

Phytoremediation of Heavy Metals: Techniques, Challenges, and Prospects

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Abstract

Heavy metals, characterized by their high atomic mass and density, can pose significant risks to soil, water, plants, and human health. Contamination sources include manufacturing activities, mining, farming practices, and improper waste management. Metals such as arsenic, mercury, lead, chromium, and cadmium are most toxic with health consequences that can result from organ dysfunction to cancer. Conventional remediation techniques usually face challenges in due to high costs and secondary pollution. Phytoremediation, an eco-friendly alternative, uses plants to absorb, stabilize, or degrade toxic metals in contaminated environments. Among the techniques found to effectively mitigate soil and water pollution are phytoextraction, phytostabilization, phytovolatilization, and rhizofiltration. On the other hand, progress in genetic engineering and the integration of plant growth-promoting rhizobacteria (PGPR) has led to a greater efficiency of phytoremediation. Nevertheless, problems such as prolonged remediation duration and poor remediation performance in heavily contaminated environments still present. This review discusses the technique, applications and developments of phytoremediation, providing insight into its utility for environmentally sustainable management.

Keywords: Phytoremediation, Heavy Metals, Contaminants, Hyperaccumulator plants, Phytoextraction

1. Introduction

Heavy metals are those elements with high atomic number, atomic weight, and density that naturally form an integral part of the Earth's geological systems. The impact of these metals on plants, animals and microbes depends on their geochemical availability and physical-chemical characteristics (1). Certain metals are essential for specific biological functions, while others remain harmless and inactive. However, some metals can be extremely toxic, even in trace amounts (2). Arsenic, mercury, lead, chromium, and cadmium are toxic heavy metals that bind to the macromolecules of human systems via the different routes of entry (3). Heavy metals often enter soil ecosystems through natural processes like geogenic contamination, as well as human activities such as air-borne deposition, waste disposal, industrial effluents, farming practices, and mining (4,5). Arsenic, fluoride, and uranium contamination is often linked to their natural presence in various regions worldwide. Additionally, atmospheric deposition, whether dry or wet, serves as another significant source of volatile toxic metals in the ground (6). Using wastewater from municipal sources for irrigation sewage along with effluents causes heavy metal contamination in agricultural lands as well (7,8). Moreover, the uses of fertilizers, insecticides, and sewage sludge in agricultural fields leads to soil contamination with heavy metals (9). Mining activities, for example, coal mining, increase metal concentration in the adjacent soils. Vehicular emissions are one of the significant factors polluting the soil on and around roads and highways (10). Improper disposal of solid and electronic waste further increases toxic metal pollution in the ground (11,12).

Metals can be toxic even in trace amounts, so their buildup in the surrounding impose threats to flora, fauna, and humans. (9,13,14). In a number of cases, metal-polluted soil completely lacks vegetation which causes intense soil degradation problems and off-site environmental contamination e.g., contamination of groundwater resources (15). Metal toxicity also affects soil microbes, changing extant microbial communities, depleting their numbers, and inhibiting their functions (16,17). Immediate and prolonged metal(loid) contact with humans results in a number of negative health impact including skin diseases, heart diseases, irritating behavior, lack of concentration, harm to the neurological and immune systems, gastrointestinal and kidney issues, along with cancer and other complications, highlighting the need for the eradication of harmful metals found in the ground to mitigate the harmful impact on surrounding and public health (18,19).

Different techniques are used to eradicate harmful metals from ground, such as precipitation, digging up soil, electro-remediation, leaching with chemicals, thermal remediation, soil flushing, landfill disposal, and immobilization techniques etc. However, these techniques still suffer from some drawbacks, including the high cost, low efficiency at low concentrations of pollutant, and the formation of secondary pollutants, as well as irreversible alteration in soil physicochemical and biological properties (20–23). Phytoremediation, a sustainable method regarding the capabilities of plants to move, stabilize, and/or bioremediate pollutants, making them environmentally inert. Using this method, it is possible to eradicate contaminants from soils and

aquatic environments, therefore contributing to environmental decontamination. Common contaminants addressed by phytoremediation include heavy metals, organic pollutants (24), and radionuclides (25). The method consists of phytoextraction, phytodegradation, phytostabilization, Phytovolatilization and rhizofiltration techniques (26,27). Phytoremediation is a economical and ecological solution to conventional remediation techniques such as excavation, incineration, leaching and landfilling (28–30). Yet this approach is usually much slower compared to classic approaches that may take years or even a lifetime to finish. Its success in remediating heavy metals is heavily dependent on the development and viability of plants used in the process (31,32).

2. Toxicity of heavy metal

The escalating emission of heavy metals into soil and water as a result of the urbanization and industrialization phenomenon is a great concern. These heavy metals affect plants, animals, and humans in different ways including inhibiting photosynthesis in plants, leading to serious human diseases, and decreasing microbial cell count and growth (33). Each heavy metal, including Pb, Cr, Cd, As and Hg possesses unique toxicity levels, effects, and acceptable limits.

2.1 Arsenic

Arsenic can be acquired by the ingestion of contaminated groundwater or some foods, such as seafood, rice, mushrooms, and poultry (1,34). Accumulations of organic arsenic compounds arsenobetaine and arsenocholine, found in shellfish and predatory fish can increase urinary arsenic level (35). Coming in contact with arsenic can result in both short-term and prolonged toxicity. Acute poisoning, which can be deadly, typically occurs at doses between 100 – 300 milligrams. For inorganic arsenic, the estimated lethal dose for humans is approximately 0.6 mg per kilogram of body mass each day (36). Prolonged contact with high levels of inorganic arsenic, generally through ingestion, can lead to initial signs such as alterations in skin color, Skin abnormalities and thickened areas on the hands and feet which may appear after five years and could indicate the onset of skin cancer (37). Diarrhea is one of the first signs of acute arsenic poisoning and tends to develop quickly. In cases of chronic exposure, it is often accompanied by vomiting (36). A variety of factors affect the toxicity of arsenic, such as its ability to dissolve, oxidation state, and various internal and external characteristics (38). Several factors affect arsenic's toxic effects, including the frequency and duration of exposure, age, gender, and individual sensitivity, with genetic and nutritional influences also being significant (39). Arsenic poisoning can denature as many as 200 enzymes, mostly related to enzymes that catalyzes DNA replication repair, cellular energy production and ATP metabolism. Moreover, unbound arsenic can stimulate the production of reactive oxygen species, which further induces DNA damage and lipid peroxidation (36,40).

2.2 Mercury

Mercury is among the most hazardous and bioaccumulative compounds and the mercury contamination is well addressed in aquatic ecosystems due to its toxic effects on marine organisms. Major mercury pollution sources are human activities compromising mining, industrial and agricultural runoff, waste incineration, and municipal wastewater discharges (41). While mercury is not essential, it is the most harmful metal to humans, existing in 3 types: elemental, inorganic, and organic mercury. Elemental mercury is primarily released into the atmosphere as vapor (42). This vapor affects the brain and nerves, leading to problems with thinking, movement, and sensing. It shows up as difficulty sleeping, forgetfulness, hand shakiness, and muscle spasms. Long term exposure may result in concentration difficulties, Distorted vision along with difficulty walking. Elevated mercury exposure can lead to severe Nervous system damage and fatality. Additionally, Exposure to mercury during pregnancy can harm the unborn baby, potentially causing brain damage, vision loss, developmental delays, and speech problems. Inhaling mercury vapor can also lead to lung issues, including inflammation, fluid buildup, and other respiratory damage. At lower exposure levels, adults may experience mood swings, shaking hands, skin irritation, and forgetfulness, while children may develop skin redness and peeling (43). Mercury is commonly used in dental amalgams, where around 50% of the material is metallic mercury. Over time, mercury is emitted as gas or inorganic charge particles from the amalgam surface due to abrasion. These vapors can be inhaled, while the metal ions can be ingested through the gastrointestinal tract. After absorption, mercury circulates throughout the body through the bloodstream (44). It also contributes to lipid peroxidation and disrupts calcium regulation by inducing Impaired mitochondrial function and increased oxidative stress. Mercury acts like catalyst for Fenton-type reactions, increasing the levels of reactive oxygen species (ROS). Organic mercury poisoning, often caused by consuming contaminated fish, has been responsible for outbreaks such as Minamata disease in Japan and Iraq. Of the organic mercury compounds, dimethyl mercury is much more toxic than methyl mercury, with even a small amount spilled on the skin potentially causing death (45).

2.3 Lead

Lead is a bluish-gray metal that naturally exists in small quantities within the earth's crust. Although it is a natural element, anthropogenic activities including mining, combustion fossil fuels, as well as industrial processes have significantly raised its concentration in the surroundings. Lead is widely used in several sectors, including industry, agriculture, and household applications. Its primary uses today include manufacturing lead-acid batteries, Firearms, metallic goods such as pipes and solder, and X-ray protection devices (46). Acute lead toxicity is marked by signs like stomach cramps, nausea, diarrhea, nerve damage, swelling, and confusion, which could progress to shaking and even death. Kids are more vulnerable to serious lead toxicity than adults as their bodies are still developing and they absorb more lead. (47). With

long-term exposure, lead gradually builds up in the bones and eventually in the kidneys. Acute lead exposure can cause symptoms like tiredness, trouble sleeping, headaches, anemia, confusion, and difficulty speaking clearly. In children, prolonged exposure often results in behavioral changes, such as irritability and a lack of interest in playing (48). Lead negatively impacts vital hormonal and nervous systems, influencing heart rate, blood vessel function, and blood flow. Research in animals has connected lead-induced high blood pressure in rats with reduced levels of nitric oxide, a crucial regulator of blood pressure. Research on drinking water has demonstrated that lead exposure causes cell death by triggering the release of mitochondrial proteins involved in cell death, inhibiting proteins that prevent cell death, and activating enzymes responsible for inducing cell death in kidney cells of treated rats (47). Furthermore, lead exposure increased the levels of vasoconstrictor peptide, nitrogen oxide, and eosinophil enzyme in the blood, while also causing lung damage in both ovalbumin-sensitive and non-sensitive guinea pigs (49). Lead disrupts heme production by inhibiting key enzymes, ferrochelatase and alpha-aminolevulinic acid dehydratase (ALAD), leading to anemia. This inhibition reduces heme synthesis, impairing red blood cell formation and function (50).

2.4 Cadmium

Cadmium (Cd) is a manufacturing waste product emerging from the refining process of Cu, Pb and Zn. Cadmium is found in rechargeable batteries, certain alloys, and is also found in Cigarettes or smoking products and emissions from burning fossil fuels. Tobacco products, is a primary source of cadmium contact, and it can also be present in products like e-cigarettes. For non-smokers, food—especially leafy greens, fruits, cereals, along with organ meats such as the heart, liver, and kidneys—serves as a significant source of cadmium (1). Cadmium is also used in Coatings, dyes, metal plating, and as a plastic stabilizer. The main routes of cadmium exposure for individuals are through breathing it in and consuming it. (51). Cadmium primarily affects the kidneys, lungs, and bones, making them the most vulnerable to its toxic effects (52). In the kidneys, cadmium binds with a protein called metallothionein, causing serious damage and leading to chronic kidney problems (53). People who work with cadmium-containing vapors face an added risk of severe lung problems, such as acute respiratory distress. Once inhaled, cadmium usually enters the bloodstream by forming complexes with a protein called cysteine (54). Beyond that, cadmium also impacts bone health by reducing bone mineral content, which raises the likelihood of bone fractures. It interferes with the natural balance of bone-building and breakdown, suppressing the formation of new bone tissue while increasing the breakdown of existing bone. This disruption can make bones weaker over time (18).

2.5 Chromium

Chromium presents in the ecosystem in oxidation states varying between Cr(II) to Cr(VI). Primary sources of chromium include coal and petroleum combustion, ferrochromate petroleum,

colorant, oxidizing agent, catalysts, chromium steels, agricultural supplements and oil well drilling, and tanneries engaged in electroplating (55). Cr(III) is generally considered safe to inhale because of its low membrane penetration ability. In contrast, Cr(VI) is more adept at penetrating cell membranes by utilizing channels designed for anions with identical electronic configurations and structural arrangements, such as SO_4^{2-} and HPO_4^{2-} , and can also enter cells via phagocytosis. Chromium(VI) is a powerful oxidant that can be reduced intracellularly to temporary forms, including pentavalent and tetravalent chromium. This reduction process is thought to detoxify Cr(VI) when it occurs away from target sites, with glutathione stabilizing the five-electron valence configuration. However, When biological reducing agents like thiols and ascorbate come into contact with Cr(VI), Oxygen-derived radicals like O_2^- , H_2O_2 , and $\bullet\text{OH}$. are generated. These ROS trigger oxidative stress, leading to cellular damage by harming proteins and DNA (56).

3. Phytoremediation Technique

Phytoremediation is an eco-friendly and cost-efficient method of waste management that uses living organisms to break down toxic substances into less harmful or harmless forms, without creating dangerous by-products. This process relies on the natural abilities of specific plants, known as hyperaccumulators, to absorb and remove pollutants such as heavy metals, pesticides, and polyaromatic hydrocarbons within ground, water and the atmosphere. The success of this approach depends on several factors, including environmental conditions, nutrient availability, the type of pollutants present, and the surrounding habitat (57).

3.1 Phytostabilization

Phytostabilization or phytoimmobilization , involves limiting the spread of contaminants in soil via the settling of pollutants in the rhizosphere or absorption by plant roots (58). This process includes several mechanisms, beginning with the adsorption of toxic metals along with other contaminants onto root surfaces via electrostatic forces and associations with substances released by the roots. After being absorbed into root cells, these pollutants are either stored contained within vacuoles as well as attached to the compounds of the cell wall, effectively limiting their movement within the plant (59). In the soil encompassing plant roots, the area driven by root activity, plants release natural substances referred to as root exudates. These substances have a vital role in immobilizing contaminants through the production of insoluble compounds with metals. Additionally, they enhance microbial activity, enabling microbes to change the chemical characteristics of pollutants, effectively minimizing their mobility and toxicity. (60,61). Additionally, plants adjust soil pH by releasing organic acids or absorbing positively and negatively charged ions, resulting in the precipitation of heavy metals, decreasing their solubility and bioavailability. Plants also influence soil redox conditions; Roots that release

oxygen have the ability to facilitate the oxidation of specific contaminants, further limiting their mobility (62,63).

3.2 Rhizofiltration

Rhizofiltration uses plant roots to take up as well as capture metal contaminants from water. The technique is especially effective in removing metals including Cd, Cr, Ni, Pb along with radioactive elements like uranium (U), strontium (Sr) as well as cesium (Cs) (64,65). Long-rooted trees are key players in this process, functioning like natural pumps that draw large amounts of water from beneath the surface. As the roots absorb water, they also take in the contaminants. Furthermore, substances released by roots, for example citric acid and malate, may boost the process by facilitating the uptake, attachment or precipitation of these contaminants. (66–68). For instance, research has shown that *Zea mays* (maize) can reduce mercury (Hg) levels by 12%, lead (Pb) by 32%, and chromium (Cr) by 30% (69). Another standout example is *Typha angustifolia*, an aquatic plant, which has an impressive ability to accumulate between 4,941.1–14,109.4 milligram of cadmium (Cd) and 14,039.3–59,360.8 milligram of zinc (Zn) per plant. Due to elevated bioconcentration factor (BCF) and minimal translocation factor (TF), *Typha angustifolia* is an excellent choice for phytoremediation efforts (70). Other commonly used aquatic plants, such as *Pistia*, *Azolla* as well as *Eichhornia*, also show remarkable potential in cleaning up pollutants. *Pistia* excels at extracting and stabilizing pollutants like As, Pb. *Eichhornia*, *Azolla* are particularly good at absorbing Ni and Cu from polluted aquatic ecosystem. Notably, *Pistia* has a translocation factor of 5.0 for fluoride, making it an exceptional hyperaccumulator for this contaminant (68).

3.3 Phytoextraction

Phytoextraction refers to a method where plants are used to clean up contaminated soil by absorbing pollutants through their roots and storing them in their above-ground parts, such as stems and leaves. These "pollutant-accumulating plants" are particularly effective at removing metals and other pollutants, providing a natural and sustainable way to remediate polluted soil and water. In some cases, pollutants may also be absorbed and processed through the plant's roots (71). The performance of this process is determined by how well a plant can intake and store metals in its aerial tissues and how accessible the metals are for absorption. Researchers are working to better understand the genetic along with biochemical mechanisms that enable certain plants, called hyperaccumulators, to take in, transport, and store metals. This knowledge is being used to develop genetically engineered plants with improved abilities for phytoremediation (72). A specialized method called chelant-enhanced phytoextraction involves adding chelating agents to the ground, which increases the availability of metals, making it easier for plants to absorb and remove them. However, the success of this technique depends on conditions like the type of

metal, the plant species, and the amount of chelant used (73). Studies on the phytoextraction potential of plants like canola and radish show that both can tolerate heavy metals to a moderate degree. Radish has slightly better tolerance compared to canola, but neither plant is highly effective at dealing with soils that contain multiple contaminants (74).

3.4 *Phytovolatilization*

Phytovolatilization is a natural cleanup process where plants absorb contaminants present in the underground and transform them into less harmful gaseous states, and discharge those into the air via their leaves while transpiration. This method is especially efficient in eliminating organic pollutants along with specific toxic metals such as arsenic, selenium and mercury through detoxification processes (75). For instance, plants from the Brassicaceae family, like *Brassica juncea*, are recognized for their capacity to volatilize selenium. They convert inorganic selenium into organic amino acids like selenocysteine and selenomethionine, which are converted into a less toxic gaseous state called dimethylselenide and discharged into the atmosphere (76,77). Mercury, a metal that can vaporize easily at room temperature, is another contaminant that plants can help mitigate. In its common environmental form, mercury often exists as a reactive cation, Hg^{2+} . Plants can intake methylmercury via their roots or leaves, convert it into elemental mercury, and release it as a gas, reducing its toxicity (78,79). One major advantage of phytovolatilization is that it eliminates heavy metal contaminants present in the underground without the need for removal or harvesting of the plants. However, while it reduces soil contamination, it doesn't entirely eradicate the contaminants from the ecosystem. Instead, it transfers them to the atmosphere, where volatile substances could contribute to air pollution or return to the soil through rainfall. Because of these potential risks, it's essential to conduct a thorough hazard assessment before implementing phytovolatilization in a specific location (80).

3.5 *phytodegradation*

Phytodegradation also known as phytotransformation, involves the breaking down of compounds absorbed by plants via their natural metabolic activities or by the action of enzymes secreted by their roots (81). Plants release enzymes like dehalogenase, oxygenase, and peroxidase that help converting organic pollutants like polycyclic hydrocarbons, into simpler, less toxic substances. These enzymes facilitate oxidation-reduction reactions, transforming pollutants either inside the plant tissues or within the area of soil surrounding plant roots (rhizosphere). This method has proven to be effective in degrading contaminants including polycyclic aromatic hydrocarbons and pesticides. For instance, research has shown that peroxidase enzymes in plants such as *Populus* species can break down organic contaminants in polluted soils. One benefit of phytodegradation is that it can work well in soils with low microbial activity, allowing plants to handle higher levels of pollutants that would typically be

harmful to microbes. However, a challenge with this method is the difficulty in tracking the by-products formed within the plant tissues, which can make it harder to confirm that the pollutants have been fully degraded (82–84).

Table 1: Some potential plant species for phytoremediation

Heavy Metal	Plant Species	Reference
As	<i>Pteris vittata</i> ,	(85)
	<i>Brassica Juncea</i>	(86)
	<i>Pteris biaurita</i>	(87)
	<i>Corrigiola telephiifolia</i>	(88)
Hg	<i>Juncus maritimus</i>	(89)
	<i>Marrubium vulgare</i>	(90)
	<i>Nephrolepis exaltata</i>	(91)
	<i>Macleaya cordata</i>	(92)
	<i>Poa pratensis</i>	(93)
	<i>Salix viminalis</i>	(94)
	<i>Silene vulgaris</i>	(94)
Pb	<i>Betula occidentalis</i>	(95)
	<i>Brassica juncea</i>	(95)
	<i>Paulownia tomentosa</i>	(96)
	<i>Deschampsia cespitosa</i>	(97)
	<i>Helianthus annuus</i>	(98)
Cd	<i>Boehmeria nivea</i>	(99)
	<i>Canna indica</i>	(100)
	<i>Lathyrus sativus</i>	(101)
	<i>Lagerstroemia indica</i>	(102)
	<i>Noccaea caerulescens</i>	(103)
Cr	<i>Genipa americana</i>	(104)
	<i>Brassica juncea</i>	(105)
	<i>Hydrocotyle umbellata</i>	(106)
	<i>Pistia sp.</i>	(107)

4. Advantages and Limitations

Phytoremediation has become widely recognized as an effective solution for reclaiming polluted and degraded sites, particularly those contaminated with heavy metals. This environmentally friendly and cost-efficient method is far more affordable than many traditional approaches to addressing heavy metal pollution (108). Beyond cleaning up contaminated soils, phytoremediation offers the added benefit of recovering valuable metals found within the earth, which can be recycled and repurposed for various applications. Research has even highlighted its potential to extract radionuclides and radioactive pollutants (109,110). Since phytoremediation works directly in the polluted region, it minimizes soil disruption and helps preserve the topsoil, unlike invasive remediation methods. This makes it ideal for restoring large areas of polluted soil, sediments, and groundwater at a small portion of the expense at a small portion of the expense. Additionally, as a green technology, phytoremediation supports afforestation efforts while contributing to carbon reduction by capturing atmospheric carbon dioxide through photosynthesis (111).

Phytoremediation offers great potential for addressing heavy metal contamination, but it does come with some challenges. One of the main limitations is the time it takes to see results. Most studies are conducted in controlled environments over relatively short periods, which may not accurately represent the long-term effectiveness of the technique in real-world conditions. This highlights the need for more extensive field studies over longer durations. Another challenge is that many metal-hyperaccumulating plants are poor choice for remediation due to their sluggish development and limited plant mass. For effective cleanup, plants need to have a larger root system and significant biomass, which can better absorb contaminants. To address this, careful planning and strategic plant placement are crucial to improving the process. Phytoremediation is also more effective in areas with lower contamination levels since highly polluted soils can inhibit plant growth. Additionally, there is a risk that animals or other organisms might consume plants containing high concentrations of toxic pollutants, potentially causing harm. Proper measures, such as carefully managing, treating, or removing these plants, are essential to ensure the process is safe and environmentally sustainable (112,113).

5. Techniques for enhancing phytoremediation efficiency

Soils contaminated with heavy metals generally have low organic matter, poor nutrient levels, and imbalanced pH, which can significantly limit plant growth (114). To overcome these challenges and enhance metal uptake, various soil improvement techniques are often employed. These include using organic soil conditioners, chemical fertilizers, synthetic chelators, surfactants, encouraging beneficial plant-microbe interactions, and introducing transgenes into plants. All of these methods play a role in boosting both plant growth and the efficiency of phytoremediation (115). A detailed discussion of these approaches is presented in the following section.

5.1 Approaches combining phytoremediation with microorganisms

In recent years, combining plants with bacteria has become a popular and effective approach for cleaning up metal-contaminated soils. Plant growth-promoting rhizobacteria (PGPR) contributes to this procedure by boosting both phytoremediation effectiveness and overall plant health. These bacteria support plant growth through several mechanisms, including converting nitrogen from the air into organic forms that plants can use, producing siderophores that help plants absorb iron, enhancing the accessibility of vital nutrients such as phosphorus along with releasing carbonic compounds for improving the absorption of toxic metals. Additionally, PGPR enhance the activity of plant antioxidant enzymes, which helps plants cope with the harmful effects of metal toxicity (116–121). For example, *Pseudomonas putida* has been proved to improve the eradication of dense toxic metals like Cd, Pb by increasing the plants' resistance to these metals as well as aiding their absorption and transport (122). Similarly, *Bacillus cereus* helps plants absorb metals like Cr, Zn enhancing plant development and making metals more accessible in the surrounding soil (123,124). Another example is *Azospirillum brasilense*, which assists the cleanup of crude oil compounds by stimulating plant development and root formation, speeding up hydrocarbon breakdown and improving their uptake by plants (125,126). Additionally, bacteria like *Enterobacter cloacae* are effective at cleaning up arsenic contaminated soils by enhancing plant development, boosting as absorption along with improving the effectiveness of phytoremediation (127,128).

5.2 Genetic engineering to enhance heavy metal uptake in plants

Genetic engineering holds significant promise for enhancing plant resistance to toxic metal stress as well as improving the effectiveness of bioremediation by plants. By altering specific genes, scientists can engineer plants to better tolerate heavy metals, produce more biomass, and enhance their ability to accumulate as well as detoxify metals. The process includes adding genes that control metal absorption, accumulation, movement, and detoxification into plants, enhancing their ability to thrive in polluted environments (129–131). For instance, transgenic *Arabidopsis* plants containing rice R1-type MYB regulatory protein showed increased resistance against chromium stress by regulating cellular balance and activating stress-responsive genes (132). The insertion of the bZIP gene present in *B. nivea* improves seed germination along with root development, increasing the *Arabidopsis* plant's resistance against Cd and drought conditions (133). In white poplar, the introduction of the PsMTA1 gene from *Pisum sativum* via *Agrobacterium tumefaciens*-mediated transformation led to the production of metallothionein-like proteins, enhancing resistance to copper (Cu) and zinc (Zn) toxicity compared to non-modified plants (134). A noteworthy example involves sunflowers engineered with the yeast metallothionein gene (CUP1), which enabled the production of metallothioneins, increasing their tolerance to cadmium (Cd) (135). Similarly, transgenic tobacco plants with gene (AhSIPR10)

from groundnut (*Arachis hypogaea*) exhibited resilience to salinity, heavy metals, and drought stress (136). Researchers boosted catalase activity in *Thlaspi caerulescens*, a natural bioaccumulator of cadmium (Cd) and zinc (Zn), by introducing the bacterium *Agrobacterium tumefaciens* EHA105, combined with a pBI121(a binary vector), which contained the selectable marker *nptII* and the reporter *gus* genes. This modification improved the plant's phytoremediation potential (137). The addition of the *gsh1* gene presents in *Escherichia coli* to *B. juncea* enhanced the production of glutathione and phytochelatin, thereby improving its tolerance to cadmium (138). Transgenic chickpea plants with the metallothionein type-1 gene demonstrated enhanced resistance to drought as well as toxic metal stress, underscoring the pivotal contribution of genetic engineering in advancing phytoremediation technologies (139).

5.3 Chelating agents for enhancing phytoremediation capacity

Phytoremediation efficiency is often enhanced through the widespread use of chemical chelators and surfactants (140). Among these EDTA is commonly used in agriculture. Research has shown that EDTA improves metal uptake by plants, especially hyperaccumulators that can absorb metal-EDTA complexes (141). For instance, in hydroponic conditions, EDTA increased lead (Pb) uptake in *Zea mays* by 6-7 times compared to untreated plants (142). Similar findings have been noted with other species, for example *Sedum alfredii* (143), *Vicia faba* seedlings (144), *Vetiveria zizanioides* (145), and *Canavalia ensiformis*, where EDTA application significantly boosted Pb accumulation in roots and shoots (146). Another chelating agent, EGTA, has shown benefits like enhancing Metal absorption as well as buildup in plants. For example, higher cadmium (Cd) levels have been recorded in *Althaea rosea* (147), *Mirabilis jalapa* (148), and *Calendula officinalis* (149) after EGTA treatment. Additionally, *Cicer arietinum* plants treated with EGTA exhibited increased Pb accumulation in their aerial parts (150). Surfactants, like sodium dodecyl sulfate, are also effective tools for improving phytoremediation. Thanks to their dual hydrophilic and lipophilic properties, SDS can alter the plant-soil interface to help remove contaminants that are otherwise hard to dissolve or degrade (151). Studies have shown that SDS boosts cadmium accumulation in *Althaea rosea* (152), specifically in both the roots and stems, also in the shoots of *Calendula officinalis* (153). Moreover, research highlighted that SDS treatment improved zinc (Zn) translocation in *Populus alba* (154).

5.4 Nanophytoremediation

The capacity of a wide variety of nanoparticles and nanomaterials to cleanup organic, inorganic, and toxic metal contaminants in land and aquatic environment have been evident from various studies (155,156). These materials are highly effective for the eradication of very large amounts of pollutants, encompassing metals and volatile organic compounds (VOCs), plants

have also demonstrated high uptake of pollutants. Nanomaterial-enhanced phytoremediation has consistently, and significantly, been useful for the remediation of organic pollutants. Recent evidences revealed that nanoparticle delivery can also enhance the plant ability to resist stress in ex situ and in situ hyperaccumulating elements by plants. The dimensions of the nanoparticles contribute to the ability to how deeply they can penetrate the plants, in how they are coupled to pollutants, and in how they are translocated apoplastically/symplastically in the downstream direction starting from the root (157). For example, As level of 705 ppm in the roots and 1188 ppm in the stems were achieved by applying salicylic acid nanoparticles to the *Isatis cappadocica* plant. (158).

6. Conclusion

The escalating levels of heavy metal pollution in soil and water have posed severe threats to ecosystems and human health, necessitating sustainable solutions for environmental remediation. Phytoremediation, as an eco-friendly and cost-effective technology, has gained significant attention for its ability to restore contaminated sites without causing further environmental damage. By utilizing natural processes in plants, such as pollutant absorption, stabilization, and degradation, this technique offers an innovative alternative to conventional methods like excavation, incineration, and chemical treatments. Advancements in phytoremediation, including genetic engineering, nanoparticles and microbial assistance through PGPR, have further enhanced its efficiency, enabling the remediation of complex pollutants in diverse settings. However, the practical application of phytoremediation is not without challenges. The extended duration required for remediation, coupled with limitations in heavily contaminated environments, reduces its feasibility for urgent pollution crises. Moreover, the proper disposal of plant biomass that has accumulated high levels of contaminants and the possible danger of contaminant transfer through the food chain underscore the importance of effective management strategies. The dependence on slow-growing plants with low biomass also restricts its large-scale use, highlighting the need for improved plant selection and enhancement methods.

Future research must focus on addressing these limitations to unlock the full potential of phytoremediation. This includes developing faster-growing, higher biomass plants through genetic modifications, optimizing the use of PGPR for enhanced pollutant uptake, and integrating phytoremediation with other complementary remediation methods. Long-term field applications and case studies will be essential to understanding the scalability and effectiveness of phytoremediation in diverse environmental conditions. With strategic advancements and support from policymakers, phytoremediation could become a critical component of global efforts to mitigate heavy metal pollution and achieve sustainable environmental restoration.

Declarations

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