

## **A Review on Phytoremediation of Heavy Metals and Utilization of It's by Products**

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**Abstract.** This review presents the status of phytoremediation technologies with particular emphasis on phytoextraction of soil heavy metal contamination. Unlike organic compounds, metals cannot be degraded, and cleanup usually requires their removal. Most of the conventional remedial technologies are expensive and inhibit the soil fertility; this subsequently causes negative impacts on the ecosystem. Phytoremediation is a cost effective, environmental friendly, aesthetically pleasing approach most suitable for developing countries. Despite this potential, phytoremediation is yet to become a commercially available technology in India. This paper reports about the mobility, bioavailability and plant response to presence of soil heavy metals. It classifies the plants according to phytoextraction mechanism and discusses the pathway of metal in plants. Various techniques to enhance phytoextraction and utilization of by-products have been elaborated. Since lot of biomass is produced during this process, it needs proper disposal and management. It also gives an insight into the work done by authors, which focuses on high biomass extractor plants. High biomass weeds were selected to restrict the passage of contaminants into the food chain by selecting non-edible, disease resistant and tolerant plants, which can provide renewable energy. Thus making phytoextraction more viable for present utilization.

**Keywords:** heavy metals, phytoextraction, hyperaccumulator, indicator, excluder species

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## Introduction

A major environmental concern due to dispersal of industrial and urban wastes generated by human activities is the contamination of soil. Controlled and uncontrolled disposal of waste, accidental and process spillage, mining and smelting of metalliferous ores, sewage sludge application to agricultural soils are responsible for the migration of contaminants into non-contaminated sites as dust or leachate and contribute towards contamination of our ecosystem. A wide range of inorganic and organic compounds cause contamination, these include heavy metals, combustible and putrescible substances, hazardous wastes, explosives and petroleum products. Major component of inorganic contaminants are heavy metals [1,2] they present a different problem than organic contaminants. Soil microorganisms can degrade organic contaminants, while metals need immobilisation or physical removal. Although many metals are essential, all metals are toxic at higher concentrations, because they cause oxidative stress by formation of free radicals. Another reason why metals may be toxic is that they can replace essential metals in pigments or enzymes disrupting their function [3]. Thus, metals render the land unsuitable for plant growth and destroy the biodiversity.

Though several regulatory steps have been implemented to reduce or restrict the release of pollutants in the soil, they are not sufficient for checking the contamination. Metal contaminated soil can be remediated by chemical, physical and biological techniques. These can be grouped into two categories [4].

### *Ex-situ method*

It requires removal of contaminated soil for treatment on or off site, and returning the treated soil to the resorted site. The conventional ex-situ methods applied for remediating the polluted soils relies on excavation, detoxification and/or destruction of contaminant physically or chemically, as a result the contaminant undergoes stabilisation, solidification, immobilisation, incineration or destruction.

### *In-situ method*

It is remediation without excavation of contaminated site. Reed et al. defined in-situ remediation technologies as destruction or transformation of the contaminant, immobilisation to reduce bioavailability and separation of the contaminant from the bulk soil [5]. In-situ techniques are favoured over the ex-situ techniques due to their low cost and reduced impact on the ecosystem. Conventionally, the ex-situ technique is to excavate soil contaminated with heavy metal and their burial in landfill site [6, 7]. But the offsite burial is not an appropriate option because it merely shifts the contamination problem elsewhere [7] and also because of hazards associated with the transport of contaminated soil [8]. Diluting the heavy metal content to safe level by importing the clean soil and mixing with the contaminated soil can be an alternative of on-site management [9]. On-site containment and barriers provide an alternative, it involves covering the soil with inert material [10]. Immobilization of inorganic contaminant can be used as a remedial method for heavy metal contaminated soils [11]. This can be achieved by complexing the contaminants, or through increasing the soil pH by liming [12]. Increased pH decreases the solubility of heavy metals like Cd, Cu, Ni and Zn in soil. Although the risk of potential exposure to plants is reduced, their concentration remains unchanged. Most of these conventional remediation technologies are costly to implement and cause further disturbance to the already damaged environment [11,12]. Plant based bioremediation Technologies have been collectively termed as phytoremediation, this refers to the use of green plants and their associated micro biota for the in-situ treatment of contaminated soil and ground water [13]. The idea of using metal accumulating plants to remove heavy

metals and other compounds was first introduced in 1983, but the concept has actually been implemented for the past 300 years [3]. The generic term 'Phytoremediation' consists of the Greek prefix phyto (plant), attached to the Latin root *remedium* (to correct or remove an evil) [14]. This technology can be applied to both organic and inorganic pollutants present in soil (solid substrate), water (liquid substrate) or the air [15,16]. The physico-chemical techniques for soil remediation render the land useless for plant growth as they remove all biological activities, including useful microbes such as nitrogen fixing bacteria, mycorrhiza, fungi, as well as fauna in the process of decontamination [17]. The conventional methods of remediation may cost from \$10 to 1000 per cubic meter. Phytoextraction costs are estimated to be as low as \$ 0.05 per cubic meter [18]. Phytoremediation consists of five main processes, shown in Table 1. This paper focuses studies on the phytoremediation especially phytoextraction of heavy metal contaminated soil using in-situ technique.

### Rhizofiltration

It is defined as the use of plants, both terrestrial and aquatic; to absorb, concentrate, and precipitate contaminants from polluted aqueous sources with low contaminant concentration in their roots. Rhizofiltration can partially treat industrial discharge, agricultural runoff, or acid mine drainage. It can be used for lead, cadmium, copper, nickel, zinc and chromium, which are primarily retained within the roots [19,20]. The advantages of rhizofiltration include its ability to be used as in-situ or ex-situ applications and species other than hyperaccumulators can also be used. Plants like sunflower, indian mustard, tobacco, rye, spinach and corn have been studied for their ability to remove lead from effluent, with sunflower having the greatest ability. Indian mustard has proven to be effective in removing a wide concentration range of lead (4 – 500 mg/l) [21]. The technology has been tested in the field with uranium (U) contaminated water at concentrations of 21-874 ug/l; the treated U concentration reported by Dushenkov was < 20 ug/l before discharge into the environment [22].

**Table 1.** Phytoremediation includes the following processes and mechanisms of contaminant removal

No.	Process	Mechanism	Contaminant
1.	Rhizofiltration	Rhizosphere accumulation	Organics/Inorganics
2.	Phytostabilisation	Complexation	Inorganics
3.	Phytoextraction	Hyper-accumulation	Inorganics
4.	Phytovolatilization	Volatilisation by leaves	Organics/Inorganics
5.	Phytotransformation	Degradation in plant	Organics

### Phytostabilisation

It is mostly used for the remediation of soil, sediment and sludges [20,23] and depends on roots ability to limit contaminant mobility and bioavailability in the soil. Phytostabilisation can occur through the sorption, precipitation, complexation, 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 [24]. 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 [20]. 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 [25]. 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 [26, 27]. Discovery of 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 [28]. Factors such as growth rate, element selectivity, resistance to disease, method of harvesting, are also important [29, 30]. However slow growth, shallow root system, small biomass production, final disposal limit the use of hyperaccumulator species [31]. Phytoextraction studies of Heavy metals have been elaborately discussed later.

### **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 as 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 [23]. 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 [3]. 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 [32]. Phytovolatilization has been successful in tritium ( $^3\text{H}$ ), a radioactive isotope of hydrogen, it is decayed to stable helium with a half-life of about 12 years reported Dushenkov [33]

### **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 [19]. 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 [34]. 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 microorganisms 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 2.

### Total and Bio-available fraction of Heavy Metals in soil

Heavy metals are elements having atomic weight between 63.54 and 200.59, and a specific gravity greater than 4 [35]. Trace amount of some heavy metals are required by living organisms, however any excess amount of these metals can be detrimental to the organisms [36]. Nonessential Heavy metals include arsenic, antimony, cadmium, chromium, mercury, lead, etc; these metals are of particular concern to surface water and soil pollution [35]. Heavy metals exist in colloidal, ionic, particulate and dissolved phase. Metals also have a high affinity for humic acids, organo clays, and oxides coated with organic matter [37,38]. The soluble forms are generally ions or unionised organometallic chelates or complexes. The solubility of metals in soil and groundwater is predominantly controlled by pH [3,4,6] amount of metal [39], cation exchange capacity [40], organic carbon content [37], the oxidation state of the mineral components, and the redox potential of the system [38]. In general, soil pH seems to have the greatest effect of any single factor on the solubility or retention of metals in soils. With a greater retention and lower solubility of metal cations occurring at high soil pH [41]. Under the neutral to basic conditions typical of most soils, cationic metals are strongly adsorbed on the clay fractions and can be adsorbed by hydrous oxides of iron, aluminum, or manganese present in soil minerals. Elevated salt concentration creates increased competition between cations and metals for binding sites. Also competitive adsorption between various metals has been observed in experiments involving various solids with oxide surfaces, in several experiments, Cd adsorption was decreased by the addition of Pb or Cu [42].

**Table 2.** Advantages and disadvantages of phytoremediation.

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

## Plant response to heavy metals

Plants have three basic strategies for growth on metal contaminated soil [16]; see Figure1.

### *Metal excluders*

They prevent metal from entering their aerial parts or maintain low and constant metal concentration over a broad range of metal concentration in soil, they mainly restrict metal in their roots. The plant may alter its membrane permeability, change metal binding capacity of cell walls or exude more chelating substances [43].

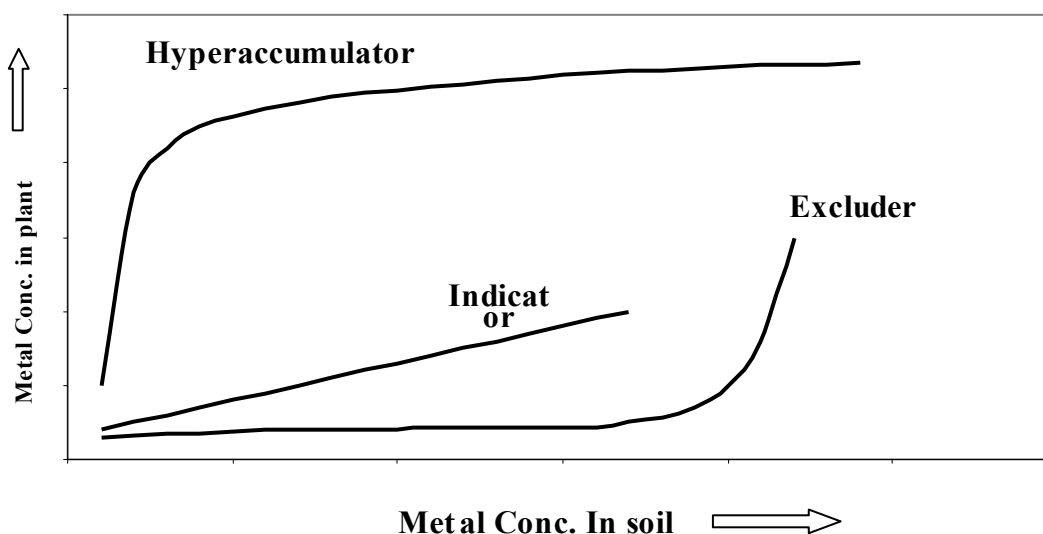
### *Metal indicators*

Species which actively accumulate metal in their aerial tissues and generally reflect metal level in the soil. They tolerate the existing concentration level of metals by producing intracellular metal binding compounds (chelators), or alter metal compartmentalisation pattern by storing metals in non-sensitive parts.

### *Metal accumulator plant species*

They can concentrate metal in their aerial parts, to levels far exceeding than soil. Hyperaccumulators are plants that can absorb high levels of contaminants concentrated either in their roots, shoots and/or leaves [16,29,30]. Baker and Brooks have defined metal hyperaccumulator as plants that contain more than or up to 0.1% i.e. more than (1000 mg/g) of copper, cadmium, chromium, lead, nickel cobalt or 1% (>10,000 mg/g ) of zinc or manganese in the dry matter. For cadmium and other rare metals, it is > 0.01% by dry weight [44]. Researchers have identified hyperaccumulator species by collecting plants from the areas where soil contains greater than usual amount of metals as in case of polluted areas or geographically rich in a particular element [45]. Approximately 400 hyperaccumulator species from 22 families have been identified. The *Brassicaceae* family contains a large number of hyperaccumulating species with widest range of metals, these include 87 species from 11 genera [44].

**Figure 1.** Conceptual response strategies of metal concentrations in plant tops in relation to increasing total metal concentrations in the soil



**Figure. 1** - Conceptual response Strategies of metal concentrations in plant tops in relation to increasing total metal concentrations in the soil

### Mechanism of Phytoextraction

The metal must mobilise into the soil solution, for the plants to accumulate metals from soil. The bioavailability of metals is increased in soil through several means. One-way plants achieve it by secreting phytosidophores into the rhizosphere to chelate and solubilise metals that are soil bound [46]. Both acidification of the rhizosphere and exudation of carboxylates are considered potential targets for enhancing metal accumulation. Following mobilization, a metal has to be captured by root cells. Metals are first bound by the cell wall, it is an ion exchanger of comparatively low affinity and low selectivity. Transport systems and intracellular high-affinity binding sites then mediate and drive uptake across the plasma membrane. Uptake of metal ions is likely to take place through secondary transporters such as channel proteins and/or H<sup>+</sup>-coupled carrier proteins. The membrane potential, that is negative on the inside of the plasma membrane and might exceed -200 mV in root epidermal cells provides a strong driving force for the uptake of cations through secondary transporters [47].

Once inside the plant, most metals are too insoluble to move freely in the vascular system, so they usually form carbonate, sulphate or phosphate precipitates immobilizing them in apoplastic (extracellular) and symplastic (intra cellular) compartments [48]. Unless the metal ion is transported as a non-cationic metal chelate, apoplastic transport is further limited by the high cation exchange capacity of cell walls [48]. The apoplast continuum of the root epidermis and cortex is readily permeable for solutes. Apoplastic pathway is relatively unregulated, because water and dissolved substance can flow and diffuse without having to cross a membrane. The cell walls of the endodermal cell layer act as a barrier for apoplastic diffusion into the vascular system.

In general, solutes have to be taken up into the root symplasm before they can enter the xylem [49]. Subsequent to metal uptake into the root symplasm, three processes govern the movement of metals from the root into the xylem: sequestration of metals inside root cells, symplastic transport into the stele and release into the xylem. The transport of ions into the xylem is generally a tightly controlled process mediated by membrane transport proteins. Symplastic transport of heavy metals probably takes place in the xylem after they cross the casparian strip. It is more regulated due to the selectively permeable plasma membrane of the cells that control access to the symplast by specific or generic metal ion carriers or channels [50]. Symplastic transport requires that metal ions move across the plasma membrane, which usually has a large negative resting potential of approximately 170 mV (negative inside the membrane). This membrane potential provides a strong electrochemical gradient for the inward movement of metal ions. Most metal ions enter plant cells by an energy dependent saturable process via specific or generic metal ion carriers or channels [51].

Non-essential heavy metals may effectively compete for the same transmembrane carriers used by essential heavy metals. Toxic heavy metals such as cadmium may effectively compete for the same transmembrane carrier as used by micronutrient heavy metal. This relative lack of selectivity in transmembrane ion transport may partially explain why non-essential heavy metals can enter cells, even against a concentration gradient. For example, kinetic data demonstrate that essential Cu<sup>2+</sup> and Zn<sup>2+</sup> and non-essential Ni<sup>2+</sup> and Cd<sup>2+</sup> compete for the same transmembrane carrier [52]. Metal chelate complexes may also be transported across the plasma membrane via specialized carriers, as is the case for Fe-phytosiderophore transport in graminaceous species [53]. After heavy metals have entered the root they are either stored in the root or translocated to the shoots. Metal ions can be actively transported across the tonoplast as free ions or as metal-chelate

complexes [54]. It is believed that in order to pass through the casparian strip, water and dissolved ions (salt and metal) require active transport, by utilising energy. For example, Cd is actively transported across the tonoplast of oat roots as either a free ion via a  $\text{Cd}/\text{H}^+$  antiport [55]. The vacuole is an important component of the metal ion storage where they are often chelated either by organic acid or phytochelatins. Insoluble precipitates may form under certain conditions. Precipitation compartmentalisation and chelating are the most likely major events that take place in resisting the damaging effects of metals [56]. Transporters mediate uptake into the symplast, and distribution within the leaf occurs via the apoplast or the symplast [57]. Plants transpire water to move nutrients from the soil solution to leaves and stems, where photosynthesis occurs. Willows, hybrid poplar are also good phytoremediators, because they take up and process large volumes of soil water. For example, data show that a single willow tree, on a hot summer day, can transpire more than 19,000 litres of water [58].

## Types of Phytoextraction

### *Natural Phytoextraction*

In the natural setting, certain plants have been identified which have the potential to uptake heavy metals. At least 45 families have been identified to have hyperaccumulate plants; some of the families are *Brassicaceae*, *Fabaceae*, *Euphorbiaceae*, *Asteraceae*, *Lamiaceae*, and *Scrophulariaceae* [15,33]. Among the best-known hyperaccumulators is *Thlaspi caerulescens* commonly known as alpine pennycress [59], without showing injury it accumulated up to 26,000 mg kg<sup>-1</sup> Zn; and up to 22% of soil exchangeable Cd from contaminated site [60,61]. *Brassica juncea*, commonly called indian mustard, has been found to have a good ability to transport lead from the roots to the shoots. The phytoextraction coefficient for *Brassica juncea* is 1.7 and it has been found that a lead concentration of 500 mg/l is not phytotoxic to *Brassica* species [3]. Phytoextraction coefficient is the ratio of the metal concentration found within the surface biomass of the plant over the metal concentration found in the soil. Some calculations indicate that *Brassica juncea* is capable of removing 1, 1550 kg of lead per acre [3].

On a worldwide basis, concentrations > 1000 mg kg<sup>-1</sup> are known for Ni in more than 320 plant species (sps.), Co (30 sps.), Cu (34 sps.), Se (20 sps.), Pb (14 sps.) and Cd (one sp.). The species involved in hyperaccumulation have recently been tabulated by Reeves and Baker [63], substantial number of these species are from Congo and Zaire. Concentration exceeding 10,000 mg kg<sup>-1</sup> has been recorded for Zn (11 sps.) and Mn (10 sps.). The hyperaccumulation threshold levels of these elements have been set higher because their normal range in plants (20 – 500 mg kg<sup>-1</sup>) are much higher than for the other heavy metals [62]. Aquatic plants such as the floating *Eichhornia crassipes* (water hyacinth), *Lemna minor* (duckweed), and *Azolla pinnata* (water velvet) have been investigated for use in rhizofiltration, phytodegradation, and phytoextraction [27]. Farago and Parsons [64] reported the bioremoval of platinum using *Eichhornia crassipes*. Many aquatic plants are used in the bioremoval of heavy metals e.g. *Azolla filliculoides*, *A. pinnata*, *Typha orientalis* and *Salvinia molesta*. Jin-Hong et al. in their study of twelve wetland species reported, *Polygonum hydropiperoides* Michx (smartweed) as the best for heavy metal phytoremediation, due to its faster growth and high plant density [65]. Recently, a fern *Pteris vitatta* has been shown to accumulate as much as 14,500 mg kg<sup>-1</sup> arsenic in fronds without showing symptoms of toxicity [66].



### ***Induced Phytoextraction or Chelate assisted Phytoextraction***

Within the plant cell heavy metal may trigger the production of oligopeptide ligands known as phytochelatin (PCs) and metallothioneins (MTs) [67]. These peptides bind and form stable complex with the heavy metal and thus neutralise the toxicity of the metal ion [68]. Phytochelatin (PCs) is synthesised with glutathione as building blocks resulting in a peptide with structure Gly-( $\gamma$ -Glu-Cys-) $_n$ ; {where,  $n = 2-11$ }. Appearance of phytochelating ligands has been reported in hundreds of plant species exposed to heavy metals [69]. Metallothioneins (MTs), are small gene encoded, Cys-rich polypeptides. PCs are functionally equivalent to MTs [68].

Chelators have been isolated from plants that are strongly involved in the uptake of heavy metals and their detoxification. Chelating agents like ethylenediamine tetra acetic acid (EDTA) are applied to Pb contaminated soils that increases the amount of bioavailable lead in the soil and a greater accumulation in plants is observed [70]. The addition of chelates to a lead contaminated soil (total soil Pb 2500 mg kg<sup>-1</sup>) increased shoot lead concentration of *Zea mays* (corn) and *Pisum sativum* (pea) from less than 500 mg kg<sup>-1</sup> to more than 10,000 mg kg<sup>-1</sup>. This was achieved by adding synthetic chelate EDTA to the soil, similar results using citric acid to enhance uranium uptake have been documented. These results indicate that chelates enhanced or facilitated Pb transport into the xylem, and increased lead translocation from roots to shoots. For the chelates tested, the order of effectiveness in increasing Pb desorption from the soil was EDTA > Hydroxyethylethylene-diaminetriacetic acid (HEDTA) > Diethylenetriaminepentaacetic acid (DTPA) > Ethylenediamine di(o-hydroxyphenylacetic acid) EDDHA [70]. Vassil et al., [71] reported that *Brassica juncea* exposed to Pb and EDTA in hydroponic solution was able to accumulate up to 55 mM kg<sup>-1</sup> Pb in dry shoot tissue (1.1% [w/w]). This represents a 75-fold concentration of lead in shoot over that in solution. A threshold conc. of EDTA (0.25 mM) was required to stimulate this dramatic accumulation of both lead and EDTA in shoots.

### ***Genetic Engineering to improve phytoremediation***

To breed plants having superior phytoremediation potential with high biomass production can be an alternative to improve phytoremediation. General plant productivity is controlled by many genes and difficult to promote by single gene insertion. Genetic engineering techniques to implant more efficient accumulator gene into other plants have been suggested by many authors [29,60,72]. Implanting more efficient accumulator genes into other plants that are taller than natural plants increases the final biomass. Zhu et al. [73] genetically engineered *Brassica juncea* to investigate rate-limiting factors for glutathione and phytochelatin production; they introduced the *Escherichia coli* -gshl- gene. The  $\gamma$ -ECS transgenic seedlings showed increased tolerance to cadmium and had higher concentrations of Phytochelatin,  $\gamma$ -GluCys, glutathione, and total nonprotein thiols compared to wild type seedlings. The potential of success of genetic engineering can be limited because of anatomical constraints [74].

### **Limitations of Phytoextraction**

Phytoextraction and plant-assisted bioremediation is most effective if soil contamination is limited to within 3 feet of the surface, and if groundwater is within 10 feet of the surface [16, 18]. It is applicable to sites with low to moderate soil contamination over large areas, and to sites with large volumes of groundwater with low levels of contamination that have to be cleaned to low (strict) standards [26]. This necessitates soil fertilization, conditioning, importance of employing effective agronomic practices [70, 72]. Scientists have investigated the effect of soil acidification on Zn and Cd phytoextraction and proposed the

use of  $(\text{NH}_4)_2\text{SO}_4$  as a soil additive to provide nutrients (N and S) needed for high yield, and to acidify the soil for greater metal bioavailability. However, there might be some negative side effects associated with soil acidification. For example, due to increased solubility some toxic metals may leach into the groundwater creating an additional environmental risk. Chaney et al. [72] indicated that following metal phytoextraction, soil could be limed to elevate the pH near a neutral value, so that normal farm uses or ecosystem development could resume. However, premature liming may increase soil capacity for metal binding and restrict the potential for phytoextraction. A similar effect can be expected following the addition of organic fertilizers [69]. Phosphorus is a major nutrient, and plants respond favorably to the application of phosphate fertilizer by increasing biomass production [71]. The addition of these fertilizers, however, can also inhibit the uptake of some major metal contaminants, such as Pb, due to metal precipitation as pyromorphite and chloropyromorphite [72]. Natural chelators of plants or microbial origin seem more promising than synthetic chemical chelators [69]. It is uncertain whether an approach based on chemical chelators is practical for improving phytoextraction, since chemical chelators have additional toxicity to plants, thus they may increase the uptake of metals but decrease plant growth thus proving to be of limited benefit.

### **Utilization of Phytoremediation by-product**

Phytoextraction involves repeated cropping of plants in contaminated soil, until the metal concentration drops to acceptable level. The ability of the plants to account for the decrease in soil metal concentrations as a function of metal uptake and biomass production plays an important role in achieving regulatory acceptance. Theoretically, metal removal can be accounted by determining metal concentration in plant, multiplied by the biomass produced; and comparing this with the reduction in soil metal concentrations. Although this sounds simple, many factors make it challenging in the field. One of the hurdles for commercial implementation of phytoextraction has been the disposal of contaminated plant material. After each cropping, the plant is removed from the site; this leads to accumulation of huge quantity of hazardous biomass. This hazardous biomass should be stored or disposed appropriately so that it does not pose any risk to the environment.

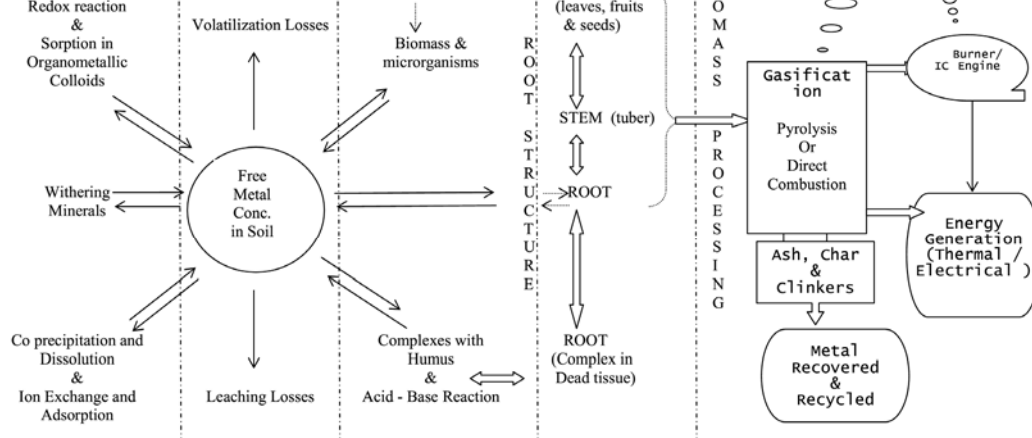
Biomass is nothing but stored solar energy in plant mass, it is also termed as materials having combustible organic matter. Biomass contains carbon, hydrogen and oxygen, it is known as oxygenated hydrocarbons. Biomass (specially wood) can be represented by the chemical formula  $\text{CH}_{1.44}\text{O}_{0.66}$  [75]. The main constituents of any biomass material are lignin, hemicellulose, cellulose, mineral matter and ash. It possesses high moisture and volatile matter constituents, low bulk density and calorific value. The percentage of these components varies from species to species. The dry weight of *Brassica juncea* for induced phytoextraction of lead amounts to 6 tonnes per hectare with 10,000 to 15,000 mg/kg of metal in dry weight [76]. Handling of huge quantity of this type of waste is a problem and hence need volume reduction [77].

Composting and compaction has been proposed as post harvest biomass treatment by some authors [48,78,79]. Leaching tests for the composted material showed that the composting process formed soluble organic compounds that enhanced metal (Pb) solubility. Studies carried out by Hetland et al., [80] showed that composting can significantly reduce the volume of harvested biomass, however metal contaminated plant biomass would still require treatment prior to disposal. Total dry weight loss of contaminated plant biomass by compaction is advantageous, as it will lower cost of transportation to a hazardous waste disposal facility. Compaction of harvested plant material was proposed by Blaylock and Huang [77] for processing metal rich phytoextraction residue.

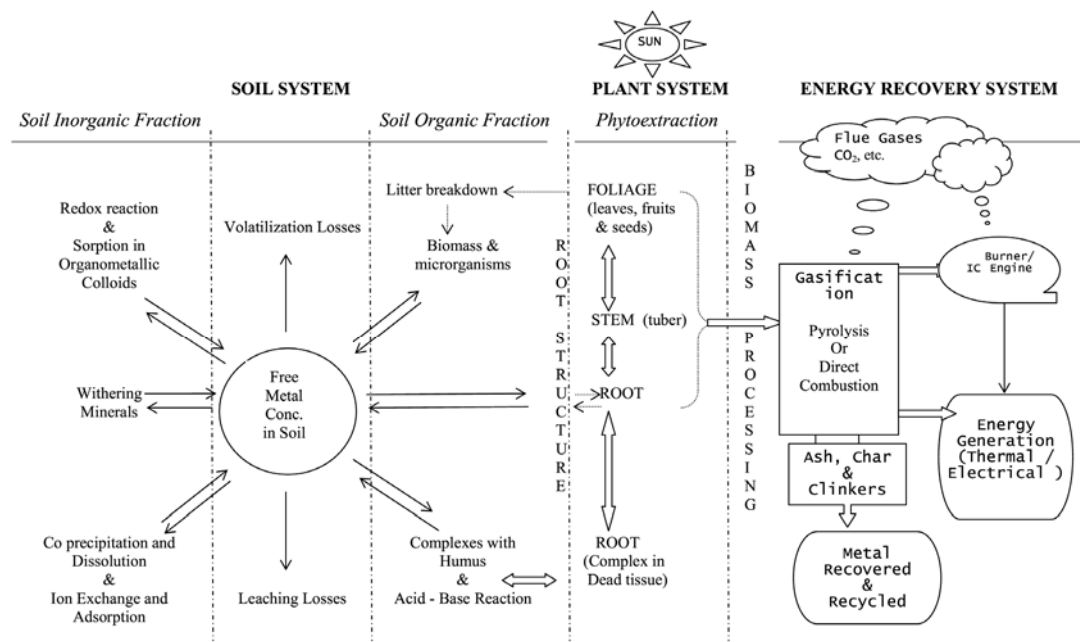
Advantages of compaction are similar as composting, the leachate will need to be collected and treated appropriately; in comparison to composting there is little information on compaction. One of the conventional and promising routes to utilize biomass produces by phytoremediation in an integrated manner is through thermochemical conversion process. If phytoextraction could be combined with biomass generation and its commercial utilization as an energy source, then it can be turned into profit making operation and the remaining ash can be used as bio-ore [28], this is also the basic principle of phytomining. Nicks and Chambers [81], reported a second potential use for hyperaccumulator plants for economic gain in the mining industry. This operation, termed phytomining includes the generation of revenue by extracting saleable heavy metals produced by the plant biomass ash, also known as bio-ore.

Combustion and gasification are the most important sub routes for organized generation of electrical and thermal energy. Recovery of this energy from biomass by burning or gasification could help make phytoextraction more cost-effective. Thermochemical energy conversion best suits the phytoextraction biomass residue because it cannot be utilized in any other way as fodder and fertilizers. Combustion is a crude method of burning the biomass, but it should be under controlled conditions, whereby volume is reduced to 2–5 % and the ash can be disposed properly. This method of plant matter disposal is often mentioned by many authors [48, 82]. It will not be favourable to burn the metal bearing hazardous waste in open, as the gases and particulates released in the environment may be detrimental; only the volume is reduced and the heat produced in the process is wasted. Gasification is the process through which biomass material can be subjected to series of chemical changes to yield clean and combustible gas at high thermal efficiencies. This mixture of gases called as producer gas and/or pyro-gas that can be combusted for generating thermal and electrical energy. The process of gasification of biomass in a gasifier is a complex phenomenon; it involves drying, heating, thermal decomposition (pyrolysis) and gasification, and combustion chemical reactions, which occurs simultaneously [75]. Hetland et al., [80] reported possibility of co-firing plant biomass with coal, the results suggested that ashing reduced the mass of lead contaminated plant material by over 90% and partitioned lead into ash. It may be possible to recycle the metal residue from the ash, however there are no estimates of the cost or feasibility of such a process [48]. Future experiments should concentrate on development of combustion system and methods to recycle different metals from ash. The process destroys organic matter, releasing metals as oxides. The liberated metals remain in the slag, modern flue gas cleaning technology assures effective capture of the metal containing dust. Considering the other technologies for disposal this method is environment-friendly.

Bridgewater et al., [82] reported that pyrolysis is a novel method of municipal waste treatment that might also be used for contaminated plant material. Pyrolysis decomposes material under anaerobic conditions; there is no emission to the air. The final products are pyrolytic fluid oil and coke; heavy metals will remain in the coke, which could be used in smelter. Koppolu et al., [83] reported that 99% of the metal recovered in the product stream was concentrated in the char formed by pyrolysing the synthetic hyperaccumulator biomass used in the pilot scale reactor. The metal component was concentrated by 3.2–6 times in the char, compared to feed. Study of the fate of the metals in various feeds during pyrolysis has been addressed in literature in different context, but results on pyrolysis of phytoextraction plant biomass are limited. Helson et al., [84] conducted low temperature pyrolysis experiments with chromium, copper and arsenate treated wood and it was concluded that most of the metal was retained in the pyrolysis residue. Influence of metal ions on the pyrolysis of wood has been studied extensively by many authors [85, 86]



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The result of their work showed that phytoextraction of Cd, Cr and Pb by *Ipomoea carnea*, *Datura innoxia* and *Phragmites karka* was higher in comparison to *Brassica juncea* and *Brassica campestris*, (known as indicator species) [3,88]. The study conducted with 10 to 200 mg kg<sup>-1</sup> of Cd, Cr and Pb (separately) indicated that *I. carnea* was more effective in extracting them from soil than *B. juncea*. Among the five species, *B. juncea* accumulated maximum Cd but *I. carnea* followed by *D. innoxia* and *P. karka* were the most suitable species for phytoextraction of cadmium, if the whole plant or above ground biomass is harvested. In the relatively short time, *I. carnea* produced more than five times more biomass in comparison *B. juncea* [89]. It was more effective at translocating Cr from soil to plant shoot. *P. karka* showed much greater tolerance to chromium than other plants, though the uptake was low. *Ipomoea* extracted maximum lead at 200 mg kg<sup>-1</sup>; *Datura* and *Phragmites* was best extractor at 100 mg kg<sup>-1</sup>, whereas Brassica species were at 50 mg Pb kg<sup>-1</sup> soil [90]. Brassica species were difficult to cultivate, as they required pesticides to protect them from army moth, and secondly they cannot grow throughout the year. Whereas high biomass species do not have these limitations and showed higher potential, the extraction capacity can be further increased by use of chelates or soil additives.

### Future of Phytoremediation

One of the key aspects to the acceptance of phytoextraction pertains to the measurement of its performance, ultimate utilization of by-products and its overall economic viability. To date, commercial phytoextraction has been constrained by the expectation that site remediation should be achieved in a time comparable to other clean-up technologies. So

far, most of the phytoremediation experiments have taken place in the lab scale, where plants grown in hydroponic setting are fed heavy metal diets. While these results are promising, scientists are ready to admit that solution culture is quite different from that of soil. In real soil, many metals are tied up in insoluble forms, and they are less available and that is the biggest problem, said Kochian [59]. The future of phytoremediation is still in research and development phase, and there are many technical barriers which need to be addressed. Both agronomic management practices and plant genetic abilities need to be optimised to develop commercially useful practices. Many hyperaccumulator plants remain to be discovered, and there is a need to know more about their physiology [16]. Optimisation of the process, proper understanding of plant heavy metal uptake and proper disposal of biomass produced is still needed.

## Conclusion

Phytoremediation is a fast developing field, since last ten years lot of field application were initiated all over the world, it includes Phytoremediation of Organic, Inorganic and Radionuclides. This sustainable and inexpensive process is fast emerging as a viable alternative to conventional remediation methods, and will be most suitable for a developing country like India. Most of the studies have been done in developed countries and knowledge of suitable plants is particularly limited in India. In India commercial application of Phytoremediation of soil Heavy metal or Organic compounds is in its earliest phase. Fast growing plants with high biomass and good metal uptake ability are needed. In most of the contaminated sites hardy, tolerant, weed species exist and phytoremediation through these and other non-edible species can restrict the contaminant from being introduced into the food web. However, several methods of plant disposal have been described but data regarding these methods are scarce. Composting and compaction can be treated as pre-treatment steps for volume reduction, but care should be taken to collect leachate resulting from compaction. Between the two methods that significantly reduce the contaminated biomass, incineration seems to be least time consuming and environmentally sound than direct burning or ashing.

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