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Phytoremediation of Metal-Contaminated Soils and Water in Pakistan: a Review

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Abstract Heavy metal pollution has increasingly deteriorated many natural ecosystems around the globe resulting in significant loss of environmental health including endangerment of many biological species. The impact of heavy metal contamination is more pronounced in the developing and resource limited countries due to poor compliance to environmental policies. Pakistan stands on the forefront of pollution threat with unprecedented levels of heavy metal contamination in soils and waters. Phytoremediation has emerged as a promising eco-friendly green technique that has proved to be inexpensive, effective, and easily applicable in different ecosystems in recent years. Different strategies of phytoremediation include phytoextraction, phytovolatilization, phytostabilization, rhizofiltration, and phytofiltration, which have shown promising results against heavy metal-contaminated areas. This paper aims to review the studies conducted in Pakistan in the last two decades that deploy phytoremediation as a green technique to remove heavy metals from the metal-contaminated environment. Merits and demerits as well as advancements and limitations of phytoremediation use in Pakistan are also discussed in the present manuscript. The role of microorganisms in metal uptake is also briefly discussed. The manuscript concludes that despite abundant availability and diversity of plant species, only a few phytoremediation studies have been conducted in Pakistan with limited scope and applicability. Nevertheless, potential of this solar driven green technique is overwhelmingly untapped and during recent years, an increasing trend on phytoremediation studies is evident in the country.

Keywords Heavy metals · Phytoremediation · Soils · Water · Pakistan

1 Introduction

The twenty-first century has been marked as an era of industrial and technological advancements by leaps and bounds. Living standards have been enormously improved. However, these achievements have greatly impacted the limited natural resources of planet Earth. Unsustainable industrial processes and agricultural practices have resulted in chronic release of contaminants from point and non-point sources in the environment. These contaminants pose serious and adverse health impacts on both wildlife and human beings (Maddela et al., 2022). With current influx in all environmental compartments, these contaminants are a serious concern to ecosystem health that

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translates their accumulation at higher trophic levels via biomagnification process (Chormare & Kumar, 2022). Environmental pollutants are broadly categorized into organics and inorganics (Akhtar et al., 2019). Organic pollutants are mostly anthropogenic and toxic/carcinogenic, whereas inorganic pollutants such as heavy metals naturally found in water and soils are persistent in our environment. These inorganic pollutants include nutrients (N and P), trace elements essential for plants (zinc (Zn), copper (Cu), manganese (Mn), iron (Fe), and boron (B), molybdenum (Mo)), non-essential elements (mercury (Hg), cadmium (Cd), chromium (Cr), cobalt (Co), lead (Pb), selenium (Se), nickel (Ni), and vanadium (V)), and radioactive isotopic elements (uranium (U), strontium (Sr) and cesium (Se)) (Dushenkov, 2003; Pilon-Smith, 2005). Organics are degradable in rhizosphere or taken up by plants and sequestered or volatilized or biodegraded, while inorganics are non-biodegradable and can be stabilized and/or sequestered in plant harvests by phytoremediation (Akhtar et al., 2019). Anthropogenic activities related to industry, agriculture, military, mining, and urban activities result in chronic release of these pollutants in ambient environment.

Countries of the global south including Pakistan have reported a consistent increase in environmental pollution caused by heavy metals in the recent past. Various sources of heavy metal pollution ranging from agricultural practices to industrial dumping heavily polluted riverine ecosystems in the country with these toxic heavy metals (Qadir et al., 2001; Shakir et al., 2016). Pakistan is an agrarian economy, and its agricultural and soil ecosystems are compromised — since they act as a sink for most if not all anthropogenic pollutants particularly toxic heavy metals (Ali et al., 2015). Naturally, heavy metal contamination of soils and waters can occur due to the natural and gradual erosion of rocks and their subsequent weathering (Sabiha-Javied et al., 2009). Anthropogenic causes include activities that involve mining as well as major industrial processes (leather tanneries, textiles, chemical, cement, fertilizer) and practices in the agriculture sector (Wuana & Okieimen, 2011).

Once these heavy metals have been introduced into the environment, even in trace amounts, these metals can be incredibly detrimental not only to human health but also for ecosystem stability and health (Kara, 2005). Heavy metals such as Hg, Sn, Cr, As, Cd, Pb, Cu, and Zn have the highest toxicity compared to other metals (Ghosh, 2010). Their toxic effects on human health are greatly significant such as hair loss in case of chromium (Salem et al., 2000), fatigue and disorientation in case of zinc (Hess & Schmid, 2002), and risk of cancer in case of cadmium and arsenic (Awofolu, 2005). Similarly, other heavy metals have been tied to different harmful ailments and disorders in humans and animals (Shakir et al., 2016).

To combat this increasingly deteriorating situation, different scientific approaches have been used to clean up contaminated sites. Phytoremediation, a cost-effective green technology, is one such emerging technique that has shown potential to mitigate pollution in both water and soil ecosystems (Cheng et al., 2002; Khan et al., 2021a, b; Meagher et al., 2000; Vithanage et al., 2012). The term phytoremediation refers to *the use of plants and associated soil microbes to reduce the concentration of toxic contaminants in the environment* (Pilon-Smith, 2005). This approach is equally applicable for solid, liquid, and gaseous pollutants. It is widely accepted as a cost-effective environmental restoration technology and regarded as a better alternative to engineering procedures that are considered destructive to the natural ecosystems. Moreover, it is solar driven biological process that has advantage (10 times cheaper) over engineering-based methods (Lin et al., 2000). Phytoremediation is proving to be an emerging technology that comes in handy to clean the soil and water bodies from deadly pollutants. In the future, it can prove to be a viable low-cost and sustainable solution to improve the deteriorating environments in the struggling economies. In Pakistan, phytoremediation applications for heavy metal removal are limited; however, in recent years, significant studies have been conducted which highlight the progress, evolution, and issues associated with phytoremediation in Pakistan. This paper reviews phytoremediation studies conducted on heavy metal-contaminated soils and water in Pakistan. The manuscript reviews the studies that have been conducted in the country during the last two decades and have shown promising results that open new avenues for consideration of phytoremediation as a reliable technique for heavy metal-polluted soil and water ecosystems. Different advantages and disadvantages of the technique and possible limitations are also addressed.

2 Types of Phytoremediation Technologies

Both plants and associated rhizospheric organisms can be deployed for phytoremediation. Different types of phytoremediation and fate of different pollutants are outlined in Fig. 1. There are several categories of phytoremediation, and the most common ones are phytoextraction, phytotransformation or phytodegradation, phytostimulation, phytovolatilization, phytostabilization, and rhizofiltration (Table 1). In Pakistan, however, most of the studies focused on phytoextraction mechanism for decontamination of polluted soil and water ecosystems. Phytoremediation processes and mechanisms for decontamination of pollutants are outlined in Fig. 2; whereas Fig. 3 categorizes the phytoremediation techniques specifically for organic and inorganic contaminants for better understanding.

3 Diversity of Higher Plants for Phytoremediation in Pakistan

A rich plant diversity in an ecosystem would help in identifying suitable plants that might have potential to clean polluted environments. From all over the world, large number of plants have been tested in Pakistan in different locations where they can grow in different climates (Kamran et al., 2014). Phytoremediation of metals has been reported by many native plant species in Pakistan (see Table S1 in supplementary information). Around 400 species

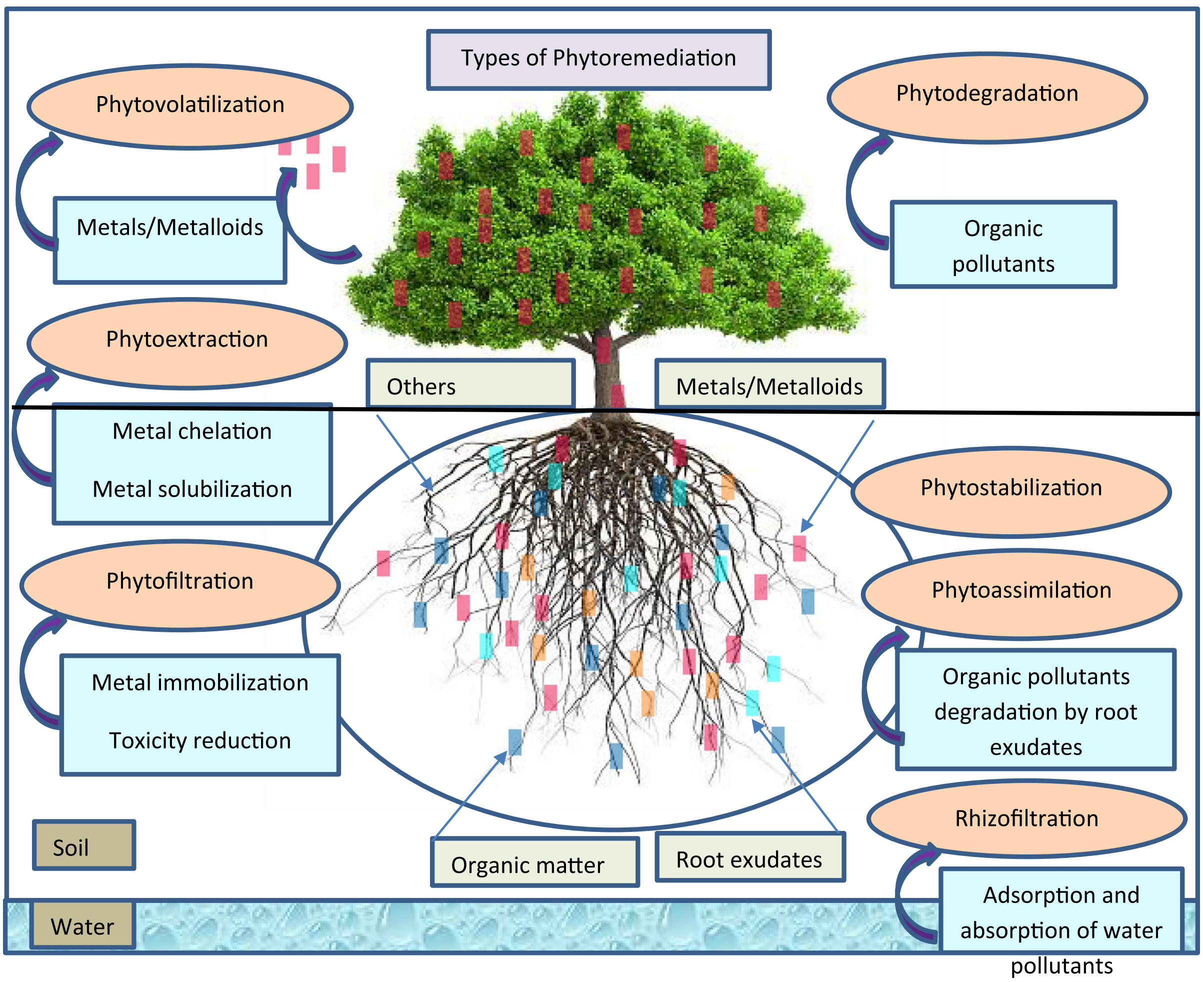

Fig. 1 Types of phytoremediation and possible fates of pollutants in phytoremediation techniques

Table 1 Most commonly used phytoremediation approaches

Technique	Description
Phytoextraction	It is the extraction and accumulation of contaminants in harvestable plant tissues including root surface and shoots. Plant biomass can be used for non-food purposes, ashing, and disposal in landfilling and phytomining
Phytotransformation	It is the degradation of complex organic molecules to simple molecules and their incorporation into plant tissues
Phytovolatalization	Volatalization of certain pollutants from plant surfaces after plant uptake. Most suitable for different volatile organics and few inorganics such as Se and Hg
Phytostimulation or rhizodegradation	It is the stimulation of microbial and fungal degradation by the release of enzymes in the root zones. Plant induced facilitated biodegradation of organic pollutants by rhizosphecric microbial community. Plant induced degradation via their own enzymes is termed as phytodegradation
Phytostabilitzation	It involves the absorption and precipitation of contaminants, i.e., metals, by plants, to prohibit their mobility to groundwater resources. Phytostabilization stabilize pollutants in soil
Rhizofiltration	It is the absorption, concentration, and accumulation of contaminants in harvestable plant roots. Suitable for removal of hazardous inorganics

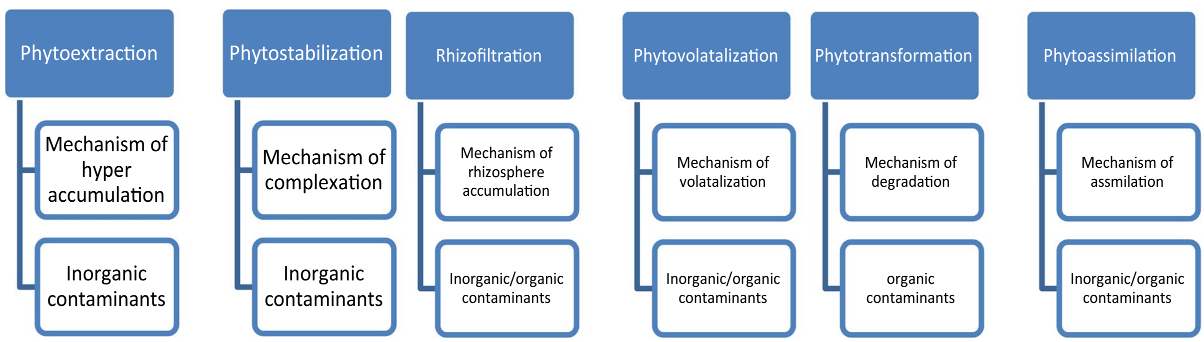

Fig. 2 Phytoremediation processes and mechanisms for decontamination

among the angiosperms are identified as hyper accumulator in the world (Mcgrath & Zhao, 2003). Pakistan has been blessed with large number of higher plant species that can be used as potential candidates for phytoremediation and only 50 species have been examined and recognized as metal accumulator that can clean and remediate polluted soils and waters. *Brassica juncea* was found in different areas in Pakistan, i.e., Islamabad Rawalpindi, Quetta, Karachi, and Lahore that were used to remediate lead (Pd), zinc (Zn), and copper (Cu) from soil. *Eichhornia crassipes*, although not common but found in many urban and industrial areas like Gujranwala, Lahore, and Rawalpindi. This plant is heavy metal accumulator of Cr, Zn, Cd, Cu, Ni, Pb, Hg, P, and pesticides. However, still, many plant species located in Pakistan are yet to be identified and tested for the phytoremediation purposes. Different plant species reported in previous studies for the phytoremediation are listed in Table S1.

4 Responses of Plants Exposed to Heavy Metals

After exposure to heavy metals, plants display either of three response strategies for growth and development on metal contaminated soils. On basis of their response, plants can be categorized as (i) metal excluders, (ii) metal indicators, and (iii) metal accumulator/hyperaccumulators (Baker & Walker, 1990; Gosh & Singh, 2005). Metal accumulators (hyperaccumulators) take up and accumulate more concentrations of heavy metals in their above-ground tissues compared to their concentrations in roots and soil. Metal translocation in above-ground parts is limited in metal excluders; however, they may still contain

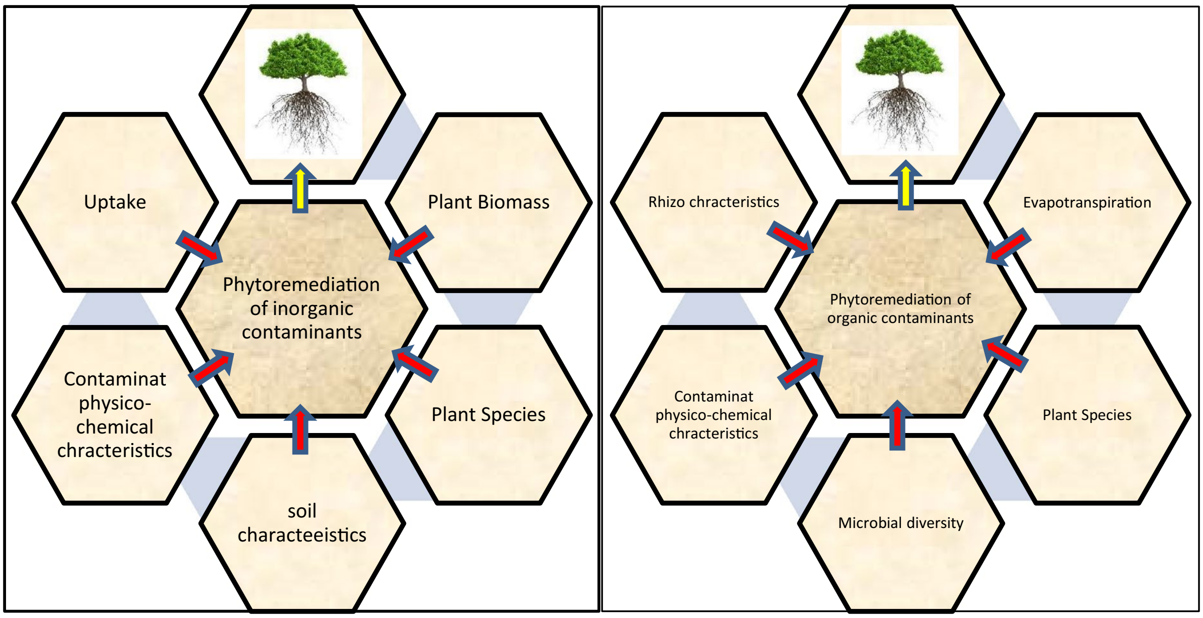

Fig. 3 Phytoremediation processes for organic and inorganic contaminants

fair concentrations in roots. Metal indicators on the other hand are plants that may accumulate metals in their above-ground parts and translocation levels are comparable to metal concentrations in the soil. Indicators may also express toxicity symptoms if they continue growing and accumulating metals and ultimately may also die-off. Response curves for plant tops concentrations vs soil metal concentrations are outlined in Fig. 4.

5 Status of Soil and Water Pollution in Pakistan

Any substance or group of substances (chemical elements, compounds, complexes) which are environmentally toxic and resist biodegradation can be listed as contaminants. These substances pose ecological threat to the environment and raise concerns regarding the sustainability of a healthy ecosystem. Major point and non-point sources of pollution are fertilizers, pesticides, spills, and industrial and urban wastes. Recent studies showed that industrial sludge, phosphate rock used to produce fertilizers, household wastes, industrial wastewater, and sewage sludge used for irrigation and medical waste incineration are the major potential sources of heavy metal pollution in Pakistan (Jamali et al., 2007; Malik & Zeb, 2009).

Almost 70% of Pakistan's underground and surface water is polluted with different types of pollutants such as (i) biological pollutants, (ii) organic pollutants, and (iii) inorganic pollutants. This is an alarming issue in Pakistan which is increasing day by day and affecting the environment very badly. The presence of different types of heavy metals in drinking water is a severe hazard for human health. Anthropogenic and many biological activities which cause increasing levels of pollutions in ground water include mining, fossil fuel combustion, pesticides, herbicides, and crop desiccants. Because of gradual increase in population level, the use of water is also increasing for agriculture and industrial demands. Water in Pakistan is contaminated and contain different metals and metalloids like Ni, As, Cr, Cu, Zn, As, Se, Pb, and Cd. Arsenic concentration is becoming a real challenge in Pakistan.

Heavy metals seem to be the dominating pollutant of soils equally especially those which are toxic in trace amounts to living organisms such as mercury, lead, antimony, bismuth, and arsenic. Gradual increases in population of Pakistan with the disposal of untreated discharges from textile industries, leather industries, municipal wastewater drains, agricultural run-off and pharmaceutical industries, etc. are the major point and non-point sources of soil pollution

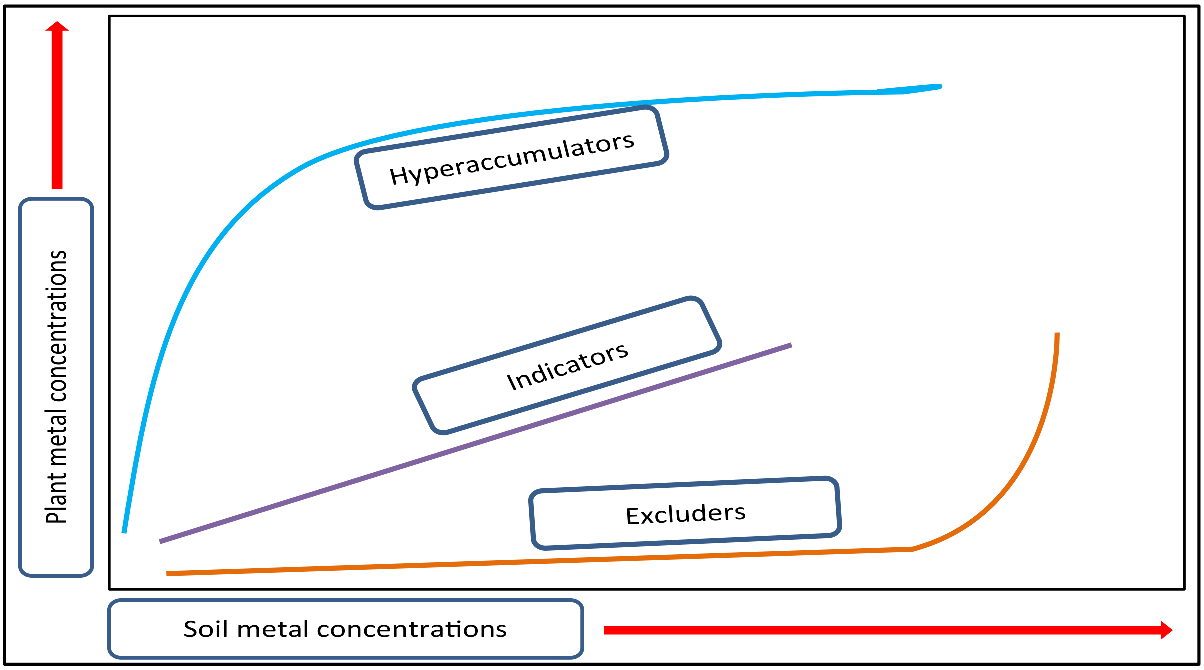

Fig. 4 Response curves for plant tops concentrations vs soil metal concentrations (adapted from Gosh & Singh, 2005)

(Jamali et al., 2007; Sial et al., 2006). For example, discharging of untreated industrial wastes from the industries has substantially increased the amount of chromium (Cr) concentrations in Pakistani soils (Sial et al., 2006).

6 Phytoremediation Studies in Contaminated Soils

KHAN (2001) conducted a study in Kala Shah Kaku area of Lahore district (31.5204° N, 74.3587° E) to determine how the diversity of arbuscular mycorrhizal (AM) fungi in the mycorrhizospheres of three tree species, i.e., *Populus euroamericana*, *Acacia arabica*, and *Dalbergia sissoo* was affected by the Cr-rich tannery effluent. They also assessed the extent of mycorrhization of roots, the populations of mycorrhizal fungal propagules in rhizospheres, and phytoextraction of Cr in tissues of selected species. The total Cr (630 ± 88 mg/kg) in the tannery effluent-contaminated soils was significantly higher ($P < 0.1$) than the reference control soil (180 ± 26 mg/kg) but were considerably less than another study reported by Raju and Tandon (1999) in the sludge formed as a result of basic tanning activities (970 mg/kg) in India. They also observed reduced plant cover on the tannery effluent-contaminated site (Khan, 2001). The Cr contents accumulated in root tissues of trees growing on tannery effluent contaminated soil were 695 µg/g by *D. sissoo*, 538 µg/g by *A. arabica*, and 459 µg/g by *P. euroamericana*. Cr contents accumulated in leaf tissues were 188 µg/g by *A. arabica*, 160 µg/g by *D. sissoo*, and 107 µg/g by *P. euroamericana*. Based on the biomagnification ration (BMR; Cr in plant tissue/available Cr in soil) and accumulation factor (ACF; Cr in mature leaves/total Cr in soil) ratios, *A. arabica* was the most efficient accumulator of Cr, as it depicted the highest values, i.e., BMR 89.1 and ACF 3.1 followed by *D. sissoo* (BMR 46.1 and ACF 2.5) and *P. euroamericana* (BMR 28.5 and ACF 1.7).

Similarly, a study of large swaths of industrial land in KPK province has identified heavy metal contamination as a major issue and metal contaminated industrial effluents cannot be used for urban agriculture which is a common practice in many developing countries (Sial et al., 2006). The soils contaminated with heavy metals normally coexist in other issues like soil salinity and sodicity (Qadir et al., 2005). These soils can be remediated based on analogous results of soils with similar physio-chemical properties. Azhar et al., (2006) reported that co-application of chelating agent (EDTA) and sunflower (*Helianthus annuus L.*) enhanced the phytoextraction of lead from lead contaminated soil. According to their results, EDTA also reduced shoot and root length and dry matter stress tolerance indices and the effect was more pronounced (50% reduction in stress tolerance) at higher levels of EDTA and concluded that addition of EDTA was effective in enhancing the uptake of Pb in sunflower plants. Jamali et al., (2007) used sorghum plants to study the mobility and transport of heavy metals such as Cu, Zn, As, Cd, Cr, Ni, and Pb, from soil and soil amended with sewage sludge in Sindh region of Pakistan and showed significant accumulation of selected metals in edible parts particularly in the presence of EDTA. Soomro et al., (2008) reported that the order of metal accumulation in black and green tea samples was K > Na > Mn > Fe > Pb > Cd from a contaminated soil. Kashif et al., (2009) reported that urban vegetables produced in urban areas of Lahore irrigated with Hudiara drain water contained significant amounts of heavy metal contents. Drain water used to irrigate soil added significant amounts of metals posing potential risk of metal accumulation by vegetables. These vegetables were suggested not fit for human consumption because of significant amounts of metal uptake. Heavy metals detected by DPTA method in drain water irrigated soil was higher (Zn, 1.35 times; Cu, 1.5 times; Fe, 1.5 times; Mn, 2 times, Cd, 2 times; Ni, 2 times; and Pb, 3.1 times) than non-drain water irrigated soil. The order of metal transfer factor in vegetables grown on contaminated soil was Spinach > ghia tori > okara > green chillies > tar > brinjal. Highest metal transfer factor for Zn (1.2), Cu (0.45), and Mn (0.25) was observed in Okra; and for Cd (1.55), Pb (0.25), Fe (0.15), Ni (0.1), and Cr (0.5) was observed in Spinach ($TF = 1.55$, 0.25, 0.15, 0.1, and 0.5 respectively) (Kashif et al., 2009). In another study conducted in Gadoon Amazai Industrial Estate (GAIE), Swabi, Khan et al., (2009) investigated the effectiveness of a continuous free surface flow wetland for removal of heavy metals from industrial wastewater. Their results reported that removal efficiencies of selected macrophytes (*Carex aquatilis*, *Typha latifolia*, *Phragmites australis*, *Pistia stratiotes*, *Juncus articulates*, *Alisma plantago-aquatica*) in constructed wetland for Pb, Cd, Fe, Ni, Cr, and Cu were 50%, 91.9%, 74.1%, 40.9%, 89%, and 48.3%, respectively. Saifullah et al., (2009) studied the lead accumulation by *Triticum aestivum* L. while using ethylenediaminetetraacetic acid (EDTA) application method and reported that EDTA application in split doses substantially increased Pb accumulation in wheat plants. In another study, same authors reported the use of elemental sulfur and EDTA enhance Pb solubilization and accumulation by wheat plants (Saifullah et al., 2010). Malik et al., (2010) studied bioconcentration factor, translocation factor, and bioaccumulation factor of 60 wild plant species grown in heavy metals (Pb, Zn, Cu, Ni, Co, Cr) contaminated soil. The order of total metal accumulation in roots was Cu > Cr > Zn > Ni > Pb > Co. Accumulation of Cu was the highest in shoots followed by Zn, Cr, Pb, Co, and Ni. Based on BCFs, TFs, and BACs' values, most of the tested species have potential for phytostabilization and phytoextraction. *Parthenium hysterophoirus* L. and *Amaranthus viridis* L. exhibited promising results for phytoextraction of Pb and Ni, whereas *Partulaca oleracea* L., *Brachiaria reptans* (L.) Gard. & Hubb., *Solanum nigrum* L., and *Xanthium stromarium* L. were more effective in phytostabilization of Pb and Cu (Malik et al., 2010). Mirza et al., (2010) used *Arundo donax species* for the phytoremediation of arsenic and Hg from contaminated soils and found that substantial levels of both toxic metals were extracted by this plant species from the soil (Mirza et al., 2010). Similarly, in another study conducted in Islamabad and Rawalpindi areas, Nazir et al., (2011) evaluated the potential of 23 plant species growing on contaminated sites for phytoremediation by calculating bioconcentration factor (BCF), biological accumulation coefficient (BAC), and biological transfer coefficient (BCF). Among 23 test plant species, 15 species were found suitable for Pb accumulation, 2 species for Cu, and 5 species for Zn. In addition to phytoaccumulation, *Brachiaria raptans* and *Malvastrum coromandelianum species* exhibited potential for phytostabilization of sites contaminated with Pb and Cu (Nazir et al., 2011). Muhammad et al., (2011) also assessed the effectiveness of wild plant species (*Plectranthus rugosus*, *Rumex hastatus*, *Fimbristylis dichotoma*, *Heteropogon conturtus*, *and Myrsine Africana*) in Mn-Zn sulfide areas of Kohistan region of northern Pakistan and reported that *P. rugosus*, *R. hastatus*, *F. dichotoma*, and *H. conturtus* are the best accumulator for Fe and Cr; K, Mg, Mn, Na, Cu, and Pb; Ni and Cd; and Ca and Zn, respectively. Bareen and Tahira (2011) investigated the clean-up potential of high biomass producing plant species such as *Suaeda fruticose* and *Calatropis procera* at tannery effluent contaminated lands ear Kasur district of Punjab. The leaves of *Suaedha fruticose* were found to accumulate more amounts of sodium (Na) and chromium (Cr) in roots and shoots, respectively. *Suaeda fruticose* showed better growth characteristics and role in phytoremediation of *Cr* compared to ten other species when grown in EDTA amended chromium-contaminated soil. At low doses of EDTA, *Suaeda fruticose* showed more Cr uptake and EDTA use also reduced Cr leaching in soil.

To identify plant species that could effectively decontaminate the heavy metal-impacted soils through phytoremediation — a "green alternative" approach was adopted in which different tree and plant species such as *Thespesia populneoides*, *Leucaena leucocephala*, *Delonex regia*, *Zea mays* and *Brassica campestris*, *Selaginella jacquemontii*, *Rumex hastatus*, *and Plectranthus rugosus* along with some wild plant species of mafic and ultramafic terrain of Kohistan region, KPK province (Ismail et al., 2013; Muhammad et al., 2013). These easily available native species showed encouraging results for phytoremediation by accumulating substantial amounts of heavy metals in different plant tissues (Akhtar & Iram, 2016). Similarly, in one study, fields were irrigated with NaCl (salt) water to remediate heavy metals such as Cd, Cu, Fe, Mn, Ni, Pb, and Zn. It showed that irrigation of brackish water from farm sludge should not be suggested because the saline water increased the movement of heavy metals and polluting the drainage water. Concentration of metal and its pattern in the plant roots were as Cu > Cr > Zn > Ni > Pb > Co. These results suggested that other environmental and edaphic factors such as soil salinity and sodicity (a major soil issue in arid areas of Pakistan) be considered for effective phytoremediation. Plants and soil samples from Kohistan and Lahore region and Northern Pakistan were analyzed for major cations (Na, K, Ca, Mg, Fe, Mn) and the heavy metals (Pd, Zn, Cd, Cu, Cr, Ni, Co). Phyto-accumulation prospects of Cd and Zn by mycorrhizal plant species growing in industrially polluted soil. Plant species which can uptake metals include *Desmostachya bipinnata*, *Dichanthium annulatums*, *Malvastrum coromandelianum*, *Saccharum bengalense*, and *Trifolium alexandrinum.*

The roots of *Trifolium alexandrinum* were found to be effective in the uptake of Zn, Pb, Cu, and Cd (following the trend of Zn > Pb > Cu > Cd). Bioconcentration factor (BCF_{root}) values were 4.242, 1.544, 1.071, and 0.604 for Zn, Pb, Cu, and Cd, respectively (Ali et al., 2012). Muhammad et al., (2013) tested wild plant species in Jijal, Dubair, and Alpuri areas of KPK of Pakistan and showed that *Selaginella jacquemontii*, *Rumex hastatus*, and *Plectranthus rugosus* were more effective in phytoremediation among all tested species. Three plant species removed Fe, Mn, Cr, and Ni more effectively from macro and trace metal-impacted soils.

Ismail et al., (2013) determined the effects of Cd and Pb on three different tree species: *Thespesia populneoides*, *Leucaena leucocephala*, and *Delonex regia*. The results showed that these metals caused metabolic disturbances and affected the growth of species — Cd being more toxic than Pb. The study found that *Delonex regia* was more endemic to metal-contaminated soil and tolerated more amount of Pb and Cd. This study was significant for its perspectives and highlighted the effect of heavy metal accumulation in plants. In conclusion, they reported that *Delonex regia* was most tolerant species (TI for Cd was 53 and for Pb was 69.1) out of three tested species and was identified as the best candidate for phytoremediation of both metals. Phyto-stabilization potential of two plant species (*Conocarpus erectus L.* and *Populus deltoides L.*) was evaluated from arsenic-contaminated soil by Hussain et al., (2017) who found that *Conocarpus erectus* was effective in tolerating arsenic concentration in soil. Lower values of TF (0.94) and BCF (0.23) depicted that these species as non-hyperaccumulators, however, were still recommended for phytostabilization of arsenic-contaminated soil. In another study conducted using pot experiment, Niazi et al., (2017) evaluated the arsenic tolerance and accumulation potential of *Brassica napus* and *Brassica juncea species* and reported that *Brassica napus* performed better ($TF = 0.44$; $BCF = 1.1$) in arsenic tolerance and accumulation. They also showed that phosphate supplementation enhanced As phytoextraction efficiency by tested plant species (Niazi et al., 2017). In a similar study, Ahmad et al., (2018) tested three species, namely, *Eucalyptus camaldulensis*, *Terminalia arjuna*, and *Salix tetrasperma* in As-stressed environment and found them as strong contenders for extraction of arsenic from soil. They also reported that *E. camaldulensis* plants accumulated maximum arsenic (21–26 mg/kg in shoots) and confirmed it a good candidate for phytoremediation (Ahmad et al., 2018).

In a study by Khalid et al., (2018), *Xanthium strumarium* was evaluated for its phytoremediation potential for cadmium, lead, nickel, and zinc from contaminated soil collected from roadside areas of Faisalabad. Their results exhibited that *X. strumarium* leaves could accumulate 0.27 ± 0.01 mg kg^{-1} cadmium, 3.3 ± 0.2 mg kg^{-1} lead, 54.5 ± 1 mg kg^{-1} zinc, and 5.9 ± 0.1 mg kg^{-1} nickel from the soils that had average metal concentration of 0.25 ± 0.2 mg kg^{-1} cadmium, 3.4 ± 0.3 mg kg^{-1} lead, 119 ± 1 mg kg^{-1} zinc, and 4.9 ± 0.1 mg kg^{-1} nickel (Khalid et al., 2018). The plant leaves showed particularly higher accumulation capacity for nickel, cadmium, and lead as indicated by their highest BCF values which were 1.7, 1.6, and 1.1, respectively (Khalid et al., 2018). Sajad et al., (2020a, b) analyzed and evaluated the roots and shoots of 61 plant species for remediation of Cr-contaminated soils in Lower Dir, KPK. *Argyrolobium stenophyllum*, *Silybum marianum*, *Bryophyllum daigremontianum*, *Limonium macrorhabdon*, *Calendula arvenis*, and *Delphinium suave* were found to be most effective for Cr phytostabilization; whereas *Rosularia adenotricha*, *Catharanthus roseus*, *Allium griffithianum*, *Himalaiella heteromalla*, *Stellaria media*, *Salvia moorcroftiana*, and *Marrubium vulgare* were observed suitable candidates for Cr phytoextraction. They recommended *Allium griffithianum* and *Catharanthus roseus* species to be hyperaccumulators for Cr-contaminated soils (Sajad et al., 2020a, b). Later that year, Ullah and Muhammad (2020) conducted a study in Zhob and Loralai valleys of Baluchistan to record heavy metal concentrations in plants and soil samples. Their findings suggested the use of *Phoenix dactylifera L.* and *Calotropis procera L.* for the uptake of heavy metals such as Cd, Cu, Ni, Mn, Fe, Pb, and Cr. Khan et al., (2021a, b) through a greenhouse environment explored the mobility of Pb from military shooting range soils in Nowshera to *Chenopodium album L.* and *Cynodon dactyl L.* Their study found significant correlation between the total concentration of Pb exposed to the plant species and the amount of Pb accumulated by the plants. In another study, Ullah et al., (2021) tested phytoremediation potential of *Xanthium strumarium* in different abandoned habitats in Khyber Pakhtunkhwa (KPK) region for Pb, Cd, Cu, and Zn. Pseudo-metallophyte nature of *X. strumarium* and soil's metalliferous nature in the study area were determining factors affecting phytoremediation potential of the tested plant. Hyperaccumulation capacity in different functional traits with comparatively high Pb, Cd, and Zn (≥ 1 TF) mobility indicating that these plants can effectively be used for Pb phytoextraction and phytostabilization of Cd, Cu, and Zn. BAF(S/R) (bioaccumulation factor from soil to roots) values for tested metals are represented in Table 2. Values higher than 1 are indicative value of plants as hyperaccumulator. In another reported study, Khan et al., (2022) used a few native plant species from northern region of Pakistan, Dargai Malakand (KPK) to evaluate their phytoremediation potential from heavy metal contaminated soils and water. The study recorded that among the tested nine plant species, *Pteris vittata* roots had the highest BCF for Zn and iron, 3.93 and 1.62, respectively while the roots of *Populus nigra* had the highest BCF for Cr (0.72). Overall, the TF was observed to be the highest among the following plants: *Pteris vittata* (0.99), *Xanthium strumarium* (0.97), and *Verbascum Thapsus* (0.94). Ali et al., (2022) also tested four different species of Brassicaceae family (i.e., *Brassica juncea*, *Brassica napus*, *Brassica rapa*, *Brassica campestris*) in pots for the remediation of heavy metal-contaminated soil of Lakki Marwat city, Pakistan that was irrigated with municipal wastewater. BAF and TF values of all species indicated that *B. juncea* accumulated maximum heavy metals (Cd, Pb, Cr, Ni). Enhanced phytoextraction ability was observed for all Brassica species in decreasing order, i.e., *B. napus* > *B. campestris* > *B. rapa*. The *B. juncea* was proved to be a better choice for phytoremediation purposes in soils of study area. These studies were highly useful for understanding the phytoremediation processes in metal-contaminated soils. More recently, Rahman et al., (2022) conducted a study in an industrial area of Faisalabad, Punjab to test six tree species for their phytoremediation capabilities to eliminate Zn, Pb, Cd, and Cu from polluted soils. Among the six evaluated tree species, the authors recommended using *Morus alba* and *Eucalyptus camaldulensis* for more "efficient" removal of heavy metals. The study, furthermore, compared how the accumulation of heavy metals differs on the basis of seasonal changes and for the industrial and the residential areas.

Table 2 Phytoremediation of heavy metal contaminated soil

Heavy metals/ metalloids	Plant species	Region	Technique	Efficacy/tolerance	Reference
Chromium (Cr)	*Dalbergia sissoo, Acacia arabica, Populus euroamericana*	Lahore, Punjab	Phytoaccumulation	459–695 µg/g (root) 107–188 µg/g (shoot)	Khan, 2001
	Sorghum bicolor	Hyderabad, Sind	Phytoaccumulation	0.28 mg/kg	Jamali et al., 2007
	Spinacia oleracea	Lahore, Punjab	Phytoaccumulation	$TF=0.5$	Kashif et al., 2009
	Alisma plantago-aquatica	Swabi, KPK	Phytoaccumulation	2.3 mg/kg (roots) 1.2 mg/kg (aerial)	Khan et al., 2009
	Chenopodium album L	Islamabad	Phytoextraction, phytostabilization	184 mg/kg (roots) 47 mg/kg (shoot)	Malik et al., 2010
	Plectranthus rugosus	Kohistan, KPK	Phytoaccumulation	BAC (20)	Muhammad et al., 2011
	Suaeda fruticosa; *Calatropis procera*	Kasur, Punjab	Phytoextraction	409–1756 µg/g (root) 436–1389 µg/g (shoot)	Bareen & Tahira, 2011
	Selaginella jacquemontii, Rumex hastatus	Kohistan, KPK	Phytoextraction	26–848 µg/g	Muhammad et al., 2013
	Allium griffithianum, Catharanthus roseus	Lower Dir, KPK	Phytoextraction	$BCF=7.4$ $TF=29.9$	Sajad et al., 2020a, b
	Leptochloa fusca	Faisalabad	Phytostabilisation	$BCF=2$ (root) $BCF=0.1$ (shoot)	Ullah et al., 2020
	Brassica juncea	KPK	Phytoaccumulation	$BAF=3.9$	Ali et al., 2022
Cadmium (Cd)					
	Sorghum bicolor	Hyderabad, Sind	Phytoaccumulation	0.85 mg/kg	Jamali et al., 2007
	Spinacia oleracea	Lahore, Punjab	Phytoaccumulation	$TF=1.55$	Kashif et al., 2009
	Juncus articulates, Pistia stratiotes	Swabi, KPK	Phytoaccumulation	5.3 mg/kg (roots) 2.5 mg/kg (aerial)	Khan et al., 2009
	R. hastatus	Kohistan, KPK	Phytoaccumulation	BAC (1.3)	Muhammad et al., 2011
	Trifolium alexandrinum	Malakand	Phytoaccumulation	$BCF=0.60$	Ali et al., 2012
	Delonex regia	Karachi	Hyper-tolerance	$TI=53$	Ismail et al., 2013
	Selaginella jacquemontii, Rumex hastatus	Kohistan	Phytoextraction	0- µg/g	Muhammad et al., 2013
	Ricinus communis L	Chakdara	Phytoaccumulation	331 mg/kg (roots) 143 mg/kg (shoot)	Hadi et al., 2016
	Alternanthera bettzickiana	Punjab	Phytoaccumulation	126 mg/kg (roots) 97.6 mg/kg (aerial)	Tauqeer et al., 2016
	Xanthium strumarium	Faisalabad	Phytoaccumulation	$BCF=1.57$	Khalid et al., 2018
	Xanthium strumarium	KPK region	Phytoaccumulation	$BAF=2.79$	Ullah et al., 2021
	Brassica juncea	KPK	Phytoaccumulation	$BAF=1.9$	Ali et al., 2022

Table 2 (continued)

Heavy metals/ metalloids	Plant species	Region	Technique	Efficacy/tolerance	Reference
Lead (Pb)					
	Helianthus annuus L	Faisalabad	Phytoextraction	More than 50% reduction	Azhar et al., 2006
	Sorghum bicolor	Hyderabad, Sind	Phytoaccumulation	3.8 mg/kg	Jamali et al., 2007
	Spinacia oleracea	Lahore, Punjab	Phytoaccumulation	*TF* = 0.25	Kashif et al., 2009
	Pistia stratiotes	Swabi, KPK	Phytoaccumulation	4.8 mg/kg (roots) 2.2 mg/kg (aerial)	Khan et al., 2009
	Amarannthus viridis L	Islamabad	Phytoextraction	43 mg/kg (roots) 39 mg/kg (shoot)	Malik et al., 2010
	Dactyloctnium aegypticum L., *Protulaca oleracea* L	Rawalpindi, Islamabad	Phytoextraction	56 mg/kg (roots) 51 mg/kg (shoot)	Nazir et al., 2011
	I. gerardiana, *F. dichotoma*	Kohistan, KPK	Phytoaccumulation	BAC (16)	Muhammad et al., 2011
	Trifolium alexandrinum	Malakand	Phytoaccumulation	*BCF* = 1.5	Ali et al., 2012
	Delonex regia	Karachi	Hyper-tolerance	*TI* = 69.1	Ismail et al., 2013
	Selaginella jacquemontii, *Rumex hastatus*	Kohistan	Phytoextraction	6–17 µg/g	Muhammad et al., 2013
	Alternanthera bettzickiana	Punjab	Phytoaccumulation	159 mg/kg (roots) 129 mg/kg (aerial)	Tauqeer et al., 2016
	Xanthium strumarium	Faisalabad	Phytoaccumulation	*BCF* = 1.05	Khalid et al., 2018
	Xanthium strumarium	KPK region	Phytoaccumulation	*BAF* = 2.2	Ullah et al., 2021
	Brassica juncea	KPK	Phytoaccumulation	*BAF* = 4.3	Ali et al., 2022
Zinc (Zn)					
	Glycine max, *Lens culinaris*	Islamabad	Phytoextraction	10–14 µg/g (shoot)	Jamal et al., 2002
	Sorghum bicolor	Hyderabad, Sind	Phytoaccumulation	68 mg/kg	Jamali et al., 2007
	Spinacia oleracea	Lahore, Punjab	Phytoaccumulation	*TF* = 1.2	Kashif et al., 2009
	Brachiaria reptans (L.) gard & hubb	Islamabad	Phytoextraction	135 mg/kg (roots) 135 mg/kg (shoot)	Malik et al., 2010
	Solanum nigrum L., *Xanthium strumarium* L	Rawalpindi, Islamabad	Phytoextraction	134 mg/kg (roots) 194 mg/kg (shoot)	Nazir et al., 2011
	R. hastatus	Kohistan, KPK	Phytoaccumulation	BAC (132)	Muhammad et al., 2011
	Trifolium alexandrinum	Malakand	Phytoaccumulation	*BCF* = 4.2	Ali et al., 2012
	Selaginella jacquemontii, *Rumex hastatus*, *Plectranthus rugosus*	Kohistan	Phytoextraction	5–54 µg/g	Muhammad et al., 2013
	Xanthium strumarium	KPK region	Phytoaccumulation	*BAF* = 2.9	Ullah et al., 2021

Table 2 (continued)

Heavy metals/ metalloids	Plant species	Region	Technique	Efficacy/tolerance	Reference
Arsenic (As)					
	Sorghum bicolor	Hyderabad, Sind	Phytoaccumulation	0.2 mg/kg	Jamali et al., 2007
	Arundo donax	Abbotabad	Phytoaccumulation	2.1 mg/kg	Mirza et al., 2010
	Conocarpus erectus	Vehari	Phytostabilization	$TF=0.94$ $BCF=0.23$	Hussain et al., 2017
	Brassica napus	Faiasalabad	Phytoextraction	$TF=0.44$ $BCF=1.1$	Niazi et al., 2017
	Eucalyptus camaldulensis	Lahore	Phytoextraction	29–37 mg/kg (roots) 21–26 mg/kg (shoots)	Ahmad et al., 2018
Copper (Cu)					
	Sorghum bicolor	Hyderabad, Sind	Phytoaccumulation	7.9 mg/kg	Jamali et al., 2007
	Spinacia oleracea	Lahore, Punjab	Phytoaccumulation	$TF=0.45$	Kashif et al., 2009
	Phragmites australis	Swabi, KPK	Phytoaccumulation	3.2 mg/kg (roots) 1.7 mg/kg (aerial)	Khan et al., 2009
	Cenchrus pennisetiformis L	Islamabad	Phytoextraction	417 mg/kg (roots) 172 mg/kg (shoot)	Malik et al., 2010
	Cenchrus pennisetiformis L., *Protulaca oleracea* L	Rawalpindi, Islamabad	Phytoextraction	417 mg/kg (roots) 172 mg/kg (shoot)	Nazir et al., 2011
	R. hastatus	Kohistan, KPK	Phytoaccumulation	BAC (45)	Muhammad et al., 2011
	Trifolium alexandrinum	Malakand	Phytoaccumulation	$BCF=1.1$	Ali et al., 2012
	Selaginella jacquemontii, *Rumex hastatus*, *Plectranthus rugosus*	Kohistan	Phytoextraction	16–146 µg/g	Muhammad et al., 2013
	Xanthium strumarium	KPK region	Phytoaccumulation	$BAF=0.9$	Ullah et al., 2021

They found that the ability of the trees to remediate the heavy metal-contaminated sites was greater in the industrial areas than in residential areas. Moreover, they showed that the concentration of accumulated heavy metals was higher in the summer season than in winter season. Nevertheless, more soil trials are direly needed for exploring potential of phytoremediation by deploying native plant species easily available in Pakistan.

7 Phytoremediation Studies in Water/Wastewater

Phytoremediation has also shown encouraging results with regard to the treatment of wastewater. Thirteen plant species from Hattar Industrial Estate were tested for their phytoremediation potential and their ability to uptake the heavy metals. Although the accumulation rate of heavy metals was different for each element — all plant species used in the study were able to remove metals in following order: Fe > Zn > Cr > Pb > Ni > Cd) (Ismail et al., 2013) in agreement with another study reported by Ali et al., (2012). *Euphorbia prostrata* (commonly called spurge) has been frequently identified as hyperaccumulator of heavy metals when it was exposed to different concentrations of polluted wastewater (Husnain et al., 2013). Irshad et al., (2015a) investigated phytoremediation potential of ten plant species in the city of Abbottabad, KPK province and their results were in the agreement with earlier findings. The order of metal concentrations among species was Fe > Zn > Cr > Ni > Cd. Among all the species investigated, *Cannabis sativa L.* accumulated more heavy metals than other species followed by *Chenopodium*

Table 2 (continued)

Heavy metals/ metalloids	Plant species	Region	Technique	Efficacy/tolerance	Reference
Nickel (Ni)					
	Sorghum bicolor	Hyderabad, Sind	Phytoaccumulation	2.45 mg/kg	Jamali et al., 2007
	Glycine max, *Lens culinaris*	Islamabad	Phytoextraction	4.5–10 µg/g (shoot)	Jamal et al., 2002
	Spinacia oleracea	Lahore, Punjab	Phytoaccumulation	*TF* = 0.1	Kashif et al., 2009
	Typha latifolia	Swabi, KPK	Phytoaccumulation	3.0 mg/kg (roots) 1.6 mg/kg (aerial)	Khan et al., 2009
	Cenchrus penniseti-formis L	Islamabad	Phytoextraction	47 mg/kg (roots) 18.2 mg/kg (shoot)	Malik et al., 2010
	F. dichotoma	Kohistan, KPK	Phytoaccumulation	BAC (14)	Muhammad et al., 2011
	Selaginella jacque-montii, *Rumex hastatus*, *Plectran-thus rugosus*	Kohistan	Phytoextraction	84–2049 µg/g	Muhammad et al., 2013
	Xanthium stru-marium	Faisalabad	Phytoaccumulation	*BCF* = 1.65	Khalid et al., 2018
	Xanthium strumar-ium, *Bryophyllum daigremontianum*	Lower Dir	Phytoextraction	541 mg/kg (roots) 296 mg/kg (shoot)	Sajad et al., 2020a, b
	Brassica juncea	KPK	Phytoaccumulation	*BAF* = 5	Ali et al., 2022
Iron (Fe)	*Spinacia oleracea*	Lahore, Punjab	Phytoaccumulation	*TF* = 0.15	Kashif et al., 2009
	Carex aquatilis	Swabi, KPK	Phytoaccumulation	4.6 mg/kg (roots) 2.5 mg/kg (aerial)	Khan et al., 2009
	Plectranthus rugosus	Kohistan, KPK	Phytoaccumulation	BAC (1190)	Muhammad et al., 2011
	Selaginella jacque-montii, Rumex hastatus	Kohistan	Phytoextraction	114–11,766 µg/g	Muhammad et al., 2013
Manganese (Mn)	*Spinacia oleracea*	Lahore, Punjab	Phytoaccumulation	*TF* = 0.25	Kashif et al., 2009
	R. hastatus	Kohistan, KPK	Phytoaccumulation	BAC (585)	Muhammad et al., 2011
	Selaginella jacque-montii, *Rumex hastatus*, *Plectran-thus rugosus*	Kohistan	Phytoextraction	116–3154 µg/g	Muhammad et al., 2013
Cobalt (Co)					
	Partulaca oleracea L., *Malvestrum coromandialinum* L	Islamabad	Phytoextraction	40 mg/kg (roots) 25 mg/kg (shoot)	Malik et al., 2010
	I. geradiana	Kohistan, KPK	Phytoaccumulation	BAC (8)	Muhammad et al., 2011
	Selaginella jacque-montii, *Rumex hastatus*, *Plectran-thus rugosus*	Kohistan	Phytoextraction	15–107 µg/g	Muhammad et al., 2013
Mercury (Hg)					
	Arundo donax	Abbotabad	Phytoaccumulation	0.92 mg/kg	Mirza et al., 2010
	Petroselinum crispum	Abbotabad	Phytoaccumulation	*BAF* = 9.32	Bibi et al., 2016

album L. and *Xanthium stramonium L.* Nonetheless, none were identified as “hyper-accumulators” because concentration of heavy metals was < 100 mg/kg in plant biomass. All plant species accumulated more metals in roots than in shoots — having a translocation factor (TF) less than 1 (Irshad et al., 2015a).

Other species like *Dalbergia sissoo* and *Acacia modesta* have also showed better performance to absorb heavy metals in their root and shoot biomass. Irshad et al., (2015b) in another study compared the potential of thirteen native plant species (including trees, bushes, and grasses) for the phytoaccumulation of heavy metals (HM). Among tested plant species, the accumulation potential for HM varied depending on the type of element. Regardless of the plant species, HM concentrations varied in the order of Fe > Zn > Cr > Pb > Ni > Cd > As. Tree species of *Ricinus communis*, *Acacia nilotica*, *Acacia modesta*, and *Dalbergia sissoo* exhibited an enhanced concentration of metals (Irshad et al., 2015b). These studies were highly useful for understanding the phytoremediation processes. Bokhari et al., (2016) also used common duckweed (*Lemna minor*) to extract heavy metals from extremely toxic wastewater. This study was significant as it tested the phytoremediation potential of the species separately in municipal and industrial wastes. The authors identified *Lemna minor* as a moderate accumulator of both affluents with high accumulation potential for heavy metals especially lead (Bokhari et al., 2016). Rapeseed *Brassica napus* showed substantial phytoremediation potential for copper and other metals in soils when citric acid was added to the growth medium. The study also found positive effects of citric acid for plant growth during phytoremediation suggesting an interesting link (Zaheer et al., 2015).

Among different techniques of phytoremediation, *phytoextraction* has been extensively studied and applied to explore heavy metal hyper-accumulators (Akhtar & Iram, 2016). Impact is substantially significant by deploying symbiotic relationship between plants and micro-organisms present in the rhizosphere (Akhtar & Iram, 2016; Ullah et al., 2014; Zubair et al., 2016). Recent studies highlighted effectiveness and assessment of phytoremediation. Molybdenum demonstrated significantly positive effect on Cd uptake and plant growth even under high cadmium stress (Hadi et al., 2016). *Alternanthera bettzickiana* plants showed tolerance for heavy metals especially for lead (Pb) and cadmium (Cd). This tolerance was observed at low heavy metal levels in soil where the plant’s physiological growth was fast and without visible effects of the heavy metals. However, after 8-week period, high levels of metal accumulation slowed impaired plant growth and considerably reduced plant biomass (Tauqeer et al., 2016). This study is significant as it highlighted the effects of phytoremediation on plant physiology as well. In another study of heavy metal accumulation by Khan et al., (2019), castor cultivators were grown in soil over a period of hundred days to estimate the behavior of seven different heavy metals. The authors found substantial accumulation of all seven heavy metals and soil quality was improved significantly after the 100-day growth period (Khan et al., 2019). The potential of *Eichhornia crassipes* commonly referred to as water hyacinth to extract heavy metals from wastewater was also estimated. Samples were collected from Narowal, Punjab Province and surrounding areas to estimate the amounts of heavy metals accumulated by hyacinth. Results highlighted that *Eichhornia crassipes* can be used as a potential plant for phytoremediation in Pakistan (Nazir et al., 2020). In addition, species ranging from parsley plant to *Zea mays* also showed significant results for metal remediation (Bibi et al., 2016; Bokhari et al., 2016; Rizwan et al., 2016; Sarwar et al., 2017). Sajad et al., (2020a, b) tested 61 plant species in a soil study in Lower Dir district area of KPK, Pakistan to estimate phytoremediation potential for nickel. Although most species showed substantial phytoextraction and phyto-stabilization, phytoaccumulation of lead was not detected. With 61 species tested, this study had a broader scope of phytoremediation application.

Novel techniques like floating wetlands and artificially constructed floating aquatic plants that extract heavy metals from polluted water are also getting well-deserved attention due to their relative success in phytoremediation (Ali et al., 2020). For successful phytoremediation, it is important to pay close attention to the effects of the process on the plant species itself and conducted studies have focused well on this aspect during quantification of impacts (Ullah et al., 2020). During the last two decades, different studies of phytoremediation conducted in Pakistan are summarized in Tables 2 and 3.

Table 3 Phytoremediation of heavy metal contaminated water

Heavy metals/metalloids	Plant species	Region	Technique	Efficacy/tolerance	Reference
Chromium (Cr)	*Euphorbia prostrata*	Layyah, Punjab	Phytoaccumulation	2.20 µg/g	Husnain et al., 2013
	Cannabis sativa L., Chenopodium album L	Abbottabad, KPK	Phytoaccumulation	187 µg/g (root) 164 µg/g (shoot)	Irshad et al., 2015a, b
	Dalbergia sissoo, Acacia modesta	Haripur, KPK	Phytoaccumulation	400 µg/g (root) 350 µg/g (shoot)	Irshad et al., 2015b
Cadmium (Cd)					
	Euphorbia prostrata	Layyah, Punjab	Phytoaccumulation	1.97 µg/g	Husnain et al., 2013
	Cannabis sativa L., Chenopodium album L	Abbottabad, KPK	Phytoaccumulation	10.1 µg/g (root) 8.5 µg/g (shoot)	Irshad et al., 2015a, b
	Acacia nilotica, Dactyloctenium aegyptium	Haripur, KPK	Phytoaccumulation	9.2 µg/g (root) 8.85 µg/g (shoot)	Irshad et al., 2015b
	Lemna minor L	Islamabad	Phytoaccumulation	24.2 µg/g	Bokhari et al., 2016
	Eichhornia crassipes	Narowal, Punjab	Phytoaccumulation	166.3 µg/g	Nazir et al., 2020
Lead (Pb)					
	Euphorbia prostrata	Layyah, Punjab	Phytoaccumulation	0.90 µg/g	Husnain et al., 2013
	Acacia nilotica, Dalbergia sissoo	Haripur, KPK	Phytoaccumulation	155 µg/g (root) 142 µg/g (shoot)	Irshad et al., 2015b
	Lemna minor L	Islamabad	Phytoaccumulation	310 µg/g	Bokhari et al., 2016
Zinc (Zn)					
	Cannabis sativa L., Chenopodium album L	Abbottabad, KPK	Phytoaccumulation	273 µg/g (root) 254 µg/g (shoot)	Irshad et al., 2015a, b
	Dalbergia sissoo, Acacia nilotica	Haripur, KPK	Phytoaccumulation	202 µg/g (root) 205 µg/g (shoot)	Irshad et al., 2015b
Arsenic (As)					
	Acacia modesta, Dactyloctenium aegyptium	Haripur, KPK	Phytoaccumulation	110 µg/g (shoot)	Irshad et al., 2015b
	Eichhornia crassipes	Dhamthal, Punjab	Phytoaccumulation	0.1 µg/g	Nazir et al., 2020
Copper (Cu)					
	Lemna minor L	Islamabad	Phytoaccumulation	35 µg/g	Bokhari et al., 2016
Nickel (Ni)					
	Cannabis sativa L., Chenopodium album L	Abbottabad, KPK	Phytoaccumulation	35.4 µg/g (root) 28.3 µg/g (shoot)	Irshad et al., 2015a, b
	Dalbergia sissoo, Acacia nilotica	Haripur, KPK	Phytoaccumulation	30 µg/g (root) 24 µg/g (shoot)	Irshad et al., 2015b
	Lemna minor L	Islamabad	Phytoaccumulation	19 µg/g	Bokhari et al., 2016
Iron (Fe)					
	Cannabis sativa L., Chenopodium album L., Xanthium stramonium L	Abbottabad, KPK	Phytoaccumulation	346 µg/g (root) 322 µg/g (shoot)	Irshad et al., 2015a, b
	Dalbergia sissoo, Acacia modesta	Haripur, KPK	Phytoaccumulation	405 µg/g (root) 312 µg/g (shoot)	Irshad et al., 2015b
Mercury (Hg)					
	Eichhornia crassipes	Dhamthal, Punjab	Phytoaccumulation	0.05 µg/g	Nazir et al., 2020

Studies have also been conducted to compare the accumulating potential of various plants for selected metals and showed discrepancies among reported results in natural and hydroponic environments (Mohtadi et al., 2012). These authors reported that some plant species seem to be hyper tolerant of one metal in one environment while in other environment, same species did not show any considerable accumulation and tolerance. This shows that site-specific environment should not be overlooked for phytoremediation. Nevertheless, phytoremediation has the potential to be integrated with other rapidly advancing engineering based clean up methods in different ecosystems (Ali et al., 2013). In addition, molecular techniques need to apply for better understanding of metal sequestration, acquisition, translocation, and tolerance in higher plants.

8 Application of Microorganisms: the Future of Phytoremediation

The ingenious use of microbes (bacteria and fungi) and chelating agents during phytoremediation enhances efficiency of plants used in phytoremediation and this co-use strategy is increasingly dominating the remediation studies owing to their success. Jamal et al., (2002) investigated the impact of mycorrhizal fungi in the plant uptake of nickel and reported that the soybean and lentil plants were effective in its uptake. Their results showed that both plant species accumulated both metals; however, soybean plants accumulated significantly ($P < 0.05$) higher Zn contents in their tissues compared to the lentil plant. Both these studies provided important insights for the application of phytoremediation as a viable method of cleaning up contaminated environments. In another study, bacterial ACC deaminase has been used to enhance the root network of plants which subsequently increased their phytoremediation potential (Arshad et al., 2007). Studies conducted prior to the application of biological and chemical approaches suggest that the shoots accumulated less concentration of the contaminants owing to hindrance in the transport of contaminants (Ali et al., 2012). Using the emerging approaches, the translocation along with other limitations can be addressed. Akhtar and Iram (2016) used diethylenetriamine pentaacetate (DTPA) along with *Aspergillus niger*, *Aspergillus flavus*, and *Aspergillus fumigatuse* — three fungi species for better translocation of heavy metals from contaminated soils of Lahore and Gujranwala, Punjab using *Zea mays* and *Brassica campestris* species. They found that *Aspergillus fumigatuse* showed better results in supporting plants root and shoot growth. In addition, it stimulated the metal accumulation, resulting in improved phytoremediation potential for the extraction of heavy metals (mainly Cr, Cd, Pb, Cu) when compared to the chemical amendment-DPTA (diethylene triamine penta acetic acid).

The bioavailability of heavy metals, type of microbes, plant potential to accumulate, and soil properties define the nature of phytoremediation (Ullah et al., 2014). The use of proper chemical and biological approaches which assist plant growth one way or another assists phytoremediation (Shahid et al., 2013; Ullah et al., 2014; Zubair et al., 2016). Rhizobacteria are such micro-organisms that under normal conditions stimulate the growth of hyperaccumulators owing to their symbiotic association and alter the bioavailability of heavy metals in soil (Zubair et al., 2016). Diazotrophs also assist in plant growth through biocontrol which helps with the heavy metal accumulation (Ullah et al., 2014). Strong organic chelating agent ethylene diamine tetra acetic acid (EDTA) is known to enhance plants' phytoextraction efficiency and reducing the time duration of the process by solubilizing soil-bound heavy metals and altering the bioavailability — making the contaminants more "phytoavailable." However, it might have toxic effects on soil microbes and plants (Shahid et al., 2013). EDTA was also used to increase the phytoremediation of *Sorghum bicolor* and *Avena sativa* in Gujranwala, Punjab province where these species were able to remediate notoriously contaminated soils from municipal and industrial waters. Although all tested heavy metals were accumulated, the uptake of cadmium was significantly higher compared to other metals (Mahmood-ul-Hassan et al., 2017). The role of *pseudomonas* has also been studied in bioremediation and has shown positive potential for remediation of contaminants (Naz et al., 2016). It is important to note that microorganisms are not the only agents to enhance phytoremediation but other stimulating agents such as organic chelates such as EDTA and DTPA are also being used (Khan et al., 2016). Bacteria have also played an important role in phytoremediation of sewage water by water hyacinth (Mahfooz et al., 2020).

9 Phytoremediation Limitations

Although phytoremediation gained much interest as a reliable green technology for decontamination of pollutants and heavy metals, however, further research is needed from molecular to ecosystem level to demonstrate its efficiency by studying plant processes and associated rhizospheric microorganism and their interactions. Generally, organics are investigated by engineers while inorganics are investigated by biologists lacking integrative approach and data. Similar situation is practiced in Pakistan. Concept and categorization of PHYTOPET (phytoremediation for petroleum hydrocarbons) and PHYTOREM (phytoremediation of metals and metalloids) are highly recommended in the country by considering climatic, biotic, and edaphic factors. Research on phytoremediation also highlighted disadvantages and challenges that phytoremediation faced in the country and addressed glaring difficulties in managing biomass after successful implementation of phytoremediation of heavy metals. Different plant parts have different accumulated metal concentrations (Jamali et al., 2007). This issue refers to the disposal of plants after the phytoremediation process. Plants in this situation have heavy concentrations of toxic metals in their various tissues and are unfit to be fed to animals or to be used by humans for any purpose. Therefore, strict guidelines need to be followed to get rid of the harvested plants after the process of phytoremediation. In addition, reservations have been reported by other scientists in Pakistan who have estimated that phytoremediation can take longer time to clean up contaminated sites. That's why there is need of integrating this solar-driven technique with latest engineering-based techniques to speed up decontamination of polluted environments in the country. Nevertheless, its inexpensive and green nature continues to make it a safe and green way of removing contaminants from our natural environment (Jabeen et al., 2009).

10 Safe Disposal of Phytoremediation Plants

10.1 Composting and Mulching

Composting and mulching are recommended in those scenarios when plants used for phytoremediation accumulate small amounts of trace elements such as Zn, Cu, Mn, Mo, B, and Fe. After composting, these trace metals will act as micronutrients for plants and will enhance soil nutrient status and plant growth of terrestrial plants.

10.2 Running Through a Woodchipper

Woodchipper can be used as a source of disposal of the phytoremediated plants. This woodchipper shreds the plants into small pieces and these pieces may be used as a fertilizer after composting.

10.3 Conversion into a Fireplace Fuel

The phytoremediated plants can be used as a source of fireplace fuel. These plants may be treated with chemicals and then subjected to fermentation process followed by heat treatment to break the molecules such as sugar, starch, and other plant molecules.

10.4 Transport and Disposal of Plants

The safe disposal of phytoremediation plants can be carried out by transferring first to facilities which are specially designed for this purpose. Designated bags are used for this purpose. These bags are then taken to waste reprocessing centers, where plants are decontaminated and processed to useful mulches.

10.5 Incineration

The phytoremediated plants can be incinerated to ash form. The ash from incinerated plants must be disposed of at a landfill, although the amount of ash generated will be even less than 1% of what would be produced if the toxic wastes were dug up for treatment. Novel approaches such as incineration in brick kilns and pyrolysis can offer promising solution for final disposal of plants used for phytoremediation of heavy metals and metalloids.

11 Conclusions and Future Perspectives

Heavy metals are becoming a part of the human environment owing to altered bio-geochemical cycles and anthropogenic activities. Hence, there is an urgent need for a socially acknowledged and economically feasible process to minimize the adverse effects of the contaminants for humans as well as for environmental health maintenance and restoration. Phytoremediation provides a green, sustainable, and inexpensive solution to our heavy metal pollution concerns. Many studies investigated the efficiency and phytoremediation potential of plant species by conducting laboratory experiments where accumulation of each contaminant is estimated without considering the role of other contaminants and rhizosphere microorganism lacking the field scale data. There is a dire need to identify the microbes closely linked with solubility of toxic heavy metals in soils. Phytoremediation is still an emerging technology. Therefore, all these studies do open avenues for future research in terms of gaining fundamental understanding of mechanisms involved in the process and identifying what other plant species can be used for pollution elimination. Efforts are also constantly being made to develop new, more effective, and economically feasible techniques for phytoremediation of metalliferous soils especially in resource limited environments.

In Pakistan, the potential of phytoremediation is not as much explored irrespective of rich diversity of plants species. Most studies conducted are limited in scope and reported contradictory results. Furthermore, a few field studies were conducted to clean up ecosystems and most studies were conducted in laboratories in the country. Although, in recent years, more research is conducted on phytoremediation, however, there is still a dire need for exploration of opportunities for practical implications of phytoremediation of heavy metals, especially on field scale. Research studies on phytoremediation of ecosystems contaminated with selenium, mercury manganese, and arsenic are less documented. Therefore, priorities should be given to explore the capability of native species to remove these metals. For a country of the global south with serious socio-economic problems, an inexpensive and green technique to clean up polluted soils and water could be a promising prospect and policy makers and scientists can use its boundless potential for the environmental cleanup of contaminants detrimental for our living ecosystems and the biodiversity. Due to heterogenous concentration and distribution of pollutants, integrative approach of phytoremediation with other engineering and chemical-based remediation techniques would provide promising solution for environmental clean-up.

Data Availability Data sharing not applicable to this article as no datasets were generated or analyzed during the current review study.

Declarations

Competing Interests The authors declare no competing interests.

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