It may be a green alternative to destructive, opencast mining practices. It could be used to mine metals that are uneconomic by conventional methods (Brooks, 1998).

Research conducted at the University of Texas in El Paso, USA, has shown that gold accumulated by alfalfa plants and stored in leaf and stem biomass can be present as discrete nanoparticles of pure metal (Gardea-Torresdey et al., 2002). This discovery was made after alfalfa sprouts germinated on gold chloride enriched agar ($320\,\mathrm{mg\,kg^{-1}}$ Au) were analysed using X-ray absorption spectroscopy (XAS) and transmission electron microscopy (TEM). The gold recovery rate in plants has been observed for many tested artificial and real substrates (Anderson et al., 1998). Preliminary studies by Gardea-Torresdey et al. (2005b) have shown that desert willow (*Chilopsis linearis*) is able to extract gold from a gold enriched medium. It grew well in the presence of NH_4SCN lower than $1 \times 10^{-4}\,\mathrm{mol\,l^{-1}}$. After two weeks the effect on plant growth and gold content was determined by inductively coupled plasma-optical emission spectroscopy (ICP-OES). The results showed that addition of $10^{-4}\,\mathrm{mol\,l^{-1}}$ NH_4SCN increased the concentration of Au by about 595, 396 and 467% in roots, stems and leaves, respectively. It was found that shoot elongation was not affected by thiocyanate. XAS studies showed that the oxidation state of gold was Au(0) and Au nanoparticles were formed inside the plants and *C. linearis* is a potential plant for Au phytomining (Gardea-Torresdey et al., 2005b). XAS has provided important information on the coordination chemistry of metals and toxic element interactions with phytoremediation systems. It has provided information in terms of the coordination environment of metals absorbed by plants and the bioreduction of metals within phytoremediation systems. It also provided information about the production of Au and Ag nanoparticles by metal interaction with the plants on phytomining systems (Gardea-Torresdey et al., 2005a). Broadhurst et al. (2004) had developed commercially viable phytomining technologies employing *Alyssum bertolonii* Ni-hyperaccumulator species, where the majority of Ni was stored either in the leaf epidermal cell vacuoles, or in the basal portions of the numerous stellate trichomes. The metal concentration in the trichome basal compartment was the highest ever reported for healthy vascular plant tissue, approximately 15–20% dry weight (Broadhurst et al., 2004).

Keeling et al. (2003) investigated the potential of South African high biomass Ni hyperaccumulator *Berkheya coddii* to phytoextract Co and Ni from artificial metalliferous media. Plant accumulation of both metals from single element substrates indicated that the bioaccumulation coefficient increased as total metal concentration increased. An important step in the phytomining operations is the recovery of metals from harvested plant material. In this work, a laboratory scale horizontal tube furnace was used to generate Ni-enriched bio-ore from the dried biomass of Ni-hyperaccumulator plants. Prior to furnace treatment, hairy roots of *Alyssum bertolonii* were exposed to Ni in liquid medium to give biomass having Ni concentration of 1.9–7.7% dry weight; whole plants of *Berkheya coddii* biomass was about 15 times greater than in *A. bertolonii.* After furnace treatment at 1200°C under air, Ni-bearing residues with crystalline morphology and containing upto 82% Ni were generated from *A. bertolonii* (Boominathan et al., 2004).

5.7. Conclusion

In recent years, phytoremediation has emerged as a promising low cost and environment friendly remediation technology, especially relevant for moderately polluted areas which is gradually approaching commercialization. Plants and associated microorganisms can remediate heavy metal contaminated soil via phytoextraction, rhizofiltration, phytostabilization, phytovolatilization and phytomining. Phytoremediation works effectively for a wide range of inorganic pollutants, the underlying biological processes are still largely unknown in most of the cases (Pilon-Smits, 2005). It is advantageous to use commonly cultivated agronomic crops such as

Table 5.1. Agronomic crops for phytoremediation of heavy metals from soil

Plant species	Heavy metals	Method/activity	Reference
Agrostis tenuis	Pb, Zn, Cu	Phytostabilization	Smith and Bradshaw (1992)
Alyssum bertolonii	Ni	Phytomining	Boominathan et al. (2004); Broadhurst et al. (2004)
Alyssum murale	Ni, Zn	Phytoextraction	Whiting et al. (2003)
Amaranthus sp.	Cd, Pb, Zn,	Phytoextraction	Kos et al. (2003)
Arabidopsis thaliana L.	Hg	Rhizofiltration	Heaton et al. (1998)
Avena sativa	Zn	Phytoextraction	Ebbs et al. (1997)
Berkheya coddii	Ni, Co	Phytoextraction	Keeling et al. (2003)
Brassica juncea	Cd, Cu	Phytoextraction	Kumar et al. (1995a)
Brassica juncea	Cd, Pb, Ni, Zn, Cr, Cu	Rhizofiltration	Dushenkov et al. (1995)
Brassica juncea	Ni, Cu	Phytoextraction	Ebbs et al. (1997)
Brassica juncea	Se	Phytovolatilization	Banuelos et al. (1997b)
Brassica napus	Zn	Phytoextraction	Ebbs et al. (1997)
Brassica napus	Cd	Phytoextraction	Wang et al. (2002)
Brassica napus	Se	Phytovolatilization	Banuelos et al. (1997b)
Brassica napus	U	Rhizofiltration	Eapen et al. (2003)
Cannabis sativa	Cd, Ni, Cr	Phytoextraction	Citterio et al. (2003)
Chenopodium amaranticolor	U	Rhizofiltration	Eapen et al. (2003)
Chilopsis linearis	Au	Phytomining	Gardea-Torresdey et al. (2005b)
Cucurbita pepo	Pb, Ni, Zn, Cu	Phytoextraction	Ciura et al. (2005)
Festuca rubra	Pb, Zn, Cu	Phytostabilisation	Smith and Bradshaw (1992)
Helianthus annuus	Cd, Ni, Cr	Phytoextraction	Turgut et al. (2004)
Iberis intermedia	Th	Phytoextraction	LaCoste et al. (2001)
Medicago sativa	Cd, Ni, Zn, Cu	Phytoextraction	Peralta-Videa and Gardea-Torresdey (2002)
Medicago sativa	Pb	Phytoextraction	Lopez et al. (2005)
Medicago sativa	Au	Phytomining	Gardea-Torresdey et al. (2002)
Nicotiana tabacum L.	Hg	Rhizofiltration	Heaton et al. (1998)
Pteris vittata	As	Phytoextraction	Ma et al. (2001)
Raphanus sativus oleiformis	Cd, Pb, Zn,	Phytoextraction	Kos et al. (2003)
Sedum alfredii	Cd, Zn	Phytoextraction	Yang et al. (2004)
Sesbania drummondii	Pb	Phytoextraction	Sahi et al. (2002)
Sinapis alba	Cd, Pb, Zn,	Phytoextraction	Kos et al. (2003)
Thlaspi caerulescens	Zn	Phytoextraction	Baker and Walker (1990); Brooks (1998)
Thlaspi caerulescens	Ni	Phytoextraction	Baker et al. (1991)
Thlaspi caerulescens	Cd	Phytoextraction	Brown et al. (1995)
Typha latifolia L.	Se	Phytovolatilization	Pilon-Smits et al. (1999)
Zea mays	Cd, Zn	Phytoextraction	Wang et al. (2002)
Zea mays	Cr	Phytoextraction	Ciura et al. (2005)

Brassica juncea, *Medicago sativa*, *Cucurbita pepo*, *Brassica napus* and *Zea mays* (Table 5.1), which are reported to accumulate many toxic metals. Agronomic crops can achieve dual purpose of treating contaminated sites alongwith farm produce and demonstration of application of phytoremediation. Identification and selection of more efficient plant varieties for phytoremediation, optimized doses of soil amendments and agronomic practices and co-ordination with developments in environmental and agricultural engineering can increase the efficiency of the phytoremediation (Salt et al., 1998). More fundamental research is required to better exploit the metabolic diversity of the plants for phytoremediation and to understand the complex interactions between metals, soil, plant roots and rhizosphere microorganisms. However major disadvantage of this technology is in requiring longer period for remediation. Well-designed and well-documented demonstration experiments and field trials are needed for public acceptance and policy makers to promote the use of agronomic crops for phytoremediation of toxic heavy metals.

REFERENCES

Adler, T., 1996. Botanical cleanup crews. Sci. News 150, 42–43.

Agostini, E., Coniglio, M.S., Milrad, S.R., Tigier, H.A., Giulietti, A.M., 2003. Phytoremediation of 2,4-dichlorophenol by *Brassica napus* hairy root cultures. Biotechnol. Appl. Biochem. 37, 139–144.

Alkorta, I., Allica, J.H., Becerril, J.M., Amezaga, I., Albizu, I., Onaindia, M., Garbisu, C., 2004. Chelate-enhanced phytoremediation of soils polluted with heavy metals. Rev. Environ. Sci. Biotechnol. 3, 55–70.

Anderson, C.W.N., Brooks, R.R., Stewart, R.B., Simcock, R., 1998. Harvesting a crop of gold in plants. Nature 395, 553–554.

Badri, M., Springuel, I., 1994. Biogeochemical prospecting in the south-easter desert of Egypt. J. Arid Environ. 28, 257–264.

Baker, A.J.M., McGrath, S.P., Sidoli, C.M.D., Reeves, R.D., 1994. The possibility of in situ heavy metal decontamination of polluted soils using crops of metal accumulating plants. Resour. Conserv. Recycl. 11, 41–49.

Baker, A.J.M., Reeves, R.D., McGrath, S.P., 1991. In situ decontamination of heavy metal polluted soils using crops of metal-accumulating plants – a feasibility study. In: Hinchee, R.E., Olfenbuttel, R.F. (Eds.), In situ Bioreclamation. Butterworth-Heinemann, Boston, pp. 600–605.

Baker, A.J.M., Walker, P.L., 1990. Ecophysiology of metal uptake by tolerant plants. In: Shaw, A.J. (Ed.), Heavy Metal Tolerance in Plants: Evolutionary Aspects. CRC Press, Boca-Raton, FL, pp. 155–177.

Banuelos, G.S., Ajwa, H.A., Mackey, L.L., Wu, C., Cook, S., Akohoue, S., 1997b. Evaluation of different plant species used for phytoremediation of high soil selenium. Environ. Qual. 26, 639–646.

Banuelos, G. S., Ajwa, H. A., Terry, N., Downey, S., 1997a. Phytoremediation of Selenium laden effluent. Fourth International in situ and on-site bioremediation symposium, April 28–May 1, New Orleans, LA. 3:303.

Baumann, A., 1885. Das Verhalten von Zinksatzen gegen Pflanzen und im Boden. Landwirtsch. Vers.-Statn. 31, 1–53.

Bennet, L.E., Burkhead, J.L., Hale, K.L., Terry, N., Pilon, M., Pilon-Smits, E.A.H., 2003. Analysis of transgenic Indian mustard plants for phytoremediation of metal-contaminated mine tailings. J. Environ. Qual. 32, 432–440.

Berti, W.R., Cunningham, S.D., 2000. Phytostabilization of metals. In: Raskin, I., Ensley, B.D. (Eds.), Phytoremediation of Toxic Metals: Using Plants to Clean up the Environment. Wiley, New York, pp. 71–88.

Blaylock, M.J., Huang, J.W., 2000. Phytoextraction of metals. In: Raskin, I., Ensley, B.D. (Eds.), Phytoremediation of Toxic Metals: Using Plants to Clean up the Environment. Wiley, New York, pp. 53–70.

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

Boominathan, R., Saha-Chaudhury, N.M., Sahajwalla, V., Doran, P.M., 2004. Production of nickel bio-ore from hyperaccumulator plant biomass: Applications in phytomining. Biotechnol. Bioeng. 86, 243–250.

Broadhurst, C.L., Chaney, R.L., Angle, J.S., Maugel, T.K., Erbe, E.F., Murphy, C.A., 2004. Simultaneous hyperaccumulation of nickel, manganese, and calcium in *Alyssum* leaf trichomes. Environ. Sci. Technol. 38, 5797–5802.

Brooks, R.R., 1977. Copper and cobalt uptake by *Haumaniastrum* species. Plant Soil 48, 541–544.

Brooks, R.R., 1983. Biological methods of prospecting for minerals. Wiley-Interscience, New York, p. 313.

Brooks, R.R., 1998. Plants that hyperaccumulate heavy metals. Wallingford CAB International, p. 380.

Brown, S.L., Chaney, R.L., Angle, J.S., Baker, A.J.M., 1995. Zinc and cadmium uptake by hyperaccumulator *Thlaspi caerulescens* and metal tolerant *Silene vulgaris* grown on sludge amended soils. Environ. Sci. Technol. 29, 1581–1585.

Burken, J.G., Schnoor, J.L., 1997. Uptake and metabolism of atrazine by poplar trees. Environ. Sci. Technol. 31, 1399–1406.

Chaney, R.L., 1983. Plant uptake of inorganic waste constitutes. In: Parr, J.F., Marsh, P.B., Kla, J.M. (Eds.), Land treatment of hazardous wastes. Noyes Data Corp., Park Ridge, NJ, pp. 50–76.

Chaney, R.L., Malik, K.M., Li, Y.M., Brown, S.L., Brewer, E.P., Angle, J.S., 1997. Phytoremediation of soil metals. Curr. Opin. Biotechnol. 8, 279–284.

Chaudhry, T.M., Hayes, W.J., Khan, A.G., Khoo, C.S., 1998. Phytoremediation-Focusing on accumulator plants that remediate metal-contaminated soils. Aust. J. Ecotoxicol. 4, 37–51.

Citterio, S., Santagostino, A., Fumagalli, P., Prato, N., Ranalli, P., Sgorbati, S., 2003. Heavy metal tolerance and accumulation of Cd, Cr and Ni by *Cannabis sativa* L. Plant Soil 256, 243–252.

Ciura, J., Poniedzialek, M., Sekara, A., Jedrszczyk, E., 2005. The possibility of using crops as metal phytoremediants. Pol. J. Environ. Stud. 14, 17–22.

Comis, D., 1996. Green remediation: Using plants to clean the soil. J. Soil Water Conserv. 51, 184–187.

Creman, H., Velikonja-Bolta, S., Vodnik, D., Kos, B., Lestan, D., 2001. EDTA enhanced heavy metal phytoextraction: Metal accumulation, leaching and toxicity. Plant Soil 235, 105–114.

Cunningham, S.D., Berti, W.R., Huang, J.W., 1995. Phytoremediation of contaminated soils. Trends Biotechnol. 13, 393–397.

Cunningham, S.D., Ow, D.W., 1996. Promises and prospects of phytoremediation. Plant Physiol. 110, 715–719.

Dierberg, F.E., Debusk, T.A., Goulet, J.R., 1987. Removal of copper and lead using a thin-film technique. In: Reddy, K.B., Smith, W.H. (Eds.), Aquatic Plants for Water Treatment and Resource Recovery. Mangolia Publisher, pp. 497–504.

Dirilgen, N., 1998. Effects of pH and chelator EDTA on Cr toxicity and accumulation in *Lemna minor*. Chemosphere 37, 771–783.

Dushenkov, S., Kapulnik, Y., 2000. Phytofiltration of metals. In: Raskin, I., Ensley, B.D. (Eds.), Phytoremediation of Toxic Metals: Using Plants to Clean up the Environment. Wiley, New York, pp. 89–106.

Dushenkov, V., Kumar, P.B.A.N., Motto, H., Raskin, I., 1995. Rhizofiltration: The use of plants to remove heavy metals from aqueous streams. Environ. Sci. Technol. 29, 1239–1245.

Eapen, S., Suseelan, K.N., Tivarekar, S., Kotwal, S.A., Mitra, R., 2003. Potential for rhizofiltration of uranium using hairy root cultures of *Brassica juncea* and *Chenopodium amaranticolor*. Environ. Res. 91, 127–133.

Ebbs, S., Brady, D., Kochian, L., 1998. Role of uranium speciation in the uptake and translocation of uranium by plants. J. Exp. Bot. 49, 1183–1190.

Ebbs, S.D., Lasat, M.M., Brandy, D.J., Cornish, J., Gordon, R., Kochian, L.V., 1997. Heavy metals in the environment: Phytoextraction of cadmium and zinc from a contaminated soil. J. Environ. Qual. 26, 1424–1430.

EPA, 1997. Electrokinetic laboratory and field processes applicable to radioactive and hazardous mixed waste in soil and groundwater. EPA/402/R-97/006. Environmental Protection Agency, Washington, DC.

EPA, 2000. Introduction to phytoremediation. EPA/600/R-99/107. Environmental Protection Agency, Washington, DC.

Ernst, W.H.O., 1996. Bioavailability of heavy metals and decontamination of soils by plants. Appl. Geochem. 11, 163–167.

Evans, G.M., Furlong, J.C., 2003. Environmental Biotechnology: Theory and applications. Wiley, West Sussex, UK.

Flathman, P.E., Lanza, G.R., 1998. Phytoremediation: Current views on an emerging green technology. J. Soil Contam. 7, 415–432.

Garbisu, C., Hernandez-Allica, J., Barrutia, O., Alkorta, I., Becerril, J.M., 2002. Phytoremediation: A technology using green plants to remove contaminants from polluted areas. Rev. Environ. Health 17, 75–90.

Gardea-Torresdey, J.L., Parsons, J.G., Gomez, E., Peralta-Videa, J., Troiani, H.E., Santiago, P., Jose, Y.M., 2002. Formation and growth of Au nanoparticles inside live alfalfa plants. Nano Letters 2, 397–401.

Gardea-Torresdey, J.L., Peralta-Videa, J.R., Rosa, G., Parsons, J.G., 2005a. Phytoremediation of heavy metals and study of the metal coordination by X-ray absorption spectroscopy. Coordin. Chem. Rev. 249, 1797–1810.

Gardea-Torresdey, J.L., Rodriguez, E., Parsons, J.G., Peralta-Videa, J.R., Meitzner, G., Cruz-Jimenez, G., 2005b. Use of ICP and XAS to determine the enhancement of gold phytoextraction by *Chilopsis linearis* using thiocyanate as a complexing agent. Anal. Bioanal. Chem. 382, 347–352.

Geebelen, W., Vangronsveld, J., Adriano, D.C., Van Poucke, L.C., Clijsters, H., 2002. Effects of Pb-EDTA and EDTA on oxidative stress reactions and mineral uptake in *Phaseolus vulgaris*. Plant Physiol. 115, 377–384.

Glass, D.J., 2000. Economic potential of phytoremediation. In: Raskin, I., Ensley, B.D. (Eds.), Phytoremediation of Toxic Metals: Using Plants to Clean up the Environment. Wiley, New York, pp. 15–32.

Grant, C.A., Bailey, L.D., 1997. Effects of phosphorus and zinc fertilizer management on cadmium accumulation in flaxseed. J. Sci. Food Agric. 73, 307–314.

Griga, M., Bjelkova, M., Tejklova, E., 2003. Potential of flax (*Linum usitatissimum*) for heavy metal extraction and industrial processing of contaminated biomass – a review. In: Mench, M., Mocquot, B. (Eds.), Risk Assessment and Sustainable Land Management Using Plants in Trace Element-Contaminated Soils. COST Action 837, 4th WG2 Workshop, Bordeaux 2002. INRA Centre Bordeaux-Aquitaine, Villenave d, Ornon, pp. 173–179.

Hartman, W. J., Jr., 1975. An evaluation of land treatment of municipal wastewater and physical siting of facility installations. US Department of Army, Washington, DC.

Heaton, A.C.P., Rugh, C.L., Wang, N., Meagher, R.B., 1998. Phytoremediation of mercury and methylmercury – polluted soils using genetically engineered plants. Soil Contamin. 7, 497–510.

Huang, J., Blaylock, M., Kapulnik, Y., Ensley, B., 1998. Phytoremediation of uranium contaminated soils: Role of organic acids in triggering uranium hyperaccumulation in plants. Environ. Sci. Technol. 32, 2004–2008.

Huang, J.W., Chen, R., Berti, W.R., Cunningham, S.D., 1997. Phytoremediation of lead contaminated soils: Role of synthetic chelators in lead phytoextraction. Environ. Sci. Technol. 31, 800–805.

Jaj, N., 2005. Heavy metals and pathogens in leafy vegetables. M.Sc. Thesis, TIET, Patiala, India.

Kay, S.H., Haller, W.T., Garrard, L.A., 1984. Effect of heavy metals on water hyacinth [*Eichhornia crassipes* (Mart.) Solms]. Aqua. Toxicol. 5, 117–128.

Keeling, S.M., Stewart, R.B., Anderson, C.W., Robinson, B.H., 2003. Nickel and cobalt phytoextraction by the hyperaccumulator *Berkheya coddii*: Implications for polymetallic phytomining and phytoremediation. Int. J. Phytoremediat. 5, 235–244.

Kos, B., Greman, H., Lestan, D., 2003. Phytoextraction of lead, zinc and cadmium from soil by selected plants. Plant Soil Environ. 49, 548–553.

Kozlov, M.V., Haukioja, E., Bakhtiarov, A.V., Stroganov, D.N., 1995. Heavy metals in Birch leaves around a nickel–copper smelter at Monchegorsk, north western Russia. Environ. Pollut. 90, 291–299.

Kumar, P.B.A.N., Dushenkov, V., Motto, H., Raskin, I., 1995a. Phytoextraction: The use of plants to remove heavy metals from soils. Environ. Sci. Technol. 29, 1232–1238.

Kumar, P.B.A.N., Motto, H., Raskin, I., 1995b. Rhizofiltration: The use of plants to remove heavy metals from aqueous streams. Environ. Sci. Technol. 29, 1239–1245.

LaCoste, C., Robinson, B., Brooks, R., 2001. Uptake of thallium by vegetables: Its significance for human health, phytoremediation and phytomining. J. Plant Nutr. 24, 1205–1215.

Landberg, T., Greger, M., 1994. Can heavy metal tolerant clones of Salix be used as vegetation filters on heavy metal contaminated land? In: Aronsson, P., Perttu, K. (Eds.), *Willow* Vegetation Filters for Municipal Wastewaters and Sludges. A Biological Purification System. Report 50, Swedish University of Agricultural Science, Uppsala, pp. 133–144.

Landberg, T., Greger, M., 1996. Differences in uptake and tolerance to heavy metals in *Salix* from unpolluted and polluted areas. Appl. Geochem. 11, 175–180.

Lasat, M.M., 2000. Phytoextraction of metals from contaminated soil: A review of plant/soil/metal interaction and assessment of pertinent agronomic issues. J. Hazar. Subst. Res. 2, 5–25.

Lasat, M.M., 2002. Phytoextraction of toxic metals: A review of biological mechanisms. J. Environ. Qual. 31, 109–120.

Lewis, B.G., Johnson, C.M., Delwiche, C.C., 1966. Release of volatile selenium compounds by plants. Collection procedures and preliminary observations. J. Agric. Food Chem. 14, 638–640.

Lopez, M.L., Peralta-Videa, J.R., Benitez, T., Gardea-Torresdey, J.L., 2005. Enhancement of lead uptake by alfalfa (*Medicago sativa*) using EDTA and a plant growth promoter. Chemosphere 61, 595–598.

Ma, L., Komar, K., Tu, C., Zhang, W., Cai, Y., Kennelley, E., 2001. A fern that hyperaccumulates arsenic. Nature 409, 579.

Maurice, C., Lagerkvist, A., 2000. Using *Betula pendula* and *Telephora caryophyllea* for soil pollution assessment. J. Soil Contam. 9, 31–50.

McCutcheon, S.C., Schnoor, J.L., 2003. Overview of phytotransformation and control of wastes. In: McCutcheon, S.C., Schnoor, J.L. (Eds.), Phytoremediation-Transformation and Control of Contaminants. Wiley, Hoboken, NJ, pp. 3–58.

McGrath, S., Sidoli, C., Baker, A.J.M., Reeves, R., 1993. The potential for the use of metal-accumulating plants for the in situ decontamination of metal-polluted soils. In: Eijsackers, H., Hamers, T. (Eds.), Integrated Soil and Sediment Research: A Basis for Proper Protection. Kluwer, Dordrecht, pp. 673–676.

Mcinnes, B.I.A., Dunn, C., Cameron, E.M., Kameko, L., 1996. Biogeochemical exploration for gold in tropical rain forest regions of Papua New Guinea. J. Geochem. Explor. 57, 227–243.

McIntyre, T.C., 2003. Databases and protocol for plant and microorganism selection: Hydrocarbons and metals. In: McCutcheon, S.C., Schnoor, J.L. (Eds.), Phytoremediation: Transformation and Control of Contaminants. Wiley, New York, pp. 887–904.

Meagher, R.B., 2000. Phytoremediation of toxic elemental and organic pllutants. Curr. Opin. Plant Biol. 3, 153–162.

Minguzzi, C., Vergnano, O., 1948. Il contento di nichel nelli ceneri di Alyssum bertlonii Desv. Atti della Societa Toscana di Science Naturali, Mem. Ser. A 55, 49–77.

Mo, S.C., Choi, D.S., Robinson, J.W., 1989. Uptake of mercury from aqueous solution by duckweed: The effect of pH, copper and humic acid. J. Environ. Health 24, 135–146.

Newman, L.A., Strand, S.E., Choe, N., Duffy, J., Ekuan, G., 1997. Uptake and biotransformation of trichloroethylene by hybrid poplars. Environ. Sci. Technol. 31, 1062–1067.

Pawlowska, T.E., Chaney, R.L., Chin, M., Charvat, I., 2000. Effects of metal phytoextraction practices on the indigenous community of arbuscular mycorrhizal fungi at a metal contaminated landfill. Appl. Environ. Microb. 66, 2526–2530.

Peralta-Videa, J.R., Gardea-Torresdey, J.L., 2002. Effect of mixed cadmium, copper, nickel and zinc at different pHs upon alfalfa growth and heavy metal uptake. Environ. Pollut. 119, 291–301.

Pierzynski, G.M., Schnoor, J.L., Youngman, A., Licht, L., Erickson, L.E., 2002. Poplar trees for phytostabilization of abandoned zinc–lead smelter. Prac. Period. Hazar. Toxic Radioact. Waste Manage. 6, 177–183.

Pilon-Smits, E., 2005. Phytoremediation. Annu. Rev. Plant Biol. 56, 15–39.

Pilon-Smits, E., de Souza, M.P., Hong, G., Amini, A., Bravo, R.C., 1999. Selenium volatilization and accumulation by twenty aquatic plant species. J. Environ. Qual. 28, 1011–1017.

Prasad, M.N.V., Freitas, H.M.O., 2003. Metal hyperaccumulation in plants – Biodiversity prospecting for phytoremediation technology. Electron. J. biotechnol. 6, 285–321.

Pulford, I.D., Watson, C., 2003. Phytoremediation of heavy metal-contaminated land by trees – a review. Environ. Int. 29, 529–540.

Punshon, T., Dickinson, N.M., 1997. Acclimation of *Salix* to metal stress. New Phytol. 137, 303–314.

Raskin, I., Ensley, B.D., 2000. Phytoremediation of toxic metals using plants to clean up the environment. Wiley, New York, p. 316.

Raskin, I., Kumar, P.B.A.N., Dushenkov, S., Salt, D.E., 1994. Bioconcentration of heavy metals by plants. Curr. Opin. Biotech. 5, 285–290.

Reeves, R.D., Baker, A.J.M., 2000. Metal-accumulating plants. In: Raskin, I., Ensley, B.D. (Eds.), Phytoremediation of Toxic Metals: Using Plants to Clean up the Environment. Wiley, New York, pp. 193–230.

Robinson, B.H., Leblanc, M., Petit, D., Brooks, R.R., Kirkman, J.H., Gregg, P.E.H., 1998. The potential of *Thlaspi caerulescenss* for phytoremediation of contaminated soils. Plant Soil 203, 47–56.

Rugh, C.L., Gragson, G.M., Meagher, R.B., Merkle, S.A., 1998. Toxic mercury reduction and remediation using transgenic plants with a modified bacterial gene. Hortsicence 33, 618–621.

Sahi, S.V., Bryant, N.L., Sharma, N.C., Singh, S.R., 2002. Characterization of a lead hyperaccumulator shrub, Sesbania drummondii. Environ. Sci. Technol. 36, 4676–4680.

Salt, D.E., Blaylock, M., Kumar, P.B.A.N., Dushenkov, V., Ensley, B.D., Chet, I., Raskin, I., 1995. Phytoremediation: A novel strategy for the removal of toxic metals from the environment using plants. Biotechnology 13, 468–474.

Salt, D.E., Smith, R.D., Raskin, I., 1998. Phytoremediation. Annu. Rev. Plant Physiol. 49, 643–668.

Schnoor, J.L., 2000. Phytostabilization of metals using hybrid poplar trees. In: Raskin, I., Ensley, B.D. (Eds.), Phytoremediation of Toxic Metals: Using Plants to Clean up the Environment. Wiley, New York, pp. 133–150.

Smith, R.A.H., Bradshaw, A.D., 1992. Stabilization of toxic mine wastes by the use of tolerant plant populations. Trans. Inst. Mining Metall. 81, A230–A237.

Sun, B., Zhao, F.J., Lombi, E., McGrath, S.P., 2001. Leaching of heavy metals in contaminated soils using EDTA. Environ. Pollut. 113, 111–120.

Tichy, R., Fajtl, J., Kuzel, S., Kolar, L., 1997. Use of elemental sulphur to enhance a cadmium solubilization and its vegetative removal from contaminated soil. Nutr. Cycl. Agroecosys. 46, 249–255.

Turgut, C., Katie, P.M., Cutright, T.J., 2004. The effect of EDTA and citric acid on phytoremediation of Cd, Cr and Ni from soil using *Helianthus annuus*. Environ. Pollut. 131, 147–154.

USEPA, 2000. Introduction to Phytoremediation. EPA/600/R-99/107. Office of Research and Development, Washington, DC.

Utsunamyia, T., 1980. Japanese Patent Application No. 55-72959.

Vangronsveld, J., van Assche, F., Clijsters, H., 1995. Reclamation of a bare industrial area contaminated by non-ferrous metals: In situ metal immobilization. Environ. Pollut. 87, 51–59.

Vassilev, A., Schwitzguebel, J.P., Thewys, T., van der Lelie, D., Vangronsveld, J., 2004. The use of plants for remediation of metal-contaminated soils. Sci. World J. 4, 9–34.

Wang, Q., Liu, X., Cui, Y., Dong, Y., Christie, P., 2002. Responses of legume and non-legume crop species to heavy metals in soils with multiple metal contamination. J. Environ. Sci. Health A 37, 611–621.

Whiting, S.N., Neumann, P.M., Baker, A.J.M., 2003. Nickel and zinc hyperaccumulation by *Alyssum murale* and *Thlaspi caerulescens* (Brassicaceae) do not enhance survival and whole plant growth under drought stress. Plant Cell Environ. 26, 351–360.

Yang, X.E., Long, X.X., Ni, W.Z., Fu, C.X., 2002. *Sedum alfredii* H: A new Zn hyperaccumulating plant first found in China. Chin. Sci. Bull. 47, 1634–1637.

Yang, X.E., Long, X.X., Ye, H.B., He, Z.L., Calvert, D.V., Stoffella, P.J., 2004. Cadmium tolerance and hyperaccumulation in a new Zn-hyperaccumulating plant species (*Sedum alfreii* Hance). Plant Soil 259, 181–189.

Yankov, B., Delibaltova, V., Bojinov, M., 2000. Content of Cu, Zn, Cd and Pb in the vegetative organs of cotton cultivars grown in industrially polluted regions. Plant Sci. 37, 525–531.

Yankov, B., Tashin, N., 2001. Accumulation and distribution of Pb, Cu, Zn and Cd in sunflower (*Helianhus annuus* L.) grown in an industrially polluted region. Helia 24, 131–136.

Zhu, Y.L., Zayed, A.M., Quian, J.H., De Souza, M., Terry, N., 1999. Phytoaccumulation of trace elements by wetland plants: II. Water hyacinth. J. Environ. Qual. 28, 339–344.