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Review

# Phytoremediation of heavy metals—Concepts and applications

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## HIGHLIGHTS

- ▶ Heavy metal pollution is a serious environmental problem.
- ▶ Phytoremediation is a better option for cleanup of metal-contaminated sites.
- ▶ Phytoremediation is a green technology with good public perception.
- ▶ Research is in progress to screen plants for hyperaccumulation of heavy metals.
- ▶ Advancement in molecular studies will improve efficiency of phytoremediation.

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## ABSTRACT

The mobilization of heavy metals by man through extraction from ores and processing for different applications has led to the release of these elements into the environment. Since heavy metals are nonbiodegradable, they accumulate in the environment and subsequently contaminate the food chain. This contamination poses a risk to environmental and human health. Some heavy metals are carcinogenic, mutagenic, teratogenic and endocrine disruptors while others cause neurological and behavioral changes especially in children. Thus remediation of heavy metal pollution deserves due attention. Different physical and chemical methods used for this purpose suffer from serious limitations like high cost, intensive labor, alteration of soil properties and disturbance of soil native microflora. In contrast, phytoremediation is a better solution to the problem. Phytoremediation is the use of plants and associated soil microbes to reduce the concentrations or toxic effects of contaminants in the environments. It is a relatively recent technology and is perceived as cost-effective, efficient, novel, eco-friendly, and solar-driven technology with good public acceptance. Phytoremediation is an area of active current research. New efficient metal hyperaccumulators are being explored for applications in phytoremediation and phytomining. Molecular tools are being used to better understand the mechanisms of metal uptake, translocation, sequestration and tolerance in plants. This review article comprehensively discusses the background, concepts and future trends in phytoremediation of heavy metals.

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**Contents**



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## 1. Introduction

Environmental pollution by heavy metals has become a serious problem in the world. The mobilization of heavy metals through extraction from ores and subsequent processing for different applications has led to the release of these elements into the environment. The problem of heavy metals' pollution is becoming more and more serious with increasing industrialization and disturbance of natural biogeochemical cycles. Unlike organic substances, heavy metals are essentially nonbiodegradable and therefore accumulate in the environment. The accumulation of heavy metals in soils and waters poses a risk to the environmental and human health. These elements accumulate in the body tissues of living organisms (bioaccumulation) and their concentrations increase as they pass from lower trophic levels to higher trophic levels (a phenomenon known as biomagnification). In soil, heavy metals cause toxicological effects on soil microbes, which may lead to a decrease in their numbers and activities (Khan et al., 2010).

Regarding their role in biological systems, heavy metals are classified as essential and non-essential. Essential heavy metals are those, which are needed by living organisms in minute quantities for vital physiological and biochemical functions. Examples of essential heavy metals are Fe, Mn, Cu, Zn, and Ni (Cempel and Nikel, 2006; Göhre and Paszkowski, 2006). Non-essential heavy metals are those, which are not needed by living organisms for any physiological and biochemical functions. Examples of non-essential heavy metals are Cd, Pb, As, Hg, and Cr (Mertz, 1981; Kärenlampi et al., 2000; Suzuki et al., 2001; Cobbett, 2003; Peng et al., 2009; Sánchez-Chardi et al., 2009; Dabonne et al., 2010). Heavy metal concentrations beyond threshold limits have adverse health effects because they interfere with the normal functioning of living systems.

## 2. Sources of heavy metals in the environment

Heavy metals enter the environment from natural and anthropogenic sources. The most significant natural sources are weathering of minerals, erosion and volcanic activity while anthropogenic sources include mining, smelting, electroplating, use of pesticides and (phosphate) fertilizers as well as biosolids in agriculture, sludge dumping, industrial discharge, atmospheric deposition, etc. (Modaihsh et al., 2004; Chehregani and Malayeri, 2007; Fulekar et al., 2009; Sabiha-Javied et al., 2009; Wuana and Okieimen, 2011). Table 1 gives anthropogenic sources of selected heavy metals in the environment.

## 3. Harmful effects of heavy metals on human health

Heavy metals have adverse effects on human health and therefore heavy metal contamination of food chain deserves special attention. Many heavy metals and metalloids are toxic and can

**Table 1**
Anthropogenic sources of specific heavy metals in the environment.

| Heavy metal | Sources | Reference |
|---|---|---|
| As | Pesticides and wood preservatives | Thangavel and Subbhuraam (2004) |
| Cd | Paints and pigments, plastic stabilizers, electroplating, incineration of cadmium-containing plastics, phosphate fertilizers | Salem et al. (2000); Pulford and Watson (2003) |
| Cr | Tanneries, steel industries, fly ash | Khan et al. (2007) |
| Cu | Pesticides, fertilizers | Khan et al. (2007) |
| Hg | Release from Au–Ag mining and coal combustion, medical waste | Memon et al. (2001), Wuana and Okieimen (2011), and Rodrigues et al. (2012) |
| Ni | Industrial effluents, kitchen appliances, surgical instruments, steel alloys, automobile batteries | Tariq et al. (2006) |
| Pb | Aerial emission from combustion of leaded petrol, battery manufacture, herbicides and insecticides | Thangavel and Subbhuraam (2004), Wuana and Okieimen (2011) |

**Table 2**
Harmful effects of specific heavy metals on human health.

| Heavy metal | Harmful effects | References |
|---|---|---|
| As | As (as arsenate) is an analogue of phosphate and thus interferes with essential cellular processes such as oxidative phosphorylation and ATP synthesis | Tripathi et al. (2007) |
| Cd | Carcinogenic, mutagenic, and teratogenic; endocrine disruptor; interferes with calcium regulation in biological systems; causes renal failure and chronic anemia | Degraeve (1981), Salem et al. (2000), and Awofolu (2005) |
| Cr | Causes hair loss | Salem et al. (2000) |
| Cu | Elevated levels have been found to cause brain and kidney damage, liver cirrhosis and chronic anemia, stomach and intestinal irritation | Salem et al. (2000), Wuana and Okieimen (2011) |
| Hg | Anxiety, autoimmune diseases, depression, difficulty with balance, drowsiness, fatigue, hair loss, insomnia, irritability, memory loss, recurrent infections, restlessness, vision disturbances, tremors, temper outbursts, ulcers and damage to brain, kidney and lungs | Neustadt and Pieczenik (2007), Ainza et al. (2010), and Gulati et al. (2010) |
| Ni | Allergic dermatitis known as nickel itch; inhalation can cause cancer of the lungs, nose, and sinuses; cancers of the throat and stomach have also been attributed to its inhalation; hematotoxic, immunotoxic, neurotoxic, genotoxic, reproductive toxic, pulmonary toxic, nephrotoxic, and hepatotoxic; causes hair loss | Salem et al. (2000), Khan et al. (2007), Das et al. (2008), Duda-Chodak and Baszczyk (2008), and Mishra et al. (2010) |
| Pb | Its poisoning causes problems in children such as impaired development, reduced intelligence, loss of short-term memory, learning disabilities and coordination problems; causes renal failure; increased risk for development of cardiovascular disease. | Salem et al. (2000), Padmavathiamma and Li (2007), Wuana and Okieimen (2011) and Iqbal (2012) |
| Zn | Over dosage can cause dizziness and fatigue. | Hess and Schmid (2002) |

cause undesirable effects and severe problems even at very low concentrations (Kara, 2005; Arora et al., 2008; Memon and Schröder, 2009). Heavy metals cause oxidative stress (Mudipalli, 2008) by formation of free radicals. Oxidative stress refers to enhanced generation of reactive oxygen species (ROS), which can overwhelm cell's intrinsic antioxidant defenses and can lead to cell damage or death (Das et al., 2008; Krystofova et al., 2009; Sánchez-Chardi et al., 2009). Furthermore, they can replace essential metals in pigments or enzymes disrupting their function (Malayeri et al., 2008). Regarding their toxicities, the most problematic heavy metals are Hg, Cd, Pb, As, Cu, Zn, Sn, and Cr (Wright, 2007; Ghosh, 2010). Out of these, Hg, Cd, Pb, and As are non-essential heavy metals while Cu and Zn are essential heavy metals (trace elements). Toxic heavy metals can cause different health problems depending on the heavy metal concerned, its concentration and oxidation state, etc. Table 2 gives harmful effects of selected heavy metals on human health.

## 4. Cleanup of heavy metal-contaminated soils

The concentrations of heavy metals in the environment increase from year to year (Govindasamy et al., 2011). The Campine region in Belgium and the Netherlands with 700 $km^2$ is diffusely contaminated by atmospheric deposition of Cd, Zn and Pb (Meers et al., 2010). In China alone, a total area of $2.88 \times 10^6$ ha of destroyed land has been produced as a result of mining and an additional mean area of 46 700 ha of destroyed land is produced annually. These destroyed lands almost completely lack vegetation due to serious pollution and ultimately cause severe soil erosion and off-site pollution (Xia, 2004). Therefore, cleanup of heavy metal-contaminated soils is utmost necessary in order to minimize their impact on the ecosystems. This is a challenging job with respect to cost and technical complexity (Barceló and Poschenrieder, 2003). So far different physical, chemical and biological approaches have been employed for this purpose. The conventional remediation methods include *in situ* vitrification, soil incineration, excavation and landfill, soil washing, soil flushing, solidification, and stabilization of electro-kinetic systems (Sheoran et al., 2011; Wuana and Okieimen, 2011). Generally, the physical and chemical methods suffer from limitations like high cost, intensive labor, irreversible changes in soil properties and disturbance of native soil microflora. Chemical methods can also create secondary pollution problems. Therefore, research is needed to develop cost effective, efficient and environment friendly remediation methods for decontamination of heavy metal-polluted soils. One such novel approach is phytoremediation, which is considered as a green alternative solution to the problem of heavy metal pollution.

## 5. Phytoremediation—a green solution to the problem of heavy metal pollution

"Phytoremediation basically refers to the use of plants and associated soil microbes to reduce the concentrations or toxic effects of contaminants in the environments" (Greipsson, 2011). It can be used for removal of heavy metals and radionuclides as well as for organic pollutants (such as, polynuclear aromatic hydrocarbons, polychlorinated biphenyls, and pesticides). It is a novel, cost-effective, efficient, environment- and eco-friendly, *in situ* applicable, and solar-driven remediation strategy (Clemens, 2001; Suresh and Ravishankar, 2004; LeDuc and Terry, 2005; Chehregani and Malayeri, 2007; Odjegba and Fasidi, 2007; Turan and Esringu, 2007; Lone et al., 2008; Kawahigashi, 2009; Saier and Trevors, 2010; Kalve et al., 2011; Sarma, 2011; Singh and Prasad, 2011; Vithanage et al., 2012). Plants generally handle the contaminants without affecting topsoil, thus conserving its utility and fertility. They may improve soil fertility with inputs of organic matter (Mench et al., 2009). The term "phytoremediation" is a combination of two words: Greek *phyto* (meaning plant) and Latin *remedium* (meaning to correct or remove an evil). Green plants have an enormous ability to uptake pollutants from the environment and accomplish their detoxification by various mechanisms. Phytoremediation technology is a relatively recent technology with research studies conducted mostly during the last two decades (1990 onwards). The concept of phytoremediation (as phytoextraction) was suggested by Chaney (1983). The idea is aesthetically pleasant and has good public acceptance. It is suitable for application at very large field sites where other remediation methods are not cost effective or practicable (Garbisu and Alkorta, 2003). Phytoremediation has low installation and maintenance costs compared to other remediation options (Van Aken, 2009). Regarding cost, phytoremediation can cost as less as 5% of alternative clean-up methods (Prasad, 2003). The establishment of vegetation on polluted soils also helps prevent erosion and metal leaching (Chaudhry et al., 1998). From an economic point of view, the purpose of phytoremediation of polluted land can be threefold: (1) risk containment (phytostabilization); (2) phytoextraction of metals with market value such as Ni, Tl and Au; (3) durable land management where phytoextraction gradually improves soil quality for subsequent cultivation of crops with higher market value (Vangronsveld et al., 2009). Furthermore, fast-growing and high-biomass producing plants such as willow, poplar and *Jatropha* could be used for both phytoremediation and energy production (Abhilash et al., 2012). Phytoremediation also enjoys popularity with the general public as a "green clean" alternative to chemical plants and bulldozers (Pilon-Smits, 2005).

## 6. Techniques/strategies of phytoremediation

Techniques of phytoremediation include phytoextraction (or phytoaccumulation), phytofiltration, phytostabilization, phytovolatilization, and phytodegradation (Alkorta et al., 2004).

### *6.1. Phytoextraction*

Phytoextraction (also known as phytoaccumulation, phytoabsorption or phytosequestration) is the uptake of contaminants from soil or water by plant roots and their translocation to and accumulation in aboveground biomass i.e., shoots (Sekara et al., 2005; Yoon et al., 2006; Rafati et al., 2011). Metal translocation to shoots is a crucial biochemical process and is desirable in an effective phytoextraction because the harvest of root biomass is generally not feasible (Zacchini et al., 2009; Tangahu et al., 2011).

### *6.2. Phytofiltration*

Phytofiltration is the removal of pollutants from contaminated surface waters or waste waters by plants (Mukhopadhyay and Maiti, 2010). Phytofiltration may be rhizofiltration (use of plant roots) or blastofiltration (use of seedlings) or caulofiltration (use of excised plant shoots; Latin *caulis* = shoot) (Mesjasz-Przybylowicz et al., 2004). In phytofiltration, the contaminants are absorbed or adsorbed and thus their movement to underground waters is minimized.

### *6.3. Phytostabilization*

Phytostabilization or phytoimmobilization is the use of certain plants for stabilization of contaminants in contaminated soils (Singh, 2012). This technique is used to reduce the mobility and bioavailability of pollutants in the environment, thus preventing their migration to groundwater or their entry into the food chain (Erakhrumen, 2007). Plants can immobilize heavy metals in soils through sorption by roots, precipitation, complexation or metal valence reduction in rhizosphere (Barceló and Poschenrieder, 2003; Ghosh and Singh, 2005; Yoon et al., 2006; Wuana and Okieimen, 2011). Metals of different valences vary in toxicity. By excreting special redox enzymes, plants skillfully convert hazardous metals to a relatively less toxic state and decrease possible metal stress and damage. For example, reduction of Cr(VI) to Cr(III) is widely studied, the latter being both less mobile and less toxic (Wu et al., 2010). Phytostabilization limits the accumulation of heavy metals in biota and minimizes their leaching into underground waters. However, phytostabilization is not a permanent solution because the heavy metals remain in the soil; only their movement is limited. Actually, it is a management strategy for stabilizing (inactivating) potentially toxic contaminants (Vangronsveld et al., 2009).

### *6.4. Phytovolatilization*

Phytovolatilization is the uptake of pollutants from soil by plants, their conversion to volatile form and subsequent release into the atmosphere. This technique can be used for organic pollutants and some heavy metals like Hg and Se. However, its use is limited by the fact that it does not remove the pollutant completely; only it is transferred from one segment (soil) to another (atmosphere) from where it can be redeposited. Phytovolatilization is the most controversial of phytoremediation technologies (Padmavathiamma and Li, 2007).

### *6.5. Phytodegradation*

Phytodegradation is the degradation of organic pollutants by plants with the help of enzymes such as dehalogenase and oxygenase; it is not dependent on rhizospheric microorganisms (Vishnoi and Srivastava, 2008). Plants can accumulate organic xenobiotics from polluted environments and detoxify them through their metabolic activities. From this point of view, green plants can be regarded as "Green Liver" for the biosphere. Phytodegradation is limited to the removal of organic pollutants only because heavy metals are nonbiodegradable. Recently, scientists have shown their interest in studying phytodegradation of various organic pollutants including synthetic herbicides and insecticides. Some studies have reported the use of genetically modified plants (e.g., transgenic poplars) for this purpose (Doty et al., 2007).

### *6.6. Rhizodegradation*

Rhizodegradation refers to the breakdown of organic pollutants in the soil by microorganisms in the rhizosphere (Mukhopadhyay and Maiti, 2010). Rhizosphere extends about 1 mm around the root and is under the influence of the plant (Pilon-Smits, 2005). The main reason for the enhanced degradation of pollutants in the rhizosphere is likely the increase in the numbers and metabolic activities of the microbes. Plants can stimulate microbial activity about 10–100 times higher in the rhizosphere by the secretion of exudates containing carbohydrates, amino acids, flavonoids. The release of nutrients-containing exudates by plant roots provides carbon and nitrogen sources to the soil microbes and creates a nutrient-rich environment in which microbial activity is stimulated. In addition to secreting organic substrates for facilitating the growth and activities of rhizospheric microorganisms, plants also release certain enzymes capable of degrading organic contaminants in soils (Kuiper et al., 2004; Yadav et al., 2010).

### *6.7. Phytodesalination*

It is a recently reported and emerging technique (Zorrig et al., 2012). Phytodesalination refers to the use of halophytic plants for removal of salts from salt-affected soils in order to enable them for supporting normal plant growth (Manousaki and Kalogerakis, 2011; Sakai et al., 2012). Halophytic plants have been suggested to be naturally better adapted to cope with heavy metals compared to glycophytic plants (Manousaki and Kalogerakis, 2011). According to an estimation, two halophytes, *Suaeda maritima* and *Sesuvium portulacastrum* could remove 504 and 474 kg of sodium chloride respectively from 1 ha of saline soil in a period of 4 months. Therefore, *S. maritima* and *S. portulacastrum* could be successfully used to accumulate NaCl from highly saline soils and enable them for crop production after a few repeated cultivation and harvest (Ravindran et al., 2007). Another study has reported accumulation of about 1 t $ha^{-1}$ of $Na^{+}$ ions in the aboveground biomass of the obligate halophyte *S. portulacastrum* cultivated on a salinized soil. The resultant decrease in salinity and sodocity of the phytodesalinized soil significantly reduced the negative effects on the growth of the test culture of the glycophytic crop, *Hordeum vulgare* (Rabhi et al., 2010).

Table 3 summarizes the different techniques of phytoremediation.

**Table 3**
Summary of the different techniques of phytoremediation.

| Technique | Description |
|---|---|
| Phytoextraction | Accumulation of pollutants in harvestable biomass i.e., shoots |
| Phytofiltration | Sequestration of pollutants from contaminated waters by plants |
| Phytostabilization | Limiting the mobility and bioavailability of pollutants in soil by plant roots |
| Phytovolatilization | Conversion of pollutants to volatile form and their subsequent release to the atmosphere |
| Phytodegradation | Degradation of organic xenobiotics by plant enzymes within plant tissues |
| Rhizodegradation | Degradation of organic xenobiotics in the rhizosphere by rhizospheric microorganisms |
| Phytodesalination | Removal of excess salts from saline soils by halophytes |

## 7. Phytoextraction of heavy metals

Phytoextraction is the main and most useful phytoremediation technique for removal of heavy metals and metalloids from polluted soils, sediments or water (Cluis, 2004; Cherian and Oliveira, 2005; Milic et al., 2012). It is the most promising for commercial application (Sun et al., 2011a). The efficiency of phytoextraction depends on many factors like bioavailability of the heavy metals in soil, soil properties, speciation of the heavy metals and plant species concerned. Plants suitable for phytoextraction should ideally have the following characteristics (Mejáre and Bülow, 2001; Tong et al., 2004; Adesodun et al., 2010; Sakakibara et al., 2011; Shabani and Sayadi, 2012):

(i) High growth rate.
(ii) Production of more above-ground biomass.
(iii) Widely distributed and highly branched root system.
(iv) More accumulation of the target heavy metals from soil.
(v) Translocation of the accumulated heavy metals from roots to shoots.
(vi) Tolerance to the toxic effects of the target heavy metals.
(vii) Good adaptation to prevailing environmental and climatic conditions.
(viii) Resistance to pathogens and pests.
(ix) Easy cultivation and harvest.
(x) Repulsion to herbivores to avoid food chain contamination.

The phytoextraction potential of a plant species is mainly determined by two key factors i.e., shoot metal concentration and shoot biomass (Li et al., 2010). Two different approaches have been tested for phytoextraction of heavy metals: (1) The use of hyperaccumulators, which produce comparatively less aboveground biomass but accumulate target heavy metals to a greater extent (2) The application of other plants, such as *Brassica juncea* (Indian mustard), which accumulate target heavy metals to a lesser extent but produce more aboveground biomass so that overall accumulation is comparable to that of hyperaccumulators due to production of more biomass (Robinson et al., 1998; Tlustoš et al., 2006). According to Chaney et al. (1997), hyperaccumulation and hypertolerance are more important in phytoremediation than high biomass. Use of hyperaccumulators will yield a metal-rich, low-volume biomass, which is economical and easy to handle in case of both metal recovery and safe disposal. On the other hand, use of non-accumulators will yield a metal-poor, large-volume biomass, which will be uneconomical to process for recovery of metals and also costly to safely dispose.

Plants, which offer multiple harvests in a single growth period (like *Trifolium* spp.) can have a great potential for phytoextraction of heavy metals (Ali et al., 2012). Grasses are more preferable for phytoextraction than shrubs or trees because of their high growth rate, more adaptability to stress environment and high biomass (Malik et al., 2010). Some researchers have evaluated the use of crops (such as maize, and barley) for phytoextraction of heavy metals. In this case, several cropping are required to lower heavy metal contamination to acceptable levels. However, the use of crops for phytoextraction of heavy metals suffers from the disadvantage of contamination of food chain. According to Vamerali et al. (2010), the use of field crops for phytoremediation purposes should not consider the use of products for animal feed or direct human consumption.

## 8. Bioavailability of heavy metals in soil—natural versus induced phytoextraction

The chemical composition and sorption properties of soil influence the mobility and bioavailability of metals (Kłos et al., 2012). The bioavailability of heavy metals in soil is a critical factor affecting the efficiency of phytoextraction of target heavy metals. Low bioavailability is a major limiting factor for phytoextraction of contaminants such as Pb. Generally, only a fraction of soil metal is bioavailable for uptake by plants (Lasat, 2002). Strong binding of heavy metals to soil particles or precipitation causes a significant fraction of soil heavy metals insoluble and therefore mainly unavailable for uptake by plants (Sheoran et al., 2011). Regarding the bioavailability of heavy metals/metalloids in soil, there can be three categories: readily bioavailable (Cd, Ni, Zn, As, Se, Cu); moderately bioavailable (Co, Mn, Fe) and least bioavailable (Pb, Cr, U) (Prasad, 2003). However, plants have developed certain mechanisms for solubilizing heavy metals in soil. Plant roots secrete metal-mobilizing substances in the rhizosphere called phytosiderophores (Lone et al., 2008). Secretion of $H^+$ ions by roots can acidify the rhizosphere and increase metal dissolution. $H^+$ ions can displace heavy metal cations adsorbed to soil particles (Alford et al., 2010). Root exudates can lower the rhizosphere soil pH generally by one or two units over that in the bulk soil. Lower soil pH increases concentration of heavy metals in solution by promoting their desorption (Thangavel and Subbhuraam, 2004). Furthermore, the rhizospheric microorganisms (mainly bacteria and mycorrhizal fungi) may significantly increase the bioavailability of heavy metals in soil (Vamerali et al., 2010; Sheoran et al., 2011). Interactions of microbial siderophores can increase labile metal pools and uptake by roots (Mench et al., 2009).

Phytoextraction of heavy metals can be practiced in two modes, natural and induced. In natural or continuous phytoextraction, plants are used for removal of heavy metals under natural conditions i.e., no soil amendment is made. In induced or chelate assisted phytoextraction, different chelating agents such as EDTA, citric acid, elemental sulfur, and ammonium sulfate are added to soil to increase the bioavailability of heavy metals in soil for uptake by plants (Elkhatib et al., 2001; Lai and Chen, 2004; Lone et al., 2008; Sun et al., 2011b). The chelates form water-soluble complexes with the heavy metals in soil and help in their desorption from soil particles. Bioavailability of the heavy metals can also be increased by lowering soil pH since metal salts are soluble in acidic media rather than in basic media. However, these chemical treatments can cause secondary pollution problems. For example, synthetic chelate EDTA is nonbiodegradable and can leach into ground-water supplies making an additional environmental hazard. Furthermore, synthetic chelating agents can also be toxic to plants at high concentrations. Thus proper care should be taken when practicing induced phytoextraction (Lombi et al., 2001; Lai and Chen, 2004; Zhuang et al., 2005; Marques et al., 2009; Ping et al., 2009; Zhao et al., 2011; Song et al., 2012). However, use of citric acid as a chelating agent could be promising because it has a natural origin and is easily biodegraded in soil. Furthermore, citric acid is not toxic to plants, therefore plant growth is not limited (Smolinska and Krol, 2012).

## 9. Metallophytes

Metallophytes are plants that are specifically adapted to and thrive in heavy metal-rich soils (Bothe, 2011; Sheoran et al., 2011). The primary sites of plants resistant to heavy metals are soils where ores are outcropping, the so-called metalliferous or orogenic soils (Ernst, 1974). Metal exposure to a surplus of various metals over thousands of years has driven the evolution of metal resistance in metallophytes under the local environmental conditions. Mining activities have destroyed and are still diminishing the metal-enriched habitats and consequently changing the niche for the metallophytes (Ernst, 2000). Metallophytes are botanical curiosities (Alford et al., 2010). These plants are concentrated in the plant family Brassicaceae. Their use, either alone or in combination with microorganisms, for phytoremediation of heavy metal-contaminated soils is an attractive idea (Bothe, 2011). Metallophytes are divided into three categories: metal excluders, metal indicators and metal hyperaccumulators.

### *9.1. Metal excluders*

Metal excluders accumulate heavy metals from substrate into their roots but restrict their transport and entry into their aerial parts (Sheoran et al., 2011; Malik and Biswas, 2012). Such plants have a low potential for metal extraction but may be efficient for phytostabilization purposes (Lasat, 2002; Barceló and Poschenrieder, 2003).

### *9.2. Metal indicators*

Metal indicators accumulate heavy metals in their aerial parts. As the name indicates, these plants generally reflect heavy metal concentrations in the substrate (Sheoran et al., 2011).

### *9.3. Metal hyperaccumulators*

Hyperaccumulators are plants, which can concentrate heavy metals in their aboveground tissues to levels far exceeding those present in the soil or in the nearby growing non-accumulating plants (Memon et al., 2001; Memon and Schröder, 2009). Hyperaccumulators can be regarded as a special and extreme case of the broader category of accumulators (Pollard et al., 2002). They are hypertolerant to the metals, which they accumulate in the shoots (McGrath et al., 2001). The standard for hyperaccumulators has not been defined scientifically (Nazir et al., 2011). However, individual authors or research groups have defined hyperaccumulators. The term "hyperaccumulator" was first coined by Brooks et al. (1977) to define plants with Ni concentrations higher than 1000 mg $kg^{-1}$ dry weigh (0.1%). Reeves (1992) attempted to define Ni hyperaccumulation with greater precision as "a Ni hyperaccumulator is a plant in which a Ni concentration of at least 1000 mg $kg^{-1}$ has been recorded in the dry matter of any aboveground tissue in at least one specimen growing in its natural habitat. For establishing hyperaccumulator status, aboveground tissue should be regarded as plant foliage only. The phrase "growing in its natural habitat" implies that hyperaccumulators must achieve metal hyperaccumulation while remaining healthy enough to maintain a self-sustaining population (van der Ent et al., 2013). To the authors' notice, the most cited criteria for hyperaccumulation of metals is that of Baker and Brooks (1989) (with 1376 citations so far), according to which "hyperaccumulators are plant species, which accumulate greater than 100 mg $kg^{-1}$ dry weight Cd, or greater than 1000 mg $kg^{-1}$ dry weight Ni, Cu and Pb or greater than 10000 mg $kg^{-1}$ dry weight Zn and Mn in their shoots when grown on metal rich soils". van der Ent et al. (2013) admit that criteria commonly used for hyperaccumulation of some metals are unnecessarily conservative and propose that criteria for hyperaccumulation of such metals be lowered. They recommend the following concentration criteria for different metals and metalloids in dried foliage with plants growing in their natural habitats: 100 mg $kg^{-1}$ for Cd, Se and Tl; 300 mg $kg^{-1}$ for Co, Cu and Cr; 1000 mg $kg^{-1}$ for Ni, Pb and As; 3000 mg $kg^{-1}$ for Zn; 10000 mg $kg^{-1}$ for Mn. Generally, hyperaccumulators achieve 100-fold higher shoot metal concentration (without yield reduction) compared to crop plants or common nonaccumulator plants (Lasat, 2002; Chaney et al., 2007). Hyperaccumulators achieve a shoot-to-root metal concentration ratio (called translocation factor, TF) of greater than 1 (Tangahu et al., 2011; Badr et al., 2012). However, TF cannot be used alone to define hyperaccumulation although it is a useful measure in supporting other evidence of hyperaccumulation (van der Ent et al., 2013).

Exploring more effective hyperaccumulators for heavy metals is a key step for successful phytoremediation of these pollutants (Wei et al., 2008; Zhang et al., 2010). van der Ent et al. (2013) point out that hyperaccumulators have to be recorded from the natural habitats. They do not regard extreme accumulation achieved through hydroponics or metal-amended spiked soils and artificially acidified soils as hyperaccumulation. They do not consider such experiments alone as capable of defining a species as a hyperaccumulator. They argue that natural populations must be studied. Literature shows that more than 400 plant species have been identified as metal-hyperaccumulators with more than 300 Ni-hyperaccumulators (Li et al., 2003; Prasad, 2005). Family Brassicaceae contains many metal-accumulating species (Poniedziałek et al., 2010). Examples of hyperaccumulators are *Thlaspi caerulescens* and *Alyssum bertolonii*. *Thlaspi caerulescens* (Alpine pennycress) is possibly the best-known metal hyperaccumulator (Lasat, 2002). This species is a hyperaccumulator for Zn, Cd and Ni (Assunção et al., 2003). The most commonly postulated hypothesis regarding the reason or advantage of metal hyperaccumulation in plants is elemental defense against herbivores (by making leaves unpalatable or toxic) and pathogens (Meharg, 2005; Prasad, 2005; Dipu et al., 2012). Table 4 gives a list of some metal hyperaccumulators.

Hyperaccumulators can be used for phytoremediation of toxic and hazardous heavy metals as well as for phytomining of precious heavy metals (such as Au, Pd and Pt). The use of hyperaccumulators for phytoremediation might result in production of a bio-ore of some commercial value to cope with some of the costs of soil remediation (Brooks et al., 1998). The amount of heavy metals removed from soil by hyperaccumulators is a function of tissue metal concentration multiplied by the quantity of biomass produced (Macek et al., 2008). Some plants have natural ability of hyperaccumulation for specific heavy metals. These plants are known as natural hyperaccumulators. On the other hand, the accumulation capacity of some plants for specific heavy metals can be enhanced by their genetic modification through biotechnological methods. Such genetically modified plants have shown promising results for phytoremediation of some heavy metals. However, since some environmental scientists are skeptic about the bio-safety of genetically modified organisms (GMOs), therefore there is a world-wide concern about the commercialization of such products.

## 10. Quantification of phytoextraction efficiency

The efficiency of phytoextraction can be quantified by calculating bioconcentration factor and translocation factor.

**Table 4**
List of some hyperaccumulator plants.

| Plant species | Metal | Metal accumulation (mg kg$^{-1}$) | Reference |
|---|---|---|---|
| *Alyssum bertolonii* | Ni | 10900 | Li et al. (2003) |
| *Alyssum caricum* | Ni | 12500 | Li et al. (2003) |
| *Alyssum corsicum* | Ni | 18100 | Li et al. (2003) |
| *Alyssum heldreichii* | Ni | 11800 | Bani et al. (2010) |
| *Alyssum markgrafii* | Ni | 19100 | Bani et al. (2010) |
| *Alyssum murale* | Ni | 4730–20100 | Bani et al. (2010) |
| | | 15000 | Li et al. (2003) |
| *Alyssum pterocarpum* | Ni | 13500 | Li et al. (2003) |
| *Alyssum serpyllifolium* | Ni | 10000 | Prasad (2005) |
| *Azolla pinnata* | Cd | 740 | Rai (2008) |
| *Berkheya coddii* | Ni | 18000 | Mesjasz-Przybylowicz et al. (2004) |
| *Corrigiola telephiifolia* | As | 2110 | (Garcia-Salgado et al., 2012) |
| *Eleocharis acicularis* | Cu | 20200 | Sakakibara et al. (2011) |
| | Zn | 11200 | |
| | Cd | 239 | |
| | As | 1470 | |
| *Euphorbia cheiradenia* | Pb | 1138 | Chehregani and Malayeri (2007) |
| *Isatis pinnatiloba* | Ni | 1441 | Altinozlu et al. (2012) |
| *Pteris biaurita* | As | ~2000 | Srivastava et al. (2006) |
| *Pteris cretica* | As | ~1800 | Srivastava et al. (2006) |
| | | 2200–3030 | Zhao et al. (2002) |
| *Pteris quadriaurita* | As | ~2900 | Srivastava et al. (2006) |
| *Pteris ryukyuensis* | As | 3647 | Srivastava et al. (2006) |
| *Pteris vittata* | As | 8331 | Kalve et al. (2011) |
| | | ~1000 | Baldwin and Butcher (2007) |
| | Cr | 20675 | Kalve et al. (2011) |
| *Rorippa globosa* | Cd | >100 | Wei et al. (2008) |
| *Schima superba* | Mn | 62412.3 | Yang et al. (2008) |
| *Solanum photeinocarpum* | Cd | 158 | Zhang et al. (2011) |
| *Thlaspi caerulescens* | Cd | 263 | Lombi et al. (2001) |

Bioconcentration factor indicates the efficiency of a plant species in accumulating a metal into its tissues from the surrounding environment (Ladislas et al., 2012). It is calculated as follows (Zhuang et al., 2007).

$$\text{Bioconcentration Factor (BCF)} = \frac{C_{\text{harvested tissue}}}{C_{\text{soil}}} \quad (1)$$

where $C_{\text{harvested tissue}}$ is the concentration of the target metal in the plant harvested tissue and $C_{\text{soil}}$ is the concentration of the same metal in the soil (substrate).

Translocation factor indicates the efficiency of the plant in translocating the accumulated metal from its roots to shoots. It is calculated as follows (Padmavathiamma and Li, 2007).

$$\text{Translocation Factor (TF)} = \frac{C_{\text{shoot}}}{C_{\text{root}}} \quad (2)$$

where $C_{\text{shoot}}$ is concentration of the metal in plant shoots and $C_{\text{root}}$ is concentration of the metal in plant roots.

Bioconcentration factor or accumulation factor ($A$) can also be represented in percent according to the following equation (Wilson and Pyatt, 2007).

$$\text{Accumulation Factor}(A) = \frac{C_{\text{plant tissue}}}{C_{\text{soil}}} \times 100 \quad (3)$$

where $A$ is accumulation factor %, $C_{\text{plant tissue}}$ is metal concentration in plant tissue and $C_{\text{soil}}$ is metal concentration in soil. Similarly, translocation factor can also be represented in percent according to the following equation (Zacchini et al., 2009).

$$\text{TF} = \frac{C_{\text{aerial parts}}}{C_{\text{roots}}} \times 100 \quad (4)$$

Both BCF and TF are important in screening hyperaccumulators for phytoextraction of heavy metals. The evaluation and selection of plants for phytoremediation purposes entirely depend on BCF and TF values (Wu et al., 2011). BCF is a more important measure than shoot metal concentration when considering the potential of a given candidate species for phytoextraction (Sakakibara et al., 2011). Translocation factor value greater than 1 indicates the translocation of the metal from root to above-ground part (Jamil et al., 2009). According to Yoon et al. (2006), only plant species with both BCF and TF greater than 1 have the potential to be used for phytoextraction. Hyperaccumulators have BCF greater than 1, sometimes reaching 50–100 (Cluis, 2004). However, high metal concentrations in soil could result in a BCF < 1, for example in ultramafic soils with 3000 mg kg$^{-1}$ Ni in the soil and 2000 mg kg$^{-1}$ in a plant or conversely plants growing on soils deficient in essential trace elements (e.g., Zn) might be very efficient in sequestration and therefore have very high BCFs yet low absolute tissue metal concentrations. Thus BCF might have use for comparisons in case of growing plants in homogenized soil or in hydroponic cultures but has little advantage over simple comparisons of foliar metal concentrations (van der Ent et al., 2013). BCF is also a convenient and reliable way for quantifying the relative difference in the bioavailability of heavy metals to plants (Naseem et al., 2009).

## 11. The fate of plants used for phytoextraction

An important question is that what will be the fate of plants after being used for phytoextraction of heavy metals? Such plants after burning, can be either disposed as hazardous waste safely in specialized dumps or if economically feasible, processed for biorecovery of precious and semiprecious metals (a practice known as phytomining) (Salt et al., 1998; Prasad, 2003; Jadia and Fulekar, 2008; Lone et al., 2008; Jadia and Fulekar, 2009; Sheoran et al., 2011). This can be outlined as in Fig. 1.

## 12. Phytomining

Plant biomass containing accumulated heavy metals can be combusted to get energy and the remaining ash is considered as “bio-ore”. This bio-ore can be processed for the recovery or

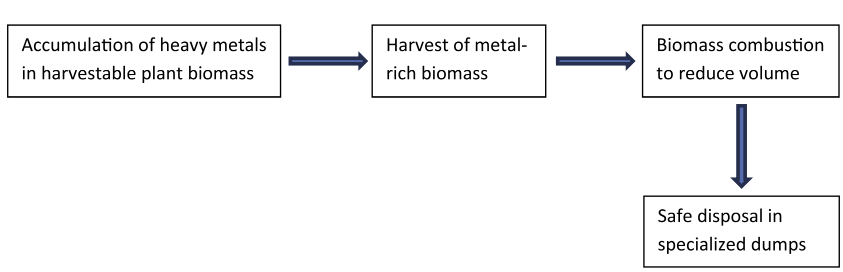


**Fig. 1.** Main route of post-harvest treatment of phytoremediator plants.

extraction of the heavy metals. An advantage of phytomining is the sale of energy from combustion of the biomass (Anderson et al., 1999). According to a field experiment conducted by Meers et al. (2010), cultivation of energy maize in the Campine region in Belgium and the Netherlands could result in the generation of 30 000–42 000 $kWh_{el+th}$ of renewable energy per hectare. By assuming the substitution of coal powered power plant, this would imply a cut of up to 21 tons $ha^{-1}$ $y^{-1}$ $CO_2$. Processing bio-ores contributes less $SO_x$ emissions to the atmosphere because of their low sulfur contents. Thus phytomining is an environment- and eco-friendly option as compared to the conventional extraction methods. However, the commercial viability of phytomining depends on many factors like the efficiency of phytoextraction and current market value of the processed metals. Phytomining has been commercially used for Ni and it is believed that it is less expensive than the conventional extraction methods. Using *Alyssum murale* and *Alyssum corsicum*, one can grow biomass containing 400 kg Ni $ha^{-1}$ with production costs of \$250–500 $ha^{-1}$. Considering Ni price of \$40 $kg^{-1}$ (in 2006, Ni metal was trading on the London Metal Exchange at more than \$40 $kg^{-1}$), Ni phytomining has become a highly profitable agricultural technology (crop value = \$16 000 $ha^{-1}$) for Ni-contaminated or mineralized soils (Chaney et al., 2007). The enforcement of more strict legislation for limiting environmental pollution would make bio-based mining more attractive (Siddiqui et al., 2009).

## 13. Use of constructed wetlands for phytoremediation

Constructed wetlands are used for clean-up of effluents and drainage waters (Vangronsveld et al., 2009). They offer a cost-effective and technically feasible technology and have proven effective and successful in remediation of heavy metal pollution and of various water quality issues (Williams, 2002; Olguin and Sanchez-Galvan, 2010; Rai, 2012). Aquatic macrophytes are more suitable for wastewater treatment than terrestrial plants due to their faster growth, production of more biomass and relative higher ability of pollutant uptake. They perform better purification due to direct contact with contaminated water (Sood et al., 2012). In constructed wetlands, different floating, emergent and submerged aquatic species are used. Poplar (*Populus* spp.) and willow (*Salix* spp.) can be used on the edges of constructed wetlands (Pilon-Smits, 2005). The floating aquatic plants accumulate metals by their roots while the submerged plants accumulate metals by their whole bodies (Rahman and Hasegawa, 2011). Water hyacinth (*Eichhornia crassipes*) has been used for phytoremediation of heavy metals at constructed wetlands. It is a fast growing, floating plant with a well-developed fibrous root system and large biomass. It also adapts easily to various aquatic conditions and plays an important role in accumulating metals from water (Liao and Chang, 2004). Similarly, water lettuce (*Pistia stratiotes*) has been pointed out as a potential phytoremediator plant for Mn contaminated waters. Its additional advantages are abundant growth in wetlands, coverage of almost the entire water surface and easy harvest (Hua et al., 2012). Another candidate for aquatic phytoremediation is *Azolla*. It is a better macrophyte for aquatic phytoremediation because of its short doubling time (2–3 d), easy harvest, nitrogen fixation ability and tolerance to and accumulation of a wide range of heavy metals (Sood et al., 2012). Wetland plants are best selected from local, endemic wetland species (Adams et al., in press).

## 14. Mechanism of heavy metals' uptake, translocation, and tolerance

Plants take heavy metals from soil solution into their roots. After entry into roots, heavy metal ions can either be stored in the roots or translocated to the shoots primarily through xylem vessels (Prasad, 2004; Jabeen et al., 2009) where they are mostly deposited in vacuoles. Vacuoles are the cellular organelles with low metabolic activities (Denton, 2007). Heavy metal sequestration in the vacuole is one of the ways to remove excess metal ions from the cytosol and may reduce their interactions with cellular metabolic processes (Assunção et al., 2003; Sheoran et al., 2011). Compartmentalization of complexed metals in vacuoles is part of the tolerance mechanism in metal hyperaccumulators (Cluis, 2004; Tong et al., 2004). The entire mechanism of phytoextraction of heavy metals has five basic aspects: mobilization of the heavy metals in soil, uptake of the metal ions by plant roots, translocation of the accumulated metals from roots to aerial tissues, sequestration of the metal ions in plant tissues and metal tolerance. Metal tolerance is a key prerequisite for metal accumulation and hence phytoremediation (Clemens, 2001; Tong et al., 2004). Mechanisms governing heavy metal tolerance in plant cells are cell wall binding, active transport of ions into the vacuole and chelation through the induction of metal-binding peptides and the formation of metal complexes (Mejáre and Bülow, 2001; Memon and Schröder, 2009).

The overall journey of heavy metals from soil solution to the vacuoles is controlled and regulated by a variety of molecules. Some molecules are involved in the cross-membrane transport of the heavy metals and others are involved in their complexation and subsequent sequestration. Uptake of heavy metal ions from soil solution is mediated by specialized transporters (channel proteins) or $H^+$ coupled carrier proteins present in the plasma membrane of the root (Greipsson, 2011). For example, transporters of the ZIP (zinc–iron permease) family contribute to the uptake of $Zn^{2+}$ and $Fe^{2+}$ (Clemens, 2001). Nramp (natural resistance-associated macrophage) is another family of proteins, which plays an important role in transport of divalent metal ions (Seth, 2012). Non-essential heavy metals may effectively compete for and enter roots through the same transmembrane transporters used by essential heavy metals having similar oxidation states and ionic radii (Thangavel and Subbhuraam, 2004; Alford et al., 2010). This relative lack of selectivity in transmembrane ion transport may partially explain the reason of entry of non-essential heavy metals in plant cells, even against a concentration gradient (Seth, 2012). Organic acids and amino acids are suggested as ligands for chelation of heavy metal ions because of the presence of donor atoms (S, N, and O) in their molecules (Shah and Nongkynrih, 2007; Sheoran et al., 2011).

## 15. Role of phytochelatins and metallothioneins in phytoextraction

The most important peptides/proteins involved in metal accumulation and tolerance are phytochelatins (PCs) and metallothioneins (MTs). Plant PCs and MTs are rich in cysteine sulfhydryl groups, which bind and sequester heavy metal ions in very stable complexes (Kärenlampi et al., 2000). PCs are small glutathione-derived, enzymatically synthesized peptides, which bind metals and are principal part of the metal detoxification system in plants (Clemens, 2001; Cobbett and Goldsbrough, 2002; Yurekli and Kucukbay, 2003; Fulekar et al., 2009). They have the general structure of $(\gamma\text{-glutamyl-cysteinyl})_n$-glycine where $n = 2–11$ (Inouhe, 2005). Chemical structure of PCs is shown in Fig. 2 (Seth, 2012). They are produced by the enzyme phytochelatin synthase (Sarma, 2011). PC synthase is activated by various heavy metal ions with *in vivo* induction of PCs (Cobbett, 2000). Mutants of *Arabidopsis thaliana* that lack PC-synthase are unable to synthesize PCs and are hypersensitive to Cd and Hg (Memon et al., 2001; Memon and Schröder, 2009). The accumulation of $Pb^{2+}$ in the aquatic fern *Salvinia minima* caused changes in the expression of the *Sm*PCS

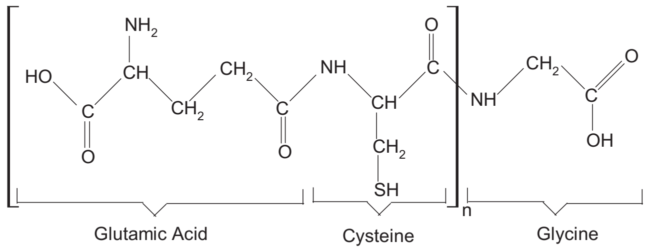


**Fig. 2.** Chemical structure of PCs (adapted from Seth, 2012).

gene. Consequently *in vivo* PCS (phytochelatin synthase) activity and PC production were increased in roots and to a lower extent in leaves (Gomez et al., 2009). MTs are gene-encoded, low molecular weight, metal-binding proteins, which can protect plants against the effects of toxic metal ions (Cobbett and Goldsbrough, 2002; Fulekar et al., 2009; Jabeen et al., 2009; Sheoran et al., 2011). By over expression of natural chelators (PCs, MTs, and organic acids), not only metal ions' entrance into plant cell but also translocation through xylem is facilitated (Wu et al., 2010). Modification or over expression of GSH (glutathione) and PCS gene has significant potential for increasing heavy metal accumulation and tolerance in plants (Seth, 2012). Studies are in progress to identify, isolate and characterize the biomolecules involved in the cross-membrane transport and vacuolar sequestration of heavy metals in plants. Advancement in such molecular studies will greatly help to improve our understanding of the complete mechanism of metal uptake, translocation and tolerance in plants, which in turn will help to enhance the efficiency of phytoremediation.

## 16. Limitations of phytoremediation

Although phytoremediation is a promising approach for remediation of heavy metal-contaminated soils, it also suffers from some limitations (Clemens, 2001; Tong et al., 2004; LeDuc and Terry, 2005; Karami and Shamsuddin, 2010; Mukhopadhyay and Maiti, 2010; Naees et al., 2011; Ramamurthy and Memarian, 2012).

- Long time required for clean-up.
- Phytoremediation efficiency of most metal hyperaccumulators is usually limited by their slow growth rate and low biomass.
- Difficulty in mobilization of more tightly bound fraction of metal ions from soil i.e., limited bioavailability of the contaminants in the soil.
- It is applicable to sites with low to moderate levels of metal contamination because plant growth is not sustained in heavily polluted soils.
- There is a risk of food chain contamination in case of mismanagement and lack of proper care.

## 17. Future trends in phytoremediation

As mentioned earlier, phytoremediation is a relatively recent field of research and application. Currently most research is limited to laboratory and greenhouse scale studies and only a few studies have been conducted to test the efficiency of phytoremediation in actual field. Results in actual field can be different from those at laboratory or greenhouse conditions (Ji et al., 2011) because field is a real world where different factors simultaneously play their role. Factors that may affect phytoremediation in the field include variations in temperature, nutrients, precipitation and moisture, plant pathogens and herbivory, uneven distribution of contaminants, soil type, soil pH, and soil structure (Vangronsveld et al., 2009). Phytoremediation efficiency of different plants for specific target heavy metals has to be tested in field conditions in order to realize the feasibility of this technology for commercialization.

After identification of desirable traits in natural hyperaccumulators, such traits can be selected either by conventional breeding techniques or by using new methods of hybridization such as protoplast fusion or by the manipulation of gene expression in transgenic plants (Pollard et al., 2002). Research is in progress to identify genes coding for hyperaccumulation of specific heavy metals in plants. Identification and successful transformation of such genes to other suitable plants makes it possible to develop "superbug" plants for phytoremediation. Transgenic plants could be developed to secrete metal selective ligands into the rhizosphere, which could specifically solubilize elements of phytoremediation interest (Thakur, 2006). Thus different desirable traits can be combined into a single plant species, which would best serve the purpose. However, gene expression and interaction has to be studied honestly and wisely in order to avoid any unintended harm to the biosphere presently or in future. An understanding of the coordination chemistry of metals within plant tissues will help researchers to finely tune the process of phytoremediation (Saraswat and Rai, 2011). In spite of the many challenges, phytoremediation is perceived as a green remediation technology with an expected great potential.

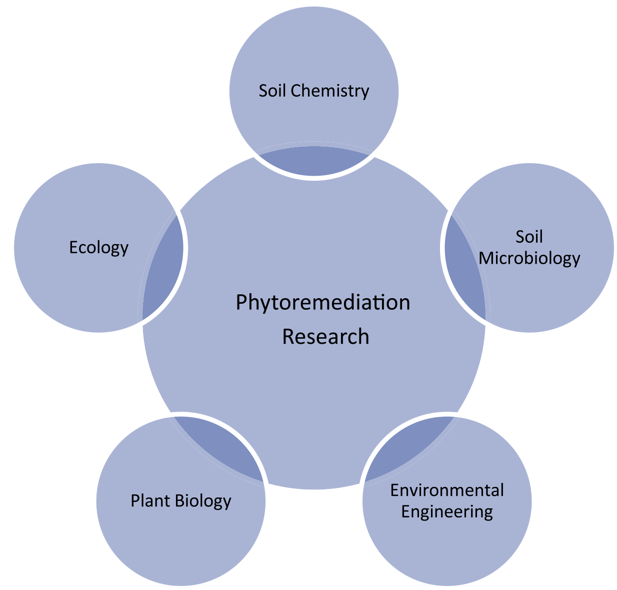


**Fig. 3.** Schematic showing interdisciplinary nature of phytoremediation research.

## 18. Interdisciplinary nature of phytoremediation research

Research in phytoremediation is truly interdisciplinary in nature and requires background knowledge in soil chemistry, plant biology, ecology and soil microbiology as well as environmental engineering (Fig. 3). In view of the current trends of integration of scientific knowledge worldwide, it is hoped that many challenging questions about commercial application of phytoremediation will be answered in future.

## 19. Conclusions

Since contamination of soils and waters by toxic heavy metals is a serious environmental problem, therefore effective remediation methods are necessary. Physical and chemical methods for clean-up and restoration of heavy metal-contaminated soils have serious limitations like high cost, irreversible changes in soil properties, destruction of native soil microflora and creation of secondary pollution problems. In contrast, phytoremediation is a better solution to the problem. Phytoremediation is environment-friendly and ecologically responsible solar-driven technology with good public acceptance. It is a relatively recent technology and is mostly in research stage. Its research is highly interdisciplinary in nature and requires background knowledge in soil chemistry, plant biology, ecology and soil microbiology as well as environmental engineering. Fortunately, interdisciplinary studies and research are appreciated and highly encouraged in broad-minded scientific communities across the globe and it is fully hoped that the integration of scientific disciplines will be highly fruitful. Research is in progress to screen native plants for phytoremediation of target heavy metals and to evaluate the effect of different parameters on phytoremediation efficiency. Furthermore, research is being conducted to genetically modify some suitable plants for better phytoremediation of heavy metals and other xenobiotics. Studies are also being done to identify and characterize the different proteins involved in cross-membrane transport and vacuolar sequestration of heavy metals. Advancement and achievements in such molecular studies will greatly help in understanding the mechanism and enhancing the efficiency of phytoremediation. An improved understanding of heavy metal uptake by plants from soil will also help in promoting phytomining—a plant-based eco-friendly mining of metals, which can be used for extraction of metals even from low-grade ores. Phytoextraction of heavy metals is expected to be a commercially viable technology for phytoremediation and phytomining of heavy metals in future.

## 20. Recommendations

1. Since phytoremediation research is truly interdisciplinary in nature, therefore researchers from different backgrounds should be welcomed and encouraged to utilize their talent and expertise in this field.
2. Existing plant diversity should be explored for hyperaccumulation of various heavy metals to find new effective metal hyperaccumulators.
3. Extensive and reliable risk assessment studies should be conducted before application of transgenic plants for phytoremediation in the field.
4. More phytoremediation studies should be conducted in the field with honest and unbiased cost-benefit analysis keeping in mind the very green nature of the technology.
5. More studies should be conducted to better understand interactions among the four players in the rhizosphere that is among metals, soil, microbes and plant roots.
6. Advancement in spectroscopic and chromatographic techniques should be exploited to improve understanding of the fate of metal ions in plant tissues, which in turn will improve understanding of metal hyperaccumulation and tolerance in plants.
7. An international forum (like IUPAC) should be developed to arrange periodic meetings to discuss and search solutions for the challenges faced by the emerging technology of phytoremediation.

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