ISSN NO. 2320-5407

Journal homepage: http://www.journalijar.com

INTERNATIONAL JOURNAL OF ADVANCED RESEARCH

RESEARCH ARTICLE

Phytoremediation : green technology to clean the environment

Mukti Gill

Department of Botany, Khalsa College for Women, Civil Lines, Ludhiana-141 001

Manuscript Info

Manuscript History:

Received: 15 June 2014
Final Accepted: 26 July 2014
Published Online: August 2014

Key words:

heavy metals, phytoremediation, phytoextraction

****Corresponding Author***

Mukti Gill

Abstract

The technology of phytoremediation is cost effective and ecologically friendly in which plant utilizes its natural abilities to restore environment. In nature there are a number of plants existing with innate mechanisms for removing heavy metals from soil, air and water as a survival strategy. Among several subsets of phytoremediation, the widely studied strategies are (a) phytoextraction (b) phytofiltration (c) phytovolatilization and (d) phytostabilization. Application of organic/inorganic chelants in soil directly affects the solubility of heavy metals and consequently increases their accumulation in plants that enhances phytoextraction. In the present review current knowledge about the phytoremediation and its techniques are discussed.

Introduction

Phytoremediation is the biotechnological application of plants to detoxify pollutants, and is an ideal and modern technique for environmental clean-up. Increased industrialization has led to the massive release of anthropogenic contaminants into the environment. Widespread pollutants include hydrocarbons, pesticides, and heavy metals (solvents and salts) showed a particular concern to the environmental issues and human health conditions. These contaminants were documented in higher amounts, low soluble in biota, highly toxic in nature, and can act as carcinogenic and mutagenic substances (Quadir et al., 2008; Malik et al., 2009; Muhammad et al., 2011). Continuous application of industrial and domestic wastewater in irrigation may cause heavy metal contamination in soils to undesirable and phyto-toxic levels. Industrial effluents logging not only destroyed the structure and fertility of soil but also causes excess loading of toxic metals from soil to plants, and at the end, heavy metals reaches human beings through food chain, causing serious health issue (Muhammad et al., 2011). Land and water pollution by heavy metals is a worldwide issue. All countries have been affected, though the area and severity of pollution vary enormously. In Western Europe, about 1,400,000 sites were affected by heavy metals of which over 300, 000 were classified as contaminated. The estimated total number of sites in Europe could be much larger as heavy metal pollution increased surprisingly in Central and Eastern Europe (Gad, 2000; McGrath et al., 2001). In the USA, there are 600,000 brown fields which are contaminated with heavy metals and need reclamation[6]. Increasing environmental pollution by heavy metal contaminants has also been reported in many developing countries (Hardy et al., 1992; Jameli et al., 2007; Quadir et al., 2008; Kausar et al., 2012).

The cleaning of heavy metal contamination from different environmental compartments, including soil, is the most difficult task, particularly on a large scale. Soil is composed of organic and inorganic solid constituents, water, and a mixture of different gases present in various proportions. The mineral components vary according to parent materials on which the soil had been developed under a particular set of climatic conditions. Therefore, soils vary enormously in physical, chemical, and biological properties. Soil water movement is controlled by physical properties, such as soil structure and texture. The soil moisture has great bearing on the controlling solute movement, salt solubility, chemical reactions, and microbiological activities and ultimately, the bioavailability of the metal ions.

Many phytoremediation technologies have been used for the remediation of polluted soils and water throughout the world. Phytoremediation is a promising technique in which different plant species are used to assimilate and detoxify metals and organic compounds. The use of plant species to treat radionuclide-contaminated soils were done in the 1950s, but the term was not introduced till the 1980s. As compared to phytoremediation, traditional techniques used for soil remediation are costly and may cause the secondary pollution. The term "phytoremediation" was coined in 1991 along with the rapid expansion in this field. Phytoremediation can be defined as "the use of green plants and their associated microorganisms, soil amendments, and agronomic techniques to remove, contain, or render harmless environmental contaminants" (Cunningham and Ow 1996). Phytoremediation is a newly evolving field of science and technology to clean up polluted soil, water, and/or air (Salt et al., 1998; Meagher et al., 2000). As compared to congenital techniques, phytoremediation is environmental friendly and is a low-cost method (Cheng et al., 2002; Malik, 2007). So, phytoremediation of heavy metals is not only an economically suitable technique, but is also important from a socioeconomic point of view (Bareen and Tahira, 2011). Various hyper accumulative plant species has been also extensively investigated that lead to the substantial progress in this field. Hyper accumulators are those plant species whose bioconcentration factor (BCF) is greater than 1,000. Accumulators are those plants whose BCF are greater than 1 but less than 1,000. Excluded are those plants whose BCF are less than 1. Bioconcentration factor indicates the efficiency of a plant in taking up heavy metals from soil and accumulating them in its tissues. It is the ratio of heavy metal concentration in the plant tissue (root, shoot, or leaves) to that in soil (Zhuang et al., 2007). It becomes clear that different mechanisms of metal accumulation, exclusion, and compartmentation exist in various plant species (Prasad 2004). Plants are susceptible to heavy metal toxicity respond by different physiological and molecular mechanisms to avoid the detrimental effects of pollutants in a variety of different ways. Toxicity depends on the type of ion, its concentration, plant species, and stage of plant growth. Tolerance and hyper accumulation of metals is based on different mechanisms such as uptake by the roots, which leads to acropetal transportation through the xylem up to the plant shoot tissues. Detoxification, chelation, degradation, transformation, and compartmentation of toxic metals further take place by cell wall binding, active transport of ions into the vacuole, and formation of complexes with organic acids or peptides. Chelation of metals by low molecular weight proteins such as metallothioneins and peptide ligands and the phytochelatins, is one of the most important mechanisms for metal detoxification in plants appears (Prasad, 2004; Memon and Schroder, 2009).

Techniques of Phytoremediation

For removal of different hazardous compounds from contaminated soil and water, plant potentials have been exploited that resulted in several technological subsets (Schwitzguebel, 2000). The following are the techniques of phytoremediation:

1) Phytoextraction: the use of pollutant-accumulating plants to remove pollutants like metal organics from soil by concentrating them in harvestable plant parts,
2) Phytotransformation: the degradation of complex organic to simple molecules or the incorporation of these molecules into plants tissues,
3) Phytostimulation: plant-assisted bioremediation or the stimulation of microbial and fungal degradation by release of exudates / enzymes into the root zone (rhizosphere),
4) Phytovolatilization: the use of plants to volatilize pollutants or metabolites,
5) Phytodegradation: enzymatic breakdown of organic pollutants such as trichloroethylene (TCE) and herbicides, both internally and externally and through secreted plant enzymes,
6) Phytorhizofiltration: The use of plant roots to ab/adsorb pollutants, mainly metals, but also organic pollutants, from water and aqueous waste streams,
7) Pump and tree (Dendroremediation): the use of trees to evaporate water and thus to extract pollutants from the soil,
8) Phytostabilization: the use of plants to reduce the mobility and bioavailability of pollutants in the environment, thus preventing their migration to groundwater or their entry into the food chain, and
9) Hydraulic Control: the control of the water and the soil field capacity by plant canopies.

Phytostabilisation

It is mostly used for the remediation of soil, sediment and sludge (Barconi et al., 2011) and depends on roots ability to limit contaminant mobility and bioavalability in the soil. Phytostabilisation can occur through the sorption, precipitation, complexaction, or metal valence reduction. The plants primary purpose is to decrease the amount of water percolating through the soil matrix, which may result in the formation of hazardous leachate and prevent soil erosion and distribution of the toxic metal to other areas. A dense root system stabilizes the soil and prevents erosion (Dalcarso et al., 2010). It is very effective when rapid immobilisation is needed to preserve ground and surface water and disposal of biomass is not required. However the major disadvantage is that, the contaminant remains in soil as it is, and therefore requires regular monitoring.

Phytoextraction

It is the best approach to remove the contamination primarily from soil and isolate it, without destroying the soil structure and fertility. It is also referred as phytoaccumulation . As the plant absorb, concentrate and precipitate toxic metals and radionuclide from contaminated soils into the biomass, it is best suited for the remediation of diffusely polluted areas, where pollutants occur only at relatively low concentration and superficially (Leyval et al., 1997). Several approaches have been used but the two basic strategies of phytoextraction, which have finally developed are; i) Chelate assisted phytoextraction or induced phytoextraction, in which artificial chelates are added to increase the mobility and uptake of metal contaminant. ii) Continuous phytoextraction in this the removal of metal depends on the natural ability of the plant to remediate; only the number of plant growth repetitions are controlled (Cobbett, 2000; Hall, 2002). Discovery of Ghosh and Singh.: A review on phytoremediation of heavy metals and utilization of its byproducts by hyperaccumulator species has further boosted this technology. In order to make this technology feasible, the plants must, extract large concentrations of heavy metals into their roots, translocate the heavy metals to surface biomass, and produce a large quantity of plant biomass. The removed heavy metal can be recycled from the contaminated plant biomass (Yang etal., 2005). Factors such as growth rate, element selectivity, resistance to disease, method of harvesting, are also important (Clemens, 2006;Yadav, 2010). However slow growth, shallow root system, small biomass production, final disposal limit the use of hyperaccumulator species (Seth et al., 2012).

Phytovolatilization

Phytovolatilization involves the use of plants to take up contaminants from the soil, transforming them into volatile form and transpiring them into the atmosphere. Phytovolatilization occurs when growing trees and other plants take up water and the organic and inorganic contaminants. Some of these contaminants can pass through the plants to the leaves and volatilise into the atmosphere at comparatively low concentrations (Barconi et al. 2011). Phytovolatilization has been primarily used for the removal of mercury, the mercuric ion is transformed into less toxic elemental mercury. The disadvantage is, mercury released into the atmosphere is likely to be recycled by precipitation and then redeposit back into ecosystem. Gary Banuelos of USDS's Agricultural Research Service have found that some plants grow in high Selenium media produce volatile selenium in the form of dimethylselenide and dimethyldiselenide (Dietz et al., 1999). Phytovolatilization has been successful in tritium (3H), a radioactive isotope of hydrogen, it is decayed to stable helium with a half-life of about 12 years (Schutzendubel and Polle, 2002).

Phytodegradation

In phytoremediation of organics, plant metabolism contributes to the contaminant reduction by transformation, break down, stabilisation or volatilising contaminant compounds from soil and groundwater. Phytodegradation is the breakdown of organics, taken up by the plant to simpler molecules that are incorporated into the plant tissues. Plants contain enzymes that can breakdown and convert ammunition wastes, chlorinated solvents such as trichloroethylene and other herbicides. The enzymes are usually dehalogenases, oxygenases and reductases (Nieboer and Richardson, 1980). Rhizodegradation is the breakdown of organics in the soil through microbial activity of the root zone (rhizosphere) and is a much slower process than phytodegradation. Yeast, fungi, bacteria and other microrganisms consume and digest organic substances like fuels and solvents. All phytoremediation technologies are not exclusive and may be used simultaneously, but the metal extraction depends on its bio available fraction in soil. The advantages and disadvantages have been discussed in Table 1.

Table 1. Advantages and disadvantages of phytoremediation.

S.No.	Advantages	Disadvantages
1.	Amendable to a variety of organic and inorganic compounds.	Restricted to sites with shallow contamination within rooting zone of remediative plants.
2.	*In Situ* / *Ex Situ* Application possible with effluent/soil substrate respectively.	May take up to several years to remediate a contaminated site.

3.	*In Situ* applications decrease the amount of soil disturbance compared to conventional methods.	Restricted to sites with low contaminant concentrations.
4.	Reduces the amount of waste to be landfilled (up to 95%), can be further utilized as bio-ore of heavy metals.	Harvested plant biomass from phytoextraction may be classified as a hazardous waste hence disposal should be proper.
5.	*In Situ* applications decrease spread of contaminant *via* air and water.	Climatic conditions are a limiting factor.
6.	Does not require expensive equipment or highly specialized personnel.	Introduction of nonnative species may affect biodiversity
7.	In large scale applications the potential energy stored can be utilized to generate thermal energy.	Consumption/utilization of contaminated plant biomass is a cause of concern.

Chelant assisted phytoextraction

The term 'chelate' denotes a complex between metal and a chelating agent and not the chelating agent itself (Nawack and Vanbriesen, 2005). A shorter word for chelating agent is 'chelant' or 'chelator'. It is therefore, suggested for using the term 'chelant-enhanced phytoextraction'. Other terms such as 'chelant-induced' and 'chelant-assisted' phytoextraction can be used as synonyms to chelant-enhanced phytoextraction. 'Chelate' should be used whenever a metal-chelating agent complex is meant, e.g., when talking about a specific complex in soil physical, chemical and biological properties. A large number but only a fraction of metals are readily available / bioavailability for transporting to the roots (Lasat, 2002). To resolve this problem, chemically enhanced phytoextraction has been developed (Huang et al., 1997). This approach utilizes high biomass crops that are induced to absorb large quantity of metals whereas metals mobility is enhanced by the treatment of different chemicals. Research into the interaction of plants with chelating agents started in the 1950s with a view to alleviating deficiencies in the essential nutrient metals Fe, Mn, Cu and Zn. Initial results also showed that chelants such as EDTA enhanced plant uptake of Pb and Hg (Hale and Wallace, 1970). Addition of chelating agents to such an extent that they might be used for cleanup plants of Pb-contaminted soils (Jorgensen, 1993; Huang and Cunninghan, 1996). Enhanced uptake was not only observed in nutrient solution and pot experiments but also in the field (Liphadzi et al., 2003). Mainly, there are three factors that control the transportation of heavy metals from soil to plants. These include the total amount of bioavailable metals / elements (quantity factor), the activity and the ratio of elements present in soil as in ionic form (intensity factor) and the rate of elements transfered from solid to liquid phases to plant roots (reaction kinetics) (Brummer et al., 1986).

I. Using organic chelants

Selection of suitable chelants for the extraction of heavy metals from a polluted site is the first issue to be considered, whereas the solubilization of heavy metals must be enhanced to increase extraction efficiency which is mainly based on the capacity of organic chelants to form water soluble organic complexes (Martel and Calvin, 1958). By the formation of complex, metals get extracted or desorbed from different components of soil. Several chelants for example Citric acid, EDTA, CDTA, DTPA, EGTA, EDDHA and NTA were studied to find out their ability of mobilizing metals and increasing metal accumulation in various plant species (Cooper et al., 1999). Different metals were focused like Pb (Blaylock et al., 1997), U, and Au (Huang et al., 1998). The complexation of heavy metals with various chelants in soil is as follows: EDTA and related synthetic chelates > nitrilotriacetic acid (NTA) > citric acid > oxalic acid > acetic acid (Hong and Pintauro, 1996; Krishnamurti et al., 1998; Wenger et al., 1998). In most of the designed experiments Pb is targeted to test the effect of applying organic chelants on the accumulation of heavy metals. The experiments were conducted with chelants of high metals binding capacity (like EDTA hydroxyethylethylenediaminetriacetic acid [HEDTA], 1,2-cyclohexylenedinitrilotetraacetic acid [CDTA], and diethylenetriaminepentaacetic acid [DTPA]). Research was conducted at highly Pb polluted soils where addition of high amount of EDTA, CDTA, or HEDTA increased Pb concentration tremendously to as much as 24g Pb Kg^{-1} dry matter. In smaller extent, artificial chelants also enhance heavy metals concentration other than Pb in the soil solution and in the biomass of several plants. These discoveries paved the way to successful Pb phytoextraction and defining development of methods to remove other toxic metals using suitable chelants. Likewise, EGTA (ethylenebis [oxyethylenetrintrilo] tetraacetic acid) has been shown to have a high affinity for Cd^{2+}, but not for Zn^{2+}. EDTA, HEDTA, and DTPA are selective for Zn^{2+}. EDTA, citric and oxalic acid increase (> 200-fold) Cr^{3+} uptake and its concentration in plant roots and shoots from a polluted soil . Acto-aminodiacetic acid stimulates Pb bioavailability whereas S-carboxy-methylcysteine is effective for Cu. In treated soil with ammonium thiocyanate, Indian mustard accumulated Au upto 57 mg/ kg. On the other hand, *Iberis intermedia* and *Biscutta lovevigata* accumulated 0.4% and 1.5% thallium on a dry weight basis, respectively. The addition of citric acid and its salts to the soil, increases uranium mobility and its uptake by the plant (Huang et al., 1998; Ebbs et al., 1998). They further suggested that the strong mobilization of uranium caused by citric acid is because of citrate-uranayl complex formation rather than decreased pH. This indicates the affinity of the chelant for the target metal. Therefore, to increase efficiency of phytoextraction, synthetic chelants with high affinity for the metal of interest should be used. In the light of aforementioned information, a hypothetical protocol for the chelant assisted phytoextraction for a contaminated area is provided as (Salt et al., 1998) : 1.Evaluation of site and determination of suitable chelant / crop combination, 2.Preparation of site and cultivation of selected crop / plant, 3.When potential bio mass is produced suitable metal chelant is applied, and 4.Plant / crop is harvested after a short metal-accumulation phase (several days or weeks). Moreover, phytoextraction could be continued by replanting on the site, depending on the crop and the season. Estimates suggest that remediation of sites contaminated with up to 2500 mg kg^{-1} Pb is possible in less than 10 years. The weight and volume of contaminated material can be further reduced by ashing or composting. Depending on economical feasibility plant residues, enriched with metals could be utilized for metal recovery. Along with the multifaceted benefits this strategy have potential risk of metal leaching to the ground water and there is still a lack of detailed studies regarding the persistence of metal chelating agent complexes in contaminated soil (Lombi et al., 2001). In addition to the risk of metal leaching, EDTA is an expensive chemical. Little discussion on the potential cost of EDTA induced phytoextraction occurred, but this issue seriously detracts from the feasibility of that technology (Chaney et al., 2007). The price of commercial quantities of EDTA and estimated the cost would be about \$30000 ha^{-1} for the amount of EDTA reportedly needed to attain over 10 kg Pb kg^{-1} dry shoots (10 mmol EDTA kg^{-1} soil for each cropping (Chanet et al., 2007).

II. Using inorganic chelants

Enhancement of phytoextraction through inorganic amendments has a different solubilization mechanism. In this strategy, instead of complex formation, the solubilization of heavy metals relies on disruption through inorganic chelants like Sulphur, Ammonium sulphate and Chloride salt (Gray et al.,1999). Metals in solubilized form are potentially bioavailable and can either be absorbed by plants, leached into the ground water, or desorbed again by the exchange sites of the soil.

Conclusions

Heavy metals are persistent environmental pollutants which cannot be degraded and require complete removal for remedial purpose. Plants are exploited to rehabilitate the contaminated environment by the scientists of different inter-related fields. This resulted in a green technology called Phytoremediation. This fast emerging, innovative technology is a cost effective, eco-friendly and viable alternative to conventional remedial methods. At the same time, it is most suitable for a developing country like Pakistan. Enhanced phytoextraction is the important aspect for the modification and implementation of phytoremediation strategies because when accumulation rate of heavy metal in plants increases, the removal of pollutants also maximizes. It is also very crucial to minimize the ecological risk linked with enhanced phytoextraction. Further research is required to optimize the ecological and economical efficiencies of Phytoremediation.

References

Barconi, D., Bernardini, G. and Santucci, A. (2011): Linking protein oxidation to environmental pollutants: redox proteome approaches. Journal of Proteomics 2011; 74(11):2324– 2337.

Bareen, F. and Tahira, S.A. (2011): Metal accumulation potential of wild plants in tannery effluent contaminated soil of Kasur, Pakistan: field trials for toxic metal cleanup using Suaeda fruticosa. J. Hazard. Matter. 186(1): 443–450.

Blaylock, M.J., Salt, D.E., Dushenkov, S., Zakharova, O., Gussman, C., Kapulnic, Y., Ensley, B.D., and Raskin, I. (1997): Enhanced accumulation of Pb in Indian mustard by soil-applied chelating agents. Environmental Science and Technology 31: 860-865.

Brümmer, G., Gerth, J. and Herms, U. (1986): Heavy metal species, mobility and availability in soils. Z. Pflanzenernaehr.Bodenkd 149:382–398.

Chaney, R.L., Angle, J.S., Broadhurst, C.L., Peters, C.A., Tappero, R.V. and Sparks, D.L. (2007): Improved understanding of hyperaccumulation yields commercial phytoextraction and phytomining technologies. J. Environ. Quality 36:1429-1443.

Cheng, S., Grosse, W., Karrenbrock, F. and Thoennessen, M. (2002): Efficiency of constructed wetlands in decontamination of water polluted by heavy metals. Ecol. Eng. 18(3):317–325.

Clemens, S. (2006): Toxic metal accumulation, responses to exposure and mechanisms of tolerance in plants. Biochimie 88(11): 1707–1719.

Cobbett, C.S. (2000): Phytochelatins and their roles in heavy metal detoxification Plant Physiology 123(3): 825–832.

Cooper, E.M., Sims, J.T., Cunningham, S.D., Huang, J.W. and Berti, W.R. (1999): Chelate-assisted Phytoextraction of lead from contaminated soils. J. Environmental Quality 28: 1709-19.

Cunningham, S.D. and Ow, D.W. (1996): Promises and prospects of phytoremediation-update on biotechnology. Plant Physiol. 110: 715–719.

Dalcorso, G., Farinati, S. and Furini, A. (2010): Regulatory networks of cadmium stress in plants. Plant Signaling and Behavior 5(6):1–5.

Dietz, K.J., Bair, M. and Kramer, U. (1999): Free radical and reactive oxygen species as mediators of heavymetal toxicity in plants.In: Prasad MNV, Hagemeyer J, editors, Heavy Metal Stress in Plants from Molecules to Ecosystems, Spinger- Verlag, Berlin, Germany p. 73–79.

Ebbs, S.E., Norvell, W.A. and Kochian, L.V. (1998): The effect of acidification and chelating agents on the solublisation of uranium from contaminated soil. J. Environ. Qual. 27:1486–1494.

Gade, L. H. (2000): Highly polar metal–metal bonds in “early–late” Heterodimetallic complexes. Angew. Chem. Int. Ed. 39(15):2658–2678.

Gray, C.W., McLaren, R.G., Roberts, A.H.C. and Condron, L.M. (1999): Solubility, sorption and desorption of native added cadmium in relation to properties of soils in New Zealand. Eur. J. Soil Sci. 50:127–137.

Hale, V.Q. and Wallace, A. (1970): Effect of chelates on uptake of some heavy metal radionuclide from oils by bush beans. Soil Sci. 109: 262-263.

Hall, J.L. (2002): Cellular mechanisms for heavy metal detoxification and tolerance. Journal of Experimental Botany 53(366):1–11.

Hardoy, J.E., Mitlin, D. and Satterthwaite, D. (1992): Environmental problems in third world cities. Earthscan, London.

Hong, J. and Pintauro, P.N. (1996): Desorption– complexation–dissolution characteristics of absorbed cadmium from kaolin by chelators. Water Air Soil Pollut. 86:35–50.

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

Huang, J.W., Blaylock, M.J., Kapulnik, Y. and Ensley, B.D. (1998): Phytoremediation of uranium-contaminated soils: Role of organic acids in triggering uranium hyperaccumulation in plants'. Environ Sci Technol 32: 2004–08.

Huang, J.W.W., Chen, 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.

Jamali, M.K., Kazi, T.G., Arain, M.B., Afridi, H.I., Jalbani, N., Memon, R.A. and Shah, A. (2007): Heavy metals from soil and domestic sewage sludge and their transfer to sorghum plants. Environ. Chem. Lett. 5:209–218.

Jorgensen, S.E. (1993): Removal of heavy metals from compost and soil by ecotechnological methods. Ecol. Eng. 2: 89-100.

Kausar, S., Mahmood, Q., Raja, I.A., Khan, A., Sultan, S., Gilani, M.A. and Shujaat, S. (2012): Potential of Arundo donax to treat chromium contamination. Ecol. Eng. 42:256–259.

Krishnamurti, G.S.R., Cielinski, G., Huang, P.M., and VanRees, K.C.J. (1998): Kinetics of cadmium release from soils as influenced by organic acids: Implementation in cadmium availability. J. Environ. Qual. 26: 271–277.

Lasat, M.M. (2002): Phytoextraction of metals from contaminated soil: A view of plant/ soil/metal interaction and assessment of pertinent agronomic issues'. J. Hazardous substance Res. 2:1-25.

Leyval. C., Turnau, K., Haselwandter, K. (1997): Effect of heavy metal pollution on mycorrhizal colonization and function:physiological, ecological and applied aspects. Mycorrhiza 7(3): 139–153.

Liphadzi, M.S., Kirkham, M.B., Mankin, K.R. and Paulsen, G.M. (2003): EDTA-assisted heavy-metal uptake by poplar and sunflower grown at a long-term sewage-sludge farm. Plant Soil 257:171-182.

Lombi, E., Zhao, F., Dunham, S. and McGrath, S. (2001): Phytoremediation of heavy metal-contaminated soils. J. Environ. Qual. 30(6):1919–1926.

Lone, M.I., He, Z.L., Hs, P. and Xiaoe, Y. (2008): Phytoremediation of heavy metal polluted soils and water: progress and perspectives. J. Zhejiung Uni. Sci. 9(3):210–220.

Malik, A. (2007): Environmental challenge vis a vis opportunity: the case of water hyacinth. Environ. Int. 33(1):122–138.

Malik, A.H., Khan, Z.M., Mahmood, Q., Nasreen, S. and Bhatti, Z. A. (2009): Perspectives of low cost arsenic remediation of drinking water in Pakistan and other countries. J. Hazard Mater. 168(1):1–12.

Martell, A.E. and Calvin, M. (1958). The chemistry of metal-chelates. (In German.) Verlag Chemie, Weinheim, Germany.

McGrath, S., Zhao, F. and Lombi, E. (2001): Plant and rhizosphere processes involved in phytoremediation of metal contaminated soils. Plant Soil. 232(1–2):207–214.

Meagher, R., Rugh, C., Kandasamy, M., Gragson, G. and Wang, N. (2000): Engineered phytoremediation of mercury pollution in soil and water using bacterial genes. Phytoremediation of Contaminated Soil and Water. Lewis Publishers, Boca Raton, FL 201–219.

Memon, A.R. and Schröder, P. (2009): Implications of metal accumulation mechanisms to phytoremediation. Environ. Sci. Pollut. Res. 16(2): 162–175.

Muhammad, S., Shah, M.T, Khan, S. (2011): Heavy metal concentrations in soil and wild plants growing around Pb–Zn sulfide terrain in the Kohistan region, northern Pakistan. Microchem. J. 99(1):67–75.

Nieboer, E., and Richardson, D.H.S. (1980): The replacement of the nondescript term % heavymetals’ by a biologically and chemically significant classification of metal ions. Environmental Pollution Series B: Chemical and Physical 1(1): 3–26.

Nowack, B. and VanBriesen, J. M. (2005): Chelating agents in the environment. In Biogeochemistry of Chelating Agents; ACS Symposium Series; American Chemical Society, Washington, DC, 910: 1-18.

Prasad, M. (2004): Phytoremediation of metals in the environment for sustainable development. Proc Indian Natl. Sci. Acad. 70(1):71–98.

Qadir, A., Malik, R.N. and Husain, S.Z. (2008): Spatio-temporal variations in water quality of Nullah Aik-tributary of the river Chenab, Pakistan. Environ. Monit. Asses. 140(1–3):43–59.

Salt, D.E., Smith, R. and Raskin, I. (1998): Phytoremediation. Annu. Rev. Plant Biol. 49(1):643–668.

Schutzendubel, A. and Polle, A. (2002): Plant responses to abiotic stresses: heavy metal-induced oxidative stress and protection by mycorrhization. J. Experimental Bot. 53(372):1351–1365.

Schwitzguebel,, J. (2000): Potential of phytoremediation, an emerging green technology. Ecosystem service and sustainable watershed management in Science 9(3): 210-220.

Seth, C.S, Remans, T. and Keunen, E. (2012): Phytoextraction of toxic metals: a central role for glutathione. Plant, Cell and Environment 35(2):334–346.

Wenger, K., Hari, T., Gupta, M.D., Krebs, R., Rammelt, R. and Leumann, C.D. (1998): Possible approaches for in situ restoration of soils contaminated by zinc'. In:Blume H P et al, editors, Toward sustainable land use. Adv. in Geoecol. ISSS, Catena Verlag, Reiskirchen, Germany; p. 745–753.

Yadav, S.K. (2010): Heavy metals toxicity in plants: an overview on the role of glutathione and phytochelatins in heavy metal stress tolerance of plants. South African Journal of Botany 2010; 76(2):167–179.

Yang, X.E., Jin, X.F., Feng, Y. and Islam, E. (2005): Molecular mechanisms and genetic basis of heavy metal tolerance/hyperaccumulation in plants. Journal of Integrative Plant Biology 47(9): 1025–1035.

Zhuang, P., Yang, Q., Wang, H. and Shu, W. (2007): Phytoextraction of heavy metals by eight plant species in the field. Water Air Soil Pollut. 184(1–4):235–242.