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Heavy Metal Phytoremediation Potential of Vetiver Grass and Indian Mustard Update on Enhancements and Research Opportunities

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Abstract Heavy metal pollution in the environment compromises environmental quality and human health. Phytoremediation is an innovative, green, and affordable technique that uses plants for the removal of contaminants from soil and water. Finding suitable plants that can adequately remove heavy metals from both soil and water has been a research hotspot in recent years, and there has been a rapid development in research on the use of high biomass producing crops for this purpose. Vetiver grass and Indian mustard have emerged as plants that are effective for phytoremediation and can serve other purposes during and after their use in phytoremediation. These plants are applicable in many areas because they can tolerate varied climatic conditions, thrive on degraded lands and contaminated water bodies, are easy to cultivate, and produce high biomass. This review article evaluates the phytoremediation potential of vetiver grass and Indian mustard by providing a synthesis of studies that have investigated their use for this purpose. The review considered research articles from the past 21 years and highlights the status and possible advancements in the efficient use of these plants for the remediation of heavy metal–contaminated sites. This work is of importance because phytoremediation is still undergoing immense research to promote its applicability and acceptability. Thus, it gives information on two important plants that are very useful for phytoremediation.

Keywords Heavy metals (HMs) · Contaminated soil · Contaminated water · Indian mustard · Phytoextraction · Vetiver grass

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1 Introduction

Industrial growth, urbanization, and resource exploitation have contributed to the environmental release of different forms of pollutants that adversely affect ecosystems and environmental health. Heavy metals (HMs) are an example of these pollutants and are a global environmental concern (Ali et al., 2013; Wuana & Okieimen, 2011) because they are not biodegradable and can be highly toxic even at very small concentrations (Graziani et al., 2016; Liu et al., 2020). They are considered priority pollutants in water (USEPA, 2014) that can be released into the environment through natural (weathering, erosion, and volcanic eruptions) and anthropogenic (ore extraction and processing, sludge, sewage effluent, fertilizers, automobiles, chemical spillage, paints) processes (Ali et al., 2013; Wuana & Okieimen, 2011).

Some HMs such as cobalt (Co), copper (Cu), iron (Fe), manganese (Mn), molybdenum (Mo), and zinc (Zn) are essential trace elements for biota metabolism (Aibibu et al., 2010; Otunola & Ololade, 2020), but excess levels can affect physiological pathways (Andra et al., 2009; Zhang et al., 2020). Overall, excessive levels of HMs in soil, water, and air have negative impacts on ecological health and human beings (Iloms et al., 2020); it is, therefore, a priority to find sustainable remediation techniques for contaminated environments that are effective, eco-friendly, and cost-effective.

Phytoremediation is a promising method of remediation because it is cost-effective and eco-friendly (Ali et al., 2013; Fornes et al., 2009; Yan et al., 2020), but there is still limited knowledge on the effective optimization of hyperaccumulators for HMs. At present, research has established that plants with high biomass and moderate to high tolerance for HMs can be used to remediate contaminated soil and water (Antiochia et al., 2007; Vardhan et al., 2019). Examples of such plants are vetiver, lemongrass, sunflower, tobacco, Indian mustard, pigweed, and butterfly stonecrop (Table 1).

The current review evaluated the potential of vetiver grass and Indian mustard for phytoremediation of HM-contaminated soil and water. Emphasis was given to the tolerance mechanisms of the two plant species and possible enhancements to make them more effective for the phytoremediation of HMs. Interest in vetiver grass and Indian mustard is linked to certain unique attributes that are common to both plants. Such attributes include (i) ease of propagation, (ii) their ability to grow in a large range of climatic conditions; (iii) ability to remove multiple contaminants and HMs from contaminated sites; (iv) ability to thrive in both soil and water; (v) effective post-remediation uses such as bioenergy, essential oils, and biochar; (vi) rapid growth; (vii) less need for water; and (viii) deep root systems suitable for deeper contaminants (Graziani et al., 2016; Napoli et al., 2019; Truong et al., 2008).

Table 1 Common plants used for phytoremediation

Plant	Heavy metals	Findings	Reference
Vetiver grass	Pb, Zn, Fe, Cd, Pb, Cu, Mn, Cr,	Promising for HM remediation in urban areas; acts as erosion control	Chen et al. (2004); Banerjee et al. (2016); Suelee et al. (2017)
Sunflower	Cu, Zn, Cd, Pb, Ni, Cr, As, Fe	Very efficient for co-contaminated media	Shahandeh & Hossner (2000); Mukhtar et al. (2010); Angelova et al. (2016)
Indian mustard	Au, Cd, Cr, Cu, Pb, Zn	Suitable for multiple heavy metals in the presence of other types of contaminants	Salido et al. (2003); Clemente et al. (2005)
Redroot pigweed	Cd, U, Cu	Shows antagonistic HM uptake in multiple contaminated sites	Vandenhove (2006); Wang et al. (2018)
Berkheya coddii	Ni, Co, Cd, Pb, Zn, Pt, Pd	Increased HM concentration decreases biomass, thus decreasing HM uptake. Multiple HMs can also reduce uptake	Keeling et al. (2003); Nemutandani et al. (2006)
Sedum Alfredii	Zn, Cd, Pb	Accumulates high levels of HM from soil and water	Yang et al. (2004); Huang et al. (2012); Chen et al. (2017)
Lemongrass	Al, Zn, Cd, Pb, Cr, As, Ni	Can control and regulate HM stress, while stabilizing soil	Gautam et al. (2017); Patra et al. (2018)
Tobacco	Cd, Mn, Cu, Zn	Effective for multiple metal-contaminated sites. HM metal absorption increases under prolonged sunlight	Nagata et al. (2006) Álvarez-López et al. (2016); Yang et al. (2019); Angelova (2018)

2 Methodology

Articles on laboratory experiments, field trials, and reviews on the remediation properties of vetiver grass and Indian mustard were considered in this study. ScienceDirect and Google Scholar were used to obtain related data; articles discussing possible ways of enhancing phytoremediation were also considered. The following search keywords were used to search the databases: vetiver grass for heavy metal remediation, Indian mustard for heavy metal remediation, vetiver grass for soil and water remediation, phytoremediation, Indian mustard for soil and water remediation, enhancements for phytoremediation by Indian mustard, enhancements for phytoremediation by Vetiver grass to retrieve relevant articles published from the year 2000 to 2021. The period was considered to be sufficiently wide to capture the state of the art with regard to the remediation of heavy metal contaminated soils and water using Indian mustard and vetiver grass. A total of 152 articles were retrieved, and the abstracts were screened for their relevance in this study. After screening, a total of 100 articles were selected as the most relevant and used in this review article.

3 The Processes Involved in Phytoremediation

The environmental implications of heavy metal pollution are well documented, hence the need for remediation solutions (Ali et al., 2013; Vardhan et al., 2019; Wuana & Okieimen, 2011). Over the years, physical, biological, and chemical-based methods have been used for the remediation of contaminated water, air, and soil (Vaca et al., 2011; Masindi and Muedi, 2018). The commonly used physical and chemical methods include soil washing, soil isolation, and chemical precipitation (Vaca et al., 2011; Zotiadis & Argyraki, 2013), but these methods have limitations that hamper their efficiency. These limitations include (i) high financial cost (Otunola & Ololade, 2020); (ii) introduction of new contaminants (secondary/by-products) when chemicals are used for cleanup (Ali et al., 2013); (iii) alteration of natural properties of the remediated environment, e.g., soil structure, microbiota, and productivity during excavation (Graziani et al., 2016); and (iv) need for continuous maintenance (Vardhan et al., 2019).

Over the past two decades, advances have been made regarding the application of phytoremediation for cleaning up HMs from the environment, especially in soil, water, and wastewater (Ali et al., 2013; Chaudhry, et al., 2020; Kamusoko & Jingura, 2017; Vardhan et al., 2019). Attention to phytoremediation has largely been driven by the pursuit of its' favorable environmentally friendly and cost-effective nature compared to conventional physical- and chemical-based methods (Ali et al., 2013; Ng et al., 2016). This method involves the utilization of plants to remove, transform, or stabilize contaminants through several processes such as phytoextraction, phytostabilization, phytovolatilization, and phytodegradation.

Phytoextraction is a process by which HMs are removed (extracted) from the soil or water through absorption and adsorption into plant tissues, predominantly roots. The extracted HMs can be stored in the roots and/or translocated into shoots (Ali et al., 2013; Suelee et al., 2017). The heavy metal ions are bound by organic acids (such as histidine, cysteine, and proline) within plant cells (Yan et al., 2020); these plants can then be harvested and disposed of accordingly or used as biofuel in some cases (Yang et al., 2019). Even particulate HM forms can be phytoextracted (Thwala et al., 2021). Phytoextraction is an attractive option for the remediation of HMs because it removes the HMs from the contaminated substrate and the biomass can be collected and disposed of, thus reducing the overall contamination in the environment (Tangahu et al., 2011; Yang et al, 2019). Phytostabilization is a process that uses suitable plant roots to stabilize HMs in soil, making them less mobile, thereby preventing further environmental release. Some plant root exudates form stable metal complexes that immobilize HMs in the rhizosphere (Yan et al., 2020). Phytovolatilization is a technique whereby plants convert HMs and other contaminants into volatile forms and release them into the atmosphere (Sakakibara et al., 2010; Vardhan et al., 2019) even though the volatilized HMs still have the potential to further pollute the atmosphere via secondary pollution (Sakakibara et al., 2010). Phytodegradation is the conversion/breakdown of contaminants into less harmful forms, but this process is irrelevant for heavy metal removal because they are nonbiodegradable (Ali et al., 2013). The choice of phytoremediation technique depends on the type and degree of contamination, future use of the remediated environment, the time frame required for remediation, as well as the environmental medium (soil or water) being remediated (Ali et al., 2013; Kamusoko & Jingura, 2017).

4 Challenges of Phytoremediation and Enhancement Strategies to Overcome Them

Some plant species thrive easily with high levels of HMs from surrounding soil and water in their harvestable parts without experiencing significant toxicity (Ali et al., 2013; Tangahu et al., 2011; Truong et al., 2008). These plants, used for phytoremediation, are known as metallophytes used for phytoremediation and can be grouped into three namely metal excluders, metal hyperaccumulators, and metal indicators. The rhizosphere is the environment where microorganisms, plant roots, and soil–water interactions occur, resulting in the formation of chelating agents, hydrogen ions, root exudates, and metabolites (Graziani et al., 2016; Kim et al., 2010). These processes can alter the pH of the soil solution, thus encouraging the solubility and bioavailability of nutrients and HMs in the soil (Chen et al., 2017; Nedjimi, 2021). During the process of phytoextraction by plants, HMs from the soil solution are absorbed into the plant roots and then transported to the shoots through the plasma membrane (symplastic pathway) or the cell wall (apoplastic pathway) (Diwan et al., 2008; Nedjimi, 2021). Thereafter, a proportion of the HMs enter the cells while the remaining are bound to cell walls (Antiochia et al., 2007; Prasad & Strzalka, 2013).

Nonetheless, phytoremediation of HMs can sometimes be limited by factors such as low biomass production of hyperaccumulator plants, reactive oxidative stress (as a result of HMs pollution), low bioavailability of HM in soil/water, low or high pH in the remediated medium, and the risk of pests and diseases that may negatively affect plants during remediation (Ali et al., 2013; Fulekar et al., 2009). Such factors restrict the upscaling of phytoremediation, although recently, innovative approaches are being developed to overcome these limitations and improve the efficiency of suitable plants for this purpose (Fulekar et al., 2009; Graziani et al., 2016; Ju et al., 2020). A summary of strategies to enhance the phytoextraction capacity of plants is given in Table 2.

5 The Use of Indian Mustard for Phytoremediation of Heavy Metals

Indian mustard is classified as a metallophyte because it can adapt and thrive in metal-contaminated soils (Ali et al., 2013; Li et al., 2019). It has attracted attention for its ability to remove HMs from naturally and artificially contaminated soil and water (Huysen et al., 2004; Li et al., 2019; Meyers et al., 2008). Indian mustard can accumulate HMs in its roots and shoots (Rathore et al., 2019), however to a higher extent in roots (Li et al., 2019; Meyers et al., 2008). However, translocation to shoots increases as root uptake increases due to increased heavy metal concentrations in the surrounding environment (Li et al., 2019; Meyers et al., 2008; Napoli et al., 2019). The HM tolerance of Indian mustard can be associated with its high concentration of antioxidants, cysteine, and ascorbic acid, which protect against reactive oxidative stress that may hamper its growth and performance (Diwan et al., 2008; Fryzova et al., 2017). Additionally, these species possess effective root cell vacuolar storage systems that aid adaptation to excessive HM levels (Graziani et al., 2016; Meyers et al., 2008).

Over the years, Indian mustard has been used to remediate single and multiple metal-contaminated sites. For example, uranium (U) was effectively removed from contaminated soils during a 60-day trial by Li et al. (2019). Direct-current voltage was applied for 9 days to enhance U uptake. This significantly enhanced plant growth and biomass yield increased by 6% in UO_3 spiked soils and 1.3% in UO_2 spiked soil, resulting in up to a 6% increase in removal efficiency. The enhanced plant growth can be attributed to changes in soil structure and conditions such as pH and nutrient mobilization after electrokinetic energy was applied (Cang et al., 2011). The type of electric current may affect the type of reaction in the soil and the behavior of Indian mustard and its metal accumulation capacity. Indian mustard has a high tolerance for Pb as it can tolerate Pb levels as high as 1000 mg/kg (Graziani et al., 2016), indicating that this plant is indeed a hyperaccumulator according to the definition of Baker and Brooks (1989). In a study by Graziani et al. (2016), 14-day-old vetiver grass seedlings with bifurcated roots were transplanted into Pb-spiked agricultural soil contained in rhizoboxes. This experiment was undertaken in order to visualize the changes in the rhizosphere as vetiver

Table 2 Possible enhancements for plants and contaminated media to improve heavy metal uptake by plants

Enhancement	Advantage	Challenges	Media (soil/water)	Reference
Electrokinetic energy	A Low-intensity current is required to lower pH. Increases biomass. It encourages biodegradation of organic co-contaminants that may be present	If too high a current is applied, this can damage the microbial community in treated media. Contaminants may diffuse to low permeable regions making remediation difficult	Soil, Water	Gill et al. (2014); Cameselle and Gouveia (2019); Li et al. (2019)
Intercropping	Reduces the risk of diseases and maximizes the availability of soil nutrients. It is an economic approach that improves plant biomass	HM accumulation may occur in some food crops, along with phytoextraction plants	Soil	Kidd et al. (2015); Luo et al. (2020); Xiao et al. (2020); Tang et al. (2020)
Amendments/ additives	Reduce stress and HMs phytotoxicity levels in plants. Can immobilize HMs in bulk soil. Generally improves plant growth and biomass also reduces pathogens	High levels of chelates can over-mobilize contaminants which can easily migrate into groundwater. Likewise, excessive addition of clays and biochar can change soil/water composition and functioning	Soil, water	Liphadzi & Kirkham (2006); Fornes et al. (2009); Ng et al. (2016)
Genetic engineering	Improves the plant's overall phytoremediation capacity. Can be designed to satisfy the specific needs of a particular remediation project. Develops plants that are resistant to diseases	Could be expensive and can negatively affect biodiversity	Soil, water	Fulekar et al. (2009); Daghan et al. (2013)

grass grew. At the end of the experiment, Graziani et al. (2016) observed that increasing soil Pb content correlated with an increase in Pb concentration in the upper parts of Indian mustard and that HM accumulation capacity may not necessarily be affected by rhizosphere pH (Chen et al., 2020; Graziani et al., 2016). Indian mustard can also be classified as a Cu tolerant plant that can translocate notable amounts of Cu from roots to shoots. An 80% of Cu uptake from soil was achieved within 32 days (Napoli et al., 2019).

Kim et al. (2010) studied HM uptake by the roots of Indian mustard in a rhizobox to understand the processes that take place in the rhizosphere, using acidic and alkaline soils. There were no significant changes in soil pH, but dissolved organic carbon significantly increased in both soils, leading to an increase in available organic acids, which in turn increased Cd, Cu, Pb, and Zn uptake 35 days after plant germination. The presence of multiple metals led to competitive uptake, with increased immobilization being observed for Cd and Zn. This is attributed to the formation of chemical bonds during the interaction of rhizosphere solutions and HMs, thus preventing the HMs from being absorbed into the shoots (Wu et al., 2020; Yang et al., 2019). According to Chigbo et al. (2013), the phytoextraction efficiency of Indian mustard may be lowered in environments contaminated with multiple HMs or contaminant types, due to competitive uptake and interactions between contaminants with different characteristics. Chigbo et al. (2013) observed an up to 85% decrease in Cu accumulation by Indian mustard and a decrease oinbiomass in the presence of pyrene. The decrease in Cu accumulation was probably due to the reactions of complexes with root exudates and pyrene (Jeelani et al., 2020), resulting in the formation of insoluble Cu complexes, thus limiting Cu uptake.

Besides Indian mustard's capability to extract HMs, it also indicates phytostabilization properties, suggesting that Indian mustard can be used for multiple phytoremediation techniques. This was confirmed by Huysen et al. (2004), but Clemente et al. (2005) concluded that Indian mustard is not a recommended plant for phytoextraction because at the end of their study, the projected average number of years for optimal remediation of Cu, Pb, and Zn was determined to be 30 575, 192 800, and 9150 years, respectively, which are not practical time frames for remediation. Clemente et al. (2005) differed from the results reported by other studies (Chen et al., 2020; Ma et al., 2009; Mohamed et al., 2012) possibly due to different experimental conditions (natural and greenhouse), soil parameters including pH, bioavailable HMs, and nutrients. Other natural factors such as light, humidity, and temperature may also be responsible for the varying results (Chintani et al., 2021).

Indian mustard possesses varying HM accumulation patterns in its roots and shoots (Clemente et al., 2005; Mhalappa et al., 2013). For instance, the accumulation of Pb and Cd is relatively higher in root systems compared to that of the stem and leaves (Meyers et al., 2008; Wu et al., 2020). Clemente et al. (2005) observed high Pb concentration in the roots of Indian mustard, whereas in the shoots, it was below the detection limit; while Cu, Fe, Mn, and As were found to have higher accumulation in the leaves. Recently, Raj et al. (2020) observed Hg uptake by Indian mustard in a 90-day study. With time, the plant demonstrated high Hg tolerance. Uptake was highest in the roots, followed by leaves and stems for the last 60 days (2 months) of the experiment. Mhalappa et al. (2013) also observed that Zn accumulation was highest in the roots, while Pb was highest in shoots, further indicating that accumulation is dependent on the metal type and influenced by other present contaminants (Masindi and Muedi, 2018). Roychowdhury et al. (2017) reported that the Indian mustard plant accumulates HMs mostly in leaves followed by roots and then stems in the order: Zn > Cu > Pb.

At times, Indian mustard may experience stunted growth, especially in environments with multiple contaminants (Chen et al., 2020; Fornes et al., 2009; Goswami & Das, 2015; Mhalappa et al., 2013). According to Goswami and Das (2015), extreme HMs toxicity can negatively affect plant biomass, root and shoot length, and chlorophyll and carotenoid performance. Mohamed et al. (2012) also observed that Cd toxicity can hamper plant growth and biomass, although Indian mustard still showed good potential as a Cd hyperaccumulator. Sridhar et al. (2005) observed that metal uptake increased with increasing concentration, although toxicity effects and slightly stunted growth were observed. Likewise, Qadir et al. (2004) observed growth inhibition of Indian mustard following 3-day exposure to Cd, although Cd accumulation increased. To avoid the possible stunted growth in Indian mustard, the phytoremediation process can be enhanced with several options discussed in Sect. 4. A significant increase in biomass has been achieved upon the application of various enhancements for phytoextraction, especially in soil (Chen et al., 2020; Fornes et al., 2009; Ng et al., 2016). In environments with multiple contaminants, Indian mustard accumulates HMs more in the shoots and soil enhancements such as biochar and clay minerals which can increase plant growth (biomass increase) significantly (Otunola & Ololade, 2020; Zhang et al., 2020). These amendments should however be minimal as some (e.g., EDTA) can lower soil pH which may result in plant mortality or increased infiltration of contaminants into groundwater. However, biochar and clays are known to improve plant growth and increase pH, thereby limiting the further release of HMs into water bodies (Otunola & Ololade, 2020; Zotiadis & Argyraki, 2013). Composting can also improve the health of Indian mustard in HM-contaminated soils. Fornes et al. (2009) observed that alperujo compost (solid–liquid waste from olive oil extraction), combined with sheep manure at a ratio of 80:20, could increase the bioavailability of aluminum (Al), As, Pb, and Zn in both acidic and calcareous soils. Although 16 weeks after compost amendment, Indian mustard did not survive in an acidic soil type, while other Brassicaceae species survived. Indian mustard may not have survived the acidic soil because the plant itself releases high organic acids in its roots (Diwan et al., 2008); therefore, the pH in the rhizosphere may have been too low for the plant to survive even in the absence of HMs. But Indian mustard survived at a 100% rate in calcareous soil of higher pH (Fornes et al., 2009).

Shahandeh & Hossner (2000) confirmed EDTA to be the most effective chelating agent for increasing the uptake of Cr in plant shoots, especially in Indian mustard and sunflower. Similarly, Mbangi et al. (2018) observed that the addition of EDTA increased Cr concentration in Indian mustard shoots by more than 50-fold and more than tenfold in sunflower, making Indian mustard a preferred candidate crop for Cr remediation. Likewise, Salido et al. (2003) used Indian mustard to extract Pb from the soil in a greenhouse experiment and found that EDTA improved Pb extraction. Initial soil Pb concentration was 338 mg/kg, and within 3 months, Indian mustard extraction of Pb increased by 125% with EDTA dosage as low as 0.6 mmol/kg. Phytoextraction rate was further increased with increasing EDTA dosage and Pb was observed to accumulate more in the shoots, confirming earlier suggestions, that EDTA enhances the accumulation of HMs in the shoots (Shahandeh & Hossner, 2000).

The ability of Indian mustard to extract Cd and U from contaminated soil with added plant growth regulators (PGR) was confirmed in a greenhouse experiment (Chen et al., 2020). The HMs initially reduced biomass, but this was later increased by 78.5% for the shoot and 55.5% for roots after the addition of 6-BA, an exogenous PGR. Chaudhry et al. (2020) exposed Indian mustard to soils with varying levels of Zn (20, 40, 80, and 160 mg/kg) for 4 weeks. The tolerance index (TI) was observed to increase with increasing Zn concentration, and the translocation factor (TF) was greater than one, indicating that the shoots can accumulate more Zn and that Indian mustard is a suitable hyperaccumulator for Zn (Minisha et al., 2020). Huysen et al. (2004) explored the selenium (Se) phytoextraction potential of transgenic Indian mustard and found that genetically modified Indian mustard can overexpress ATP sulfurylase, a plant enzyme that promotes translocation of Se into Indian mustard shoots. After 70 days, transgenic Indian mustard had a 2.5% higher Se extraction rate than the unmodified plants. This result was achieved in both soil and water remediation experiments (Huysen et al., 2004).

Genetic modification has been used extensively to enhance phytoextraction by Indian mustard (Bañuelos et al., 2005; Ma et al., 2009). Indian mustard seeds were inoculated with a strain of AX10 plant growth-promoting bacteria by Ma et al. (2009). This bacterial strain is Cu tolerant and thus prevented HMs stress in Indian mustard. Up to 56% more Cu was extracted by the inoculated crop compared to the non-inoculated counterparts (Ma et al., 2009). Bañuelos et al. (2005) got similar results after using transgenic plants to improve Se uptake: transgenic plants exhibited a 30% increase in Se uptake compared to the normal, unmodified plant.

Most studies on the use of Indian mustard for the phytoremediation of HMs have focused on its usefulness in soil remediation. Few studies in which Indian mustard was considered for water and wastewater remediation confirm that this plant can take up high amounts of Pb (up to 138 g/kg Pb) in its roots (Huysen et al., 2004; Meyers et al., 2008; Qadir et al., 2004). Since accumulation was more in the roots, root microbial inoculation could be a good enhancement to improve the root system and improve HM removal in water (Ma et al., 2009). Mercury (Hg) is a highly toxic water contaminant that is not often treated by phytoremediation, but some studies have used Indian mustard to treat Hg-contaminated water and wastewater. They found that Indian mustard is an efficient plant with adequate metabolic defense and adaptation to Hg stress (Ansari et al., 2021; Shiyab et al., 2009). Ansari et al. (2013) confirmed the absorption of Cu by Indian mustard within 20 days in a hydroponic setup. According to Khan et al. (2009) and Yang et al. (2021), Indian mustard is a suitable candidate for the removal of As from water, and the presence of As increased the activities of antioxidative enzymes, making the plant more HM tolerant. Yang et al. (2021) carried out a greenhouse experiment that was set up to investigate HM absorption by Indian mustard. Seedlings were cultivated in 250-ml Hoagland's solution for 2 weeks, then transferred into Hoagland's solution spiked with As and Pb of up to 30 mg/L and 82 mg/L, respectively. After 3 days, the plants were harvested and tested for As and Pb absorption. Concentrations of 1,786 mg/kg and 47,200 mg/kg of As and Pb, respectively, were absorbed by Indian mustard. These concentrations, however, were reduced by > 90% for As, and ~10–30% for Pb when both of the HMs were present in the same solution. This is an indication that the tolerance capacity of Indian mustard may be reduced when certain HMs are present simultaneously (Yang et al., 2021). It can be said that Indian mustard is a reliable hyperaccumulator for HMs, suitable for the remediation of both soil and water.

6 The Use of Vetiver Grass for Phytoremediation of Heavy Metals

Vetiver grass (*Chrysopogon zizanioides*) belongs to the family Poaceae along with maize, sorghum, sugarcane, and lemongrass (Truong et al., 2008; Danh et al., 2009). Vetiver is a herbaceous, perennial crop native to India that has been adopted in many countries, being a noninvasive species (Truong et al., 2008). Given its relatively rapid growth, high biomass, and good tolerance to a wide array of pollution effects, vetiver holds attractive characteristics for phytoremediation of HMs (Lai & Chen, 2004; Danh et al., 2009; Aibibu et al., 2010; and Singh et al., 2017). Vetiver is recognized as an ideal species for revegetation in tailings (Chiu et al., 2006; Vargas et al., 2016) and has been found superior to other grass crops including maize, rainbow pink, and common reed (Phragmites australis) for HM removal in soil and water (Chiu et al., 2005; Danh et al., 2009; Wilde et al., 2005). Similar to many other phytoremediation plants, Vetiver roots are HM accumulation sites, accumulating as many as two-fold shoot concentrations in its roots (Singh et al., 2017) and exhibiting low translocation rates (Aibibu et al., 2010; Andra et al., 2009; Chiu et al., 2005; Gravand et al., 2021). Although vetiver has been observed to be an efficient hyperaccumulator of HMs with high translocation rates for some HMs (Banerjee et al., 2016; Gautam & Agrawal, 2017; Ng et al., 2016), others have observed TF below one (Chen et al., 2004; Gravand et al., 2021).

Vetiver has been evaluated for its potential application in water and wastewater remediation and was found to be effective for the removal of HMs (Aibibu et al., 2010; Zhang et al., 2014). Singh et al. (2017), investigated the absorption of As by Vetiver grass in a hydroponic system with 61 ppm As over 14 days. Up to 37 ppm was accumulated with the plants not exhibiting significant toxicity effects due to enhanced antioxidative capacity. Likewise, in soils, Cd has been observed to improve chlorophyll and the rate of photosynthesis of Vetiver grass (Zhang et al, 2014). Cd did not influence the translocation of most other HMs in vetiver; Fe was the only heavy metal whose translocation rate was lower in the presence of Cd (Zhang et al, 2014). In a hydroponic system observed by Aibibu et al. (2010), 1 mg/L Cd exposure over 15 days enhanced vetiver chlorophyll contents, root activity, and growth (2.2% biomass increase). Cd accumulation increased with increasing Cd solution concentration; however, the translocation factor was less than one; hence, they conclude that vetiver grass is a metal excluder.

Vetiver grass has also been applied for the remediation of acid mine water. It increased the pH, while reducing concentrations of Fe, Zn, Cu, and SO_4^{2-}, but the uptake of Pb, Al, and Ni was lower (Kiiskila et al., 2019). In another study, Vetiver grass removed Cu and Zn to a higher extent than Fe (Roongtanakiat et al., 2007). This could be explained by the formation of iron plaques on the roots of vetiver grass, thus preventing uptake (Yang et al., 2019). It was shown that 27%, 53%, and 88% of Fe, Zn, and Cu, respectively, were removed from industrial wastewaters after planting for 120 days. Iron plaque formation is suspected to be responsible for the lowered root-to-shoot movement of HMs (Roongtanakiat et al., 2007) because it increases the metal stabilization in the roots, thus preventing translocation (Kiiskila et al., 2019). Wastewater from the manufacture of batik fabrics was also successfully remediated using vetiver grass. Vetiver successfully removed 40% Cr within 56 days, while improving ammonia, biochemical oxygen demand (BOD), and chemical oxygen demand (COD) contents in the wastewater (Tambunan et al., 2018), illustrating that vetiver can be applied for remediation of multiple contaminants. The optimal pH for vetiver has been suggested to be pH 4–6, whereas above pH 9 performance could be inhibited (Kiiskila et al., 2019; Tambunan et al., 2018).

Several approaches have been applied to enhance the phytoremediation capacity of vetiver in soil and water. Commonly applied is the addition of organic acids, manure, biochar, and composts in the environmental media (Andra et al., 2009; Chiu et al., 2005; Ng et al., 2016). Specifically, Ng et al (2016) conducted a greenhouse experiment to investigate the effects of EDTA, elemental sulfur, and ammonium nitrate fertilizer (N-fertilizer). In this experiment, topsoil from a field was spiked with 50 mg/kg Cd and 100 mg/kg Pb using $Cd(NO_3)_2.H_2O$ and $Pb(NO_3)_2$. One-week-old seedlings were transplanted into 2-kg plastic pots containing the spiked soil and 50-mL water was supplied per day for 60 days. The application of phytochelatins improved vetiver tolerance of Pb, and up to 3 000 mg/kg was accumulated without signs of toxicity exhibited by vetiver after 7 days, although phosphorus in soils can inhibit Pb uptake (Andra et al., 2009). The application of direct and alternating current can significantly increase vetiver bioconcentration factor and HM uptake by up to 65% (Siyar et al., 2020). Studies have confirmed that the effectiveness of vetiver grass for HMs extraction can be enhanced by increasing planting density. Longer roots and higher plant densities increase the surface area for absorption of HMs, thereby increasing the amount of metals absorbed per time (Hasan et al., 2017; Suelee et al., 2017).

In a 60-day pot experiment, vetiver grass was grown in Mn-, Zn-, Pb-, Cu-, and Cd-polluted soil amended with vermicompost (Jayashree et al., 2011). It was observed that vetiver grass tolerated high levels of HMs and biomass yield increased as vermicompost dosage increased. Plant growth reduced soil pH, resulting in higher HM extraction rates because lower pH increased the bioavailable fraction of the HMs. Lai and Chen (2004) used EDTA to increase the bioavailability of HMs in soils in a 14-day greenhouse experiment. The addition of EDTA significantly improved Cd and Zn uptake by Vetiver grass, but the roots of vetiver could not accumulate Pb, possibly because plants generally do not readily take up soil Pb fractions (Wuana & Okieimen, 2011).

In another study, the addition of EDTA 1 week before harvest significantly increased the amount of Pb phytoextraction from the soil; Pb levels of 1390–1450 ppm were observed in plant tissue samples (Wilde et al., 2005). Ye et al. (2014) also observed that soil washing using maize oil before planting vetiver significantly improved microbial function, growth of vetiver, and therefore, HM uptake in Cd- and Pb-contaminated soil after 30 days. The addition of nitrilotriacetic acid can enhance the uptake of As, Zn, and Cu by vetiver, while hydroxyethyl-iminodiacetate can increase Cu bioavailability in the soil solution (Chiu et al., 2005). Exposure duration may be considered an important factor in successful phytoremediation. Chiu et al. (2005) highlighted that the highest HMs' uptake occurs between 16 and 20 days, since, on the first day, 15.749 mg/kg Cu was extracted by vetiver, followed by 54.427 mg/kg on the 16th day and finally 55.18 mg/kg on day 20. Similar to the observation of other researchers, root uptake of Cu was significantly greater than shoot uptake, but nitrilotriacetic acid increased the translocation of HMs from roots to shoots and no stress was observed in vetiver.

According to Lai & Chen (2004), vetiver grass can be regarded as a potential phytostabilization plant that can be grown in a site contaminated with multiple HMs except for its low preference for Pb, whereas Ng et al. (2016) successfully achieved Pb accumulation in the shoots of vetiver, with Pb being the only contaminant present. EDTA and N-fertilizer enhanced the remediation capacity of vetiver in Pb-polluted soil, thereby increasing Pb uptake in shoots and translocation factor to 1.72 and 2.15 in both treatments. They also noted that elemental sulfur may inhibit the bioavailability of Pb and its translocation from soil to root and root to shoot (Ng et al., 2016). Likewise, Gravand et al. (2021) achieved 83% Pb uptake from soils within 5 months of planting vetiver, while the percentage of absorption for Cd, Ni, and Mn were 53.2%, 65.5%, and 61%, respectively. There was also a minimal difference in the amount of Pb extracted by the root and shoot of vetiver. The Pb concentrations in the roots and shoots were 363.01 mg/kg and 300.39 mg/kg, respectively.

Vetiver grass can grow well in mine tailings containing high levels of HMs. For over 4 months, Chiu et al. (2006), improved the tailing conditions by adding manure compost and sewage sludge. This significantly increased the vetiver biomass yield compared to sewage sludge. The study concluded that manure compost and sewage sludge could significantly reduce Pb uptake and accumulation by vetiver, whereas Cu and Zn uptake was improved by more than 10%. Such findings suggest that the application of amendments to enhance phytoremediation capacity still requires further refinements as different amendment types have varying effects on the behavior of HMs. Banerjee et al. (2016) also confirmed the suitability of vetiver for mine soil and waste dumps. They observed that although HMs reduced chlorophyll contents, certain enzyme activities were improved, and the translocation factor of Cu was greater than one, likewise its bioconcentration factor in vetiver shoot.

According to Gautam & Agrawal (2017), red mud (a by-product of alumina production from bauxite) can significantly improve the growth of vetiver in contaminated soil. It improved the metal tolerance index of vetiver by over 100%. Red mud and sludge increased soil organic matter, but soil HM contents (Fe, Mn, Mg, Zn, Cu, Ni, Pb, Cd, and Cr) increased with increasing red mud dosage, although their bioavailability was reduced. Vetiver effectively removed Mn and Cu with translocation factors between 1 and 1.3 for both metals, but the bioconcentration factors observed for all HMs were less than one, indicating that vetiver is a metal excluder. Its inability to extract any of the HMs is possibly due to the adsorptive properties of red mud and sludge which reduce the bioavailable fractions of the HMs.

Antiochia et al. (2007) considered vetiver to be an excellent hyperaccumulator for Pb and Zn, but not efficient for Cr and Cu uptake. Vetiver grass was irrigated with HM solution containing 623 ppm Cr, 190 ppm Cu, 621 ppm Pb, and 653 ppm Zn for 30 days. Shoot uptake of Cu was about 3 times higher than root uptake. Better shoot uptake might be because the contaminants were from irrigation water and are not yet bound to soil particles, thus rapidly adsorbed to shoots. In comparison to common reed (*Phragmites australis*), Danh et al. (2009) found that vetiver grew better and produced double the biomass of common reed. Amendments, however, reduced Pb uptake and accumulation, but not Cu in both vetiver and common reed. The shoot Pb concentrations of vetiver exposed to 400 and 1,200 mg/L Pb were 25 and 150 mg/kg Pb, respectively. Vetiver grass can be considered a good hyperaccumulator for HMs and other contaminants. Asides from this, the plant is useful for slope stabilization, flood control, etc. Therefore, it is multifunctional in the environment.

7 Conclusion and Recommendations

Phytoremediation has been proven to be a cost-effective and eco-friendly, easy-to-manage technique. The use of fast-growing terrestrial crops such as vetiver grass and Indian mustard for soil and water remediation is gaining attention. This review revealed that vetiver grass and Indian mustard use different mechanisms to tolerate HMs. This depends on the substrate being remediated, heavy metal type, presence or absence of co-contaminants, plant cultivar type, climate, and enhancements applied. This study highlights that suitable enhancements can improve the biomass production and heavy metal uptake capacity of vetiver grass and Indian mustard, making both plants affordable and eco-friendly remediation options. However, the following recommendations will be relevant for future studies to promote these plants' real-life, field-scale use for phytoextraction purposes:

- There are various options to improve/enhance the performance of Indian mustard and vetiver as heavy metal remediation plants. However, the use of amendments and intercropping may be the best option because they are effective, more straightforward to apply, and less costly compared to microbial inoculation, genetic modification, and electrokinetic enhancement.
- Amendments such as clay minerals with adsorptive properties can serve multiple purposes: (i) improve plant growth, therefore biomass and capacity to extract HMs, (ii) reduce the mobility of HMs to prevent further contamination of water resources before plants completely take up the

HMs, (iii) improve entire soil structure. Therefore, the use of clay minerals as soil amendments to enhance the remediation capacity of Indian mustard and Vetiver could be investigated in future studies.

- Intercropping can be very efficient for co-contaminated or multiple heavy metal contaminated sites. Elucidating the effectiveness of intercropping on phytoremediation still requires extensive studies.
- Unlike the case of vetiver grass, a limited number of studies have investigated Indian mustard for water and wastewater remediation. Therefore, future studies could focus on investigating the application of Indian mustard for water remediation.
- The variations in experimental conditions such as a greenhouse, naturally and artificially contaminated media, duration of the experiment, and environmental factors make it difficult to formally establish the application of these plants in real-life scenarios. With this in mind, more research on the field-scale application of Indian mustard and vetiver for heavy metal remediation is needed.

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Declarations

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