CHAPTER 4

Recent advances in phytoremediation of arsenic-contaminated soils

Xin Wang & Lena Qiying Ma

4.1 INTRODUCTION

Arsenic contamination in soils occurs widely in a range of ecosystems resulting from geological origins and anthropogenic activities. On average, arsenic concentration ranges from 5 to 10 mg kg\(^{-1}\) in uncontaminated soils and above 10 mg kg\(^{-1}\) in contaminated soils (Hossain, 2006). Increased buildup of arsenic in irrigated soils has been widely recognized in South and South-east Asia (Brammer and Ravenscroft, 2009), posing significant threats to agriculture sustainability. In Bangladesh, long-term irrigation with arsenic-rich groundwater from shallow aquifers in dry season adds >1000 tons of arsenic to the agricultural soils (Ali et al., 2003). In addition, arsenic contamination in soils results from various anthropogenic activities, such as mining and smelting (Williams et al., 2009), and using arsenic-containing wood preservatives (Chirenje et al., 2003), pigment, pesticides, herbicide (Sarkar et al., 2005) and feed additives (Arai et al., 2003).

As a cost-effective and ecology-friendly technology, phytoremediation of arsenic-contaminated soils has been widely studied. Among phytoremediation technologies, phytoextraction and phytostabilization are two predominant approaches in remediation of soils contaminated with heavy metals. Phytoextraction takes advantage of plants to remove contaminants from soils by concentrating the targeted contaminant to the harvestable tissues (Salt et al., 1998). To achieve effective arsenic removal from soils, the plant should be highly tolerant to arsenic and efficient in accumulating arsenic into sufficient aboveground biomass. Therefore, phytoextraction efficiency depends on both aboveground biomass yield and plant arsenic concentration. Bioconcentration factor (\(BF\)), which is defined as the ratio of element concentration in plant shoots to that in soil, has been used to measure a plant’s efficiency in phytoextraction. Based on mass balance calculation, phytoextraction is feasible only by using plants with \(BF\) much greater than 1, regardless of how large the harvestable biomass (McGrath and Zhao, 2003). Furthermore, to achieve efficient removal of contaminant in a reasonable time frame with high plant survival and biomass yield, the initial and target soil contaminant concentrations should be taken into account to predict the applicability of phytoextraction, which is in most cases appropriate for soils with low contamination (Zhao and McGrath, 2009).

For heavily contaminated sites (e.g., industrial and mining degraded sites), indigenous tolerant species with extensive root system and low translocation factor (\(TF\), the ratio of contaminant concentration in shoots to that in roots) provide valuable plant resources to immobilize the pollutant in the rhizosphere, and simultaneously stabilize the degraded sites by establishing vegetation cover. Soil amendments, in some cases, are essential to assist the success of the survival of pioneering species by mitigating contaminant toxicity and improving substrate conditions (Vangronsveld et al., 2009). In this way, ecological restoration of contaminated sites can be gradually achieved through revegetation, which is termed as phytostabilization.

Besides these two major phytoremediation techniques, other methods include phytoexclusion and rhizofiltration. To remediate large-scale agricultural soils contaminated by arsenic, phytoexclusion is more practical to reduce arsenic transfer from soil to crops. Based on the well-established knowledge with regard to arsenic biogeochemistry and arsenic transport mechanisms in rice, a range of strategies including water management, Si fertilization, and rhizosphere manipulation...
have been studied to mitigate arsenic contamination in soil-rice system (Kertulis et al., 2005; Ma et al., 2008; Zhao et al., 2010). To prevent loading of arsenic into soils from irrigating water, which has been well recognized in South and South-east Asia, arsenic phytofiltration technique, using arsenic hyperaccumulators or aquatic plants with substantial arsenic accumulating ability can be taken into account for arsenic reduction in irrigation water (Rahman et al., 2007).

In this chapter, phytoextraction and phytostabilization using As hyperaccumulators and tolerant species in both greenhouse- and field-scale studies are discussed from different perspectives. With prevalent hazards of arsenic in South Asia, potential phytoexclusion strategies are proposed to mitigate arsenic contamination in a soil-rice system. Furthermore, the feasibility of using phytofiltration to reduce continuous loading of arsenic into agricultural soils is discussed.

4.2 PHYTOEXTRACTION OF ARSENIC CONTAMINATED SOILS

As the first known arsenic hyperaccumulator, *Pteris vittata* L. (Chinese brake fern) exhibits great potentials to extract arsenic efficiently from contaminated soils and concentrate >90% of the arsenic in the fronds with BF up to 126 and little toxicity (Ma et al., 2001). Furthermore, the plant grows fast and yields considerable biomass, particularly in tropical and subtropical areas. These constitutive traits of *P. vittata* have attracted extensive attention regarding its application in phytoextraction of arsenic-contaminated sites. Most studies have demonstrated effective extraction of arsenic by *P. vittata* in a reasonable time frame with different experiment scales (Gonzaga et al., 2008; Kertulis-Tartar et al., 2006; Tu et al., 2002). The effective decontamination of arsenic-contaminated soil by *P. vittata* is largely attributed to its unique metabolism traits, through effective mobilization of arsenic in the rhizosphere, efficient uptake by the roots and translocation to the fronds. Following the discovery of *P. vittata*, additional 11 fern species belonging to Pteridaceae family have been identified to hyperaccumulate arsenic, providing alternative plant resources for phytoextraction of arsenic from contaminated soils (Table 4.1). As detailed in this section, phytoextraction using *P. vittata* is discussed with the underlying mechanisms.

4.2.1 Efficient arsenic extraction by *P. vittata*

*P. vittata* is known for its effectiveness in arsenic phytoextraction from soils. In a greenhouse experiment, *P. vittata* were grown in an arsenic-contaminated soil containing 98 mg As kg$^{-1}$. After 20 weeks of growth, ~26% of soil arsenic from each pot (1.5 kg soil pot$^{-1}$) was removed.

| Table 4.1. Arsenic hyperaccumulators identified up to date in Pteridaceae family. |
|----------------------------------|----------------|----------------|----------------|
| Species                         | Frond arsenic | Soil arsenic  | Reference      |
|                                 | concentration | [mg As kg$^{-1}$] |               |
| *Pteris vittata*                | 2500–22630    | 50–1500        | Ma et al. (2001) |
| *Pteris cretica*                | 6200–7600     | 500            | Zhao et al. (2002) |
| *Pteris umbrosa*                |               |                |                 |
| *Pteris longifolia*             |               |                |                 |
| *Pteris biaurita*               | 1770–3650     | 100            | Srivastava et al. (2006) |
| *Pteris quadriaurita*           |               |                |                 |
| *Pteris ryukyuensis*            | 4000          | 200            | Wang et al. (2007) |
| *Pteris aspericaulis*           | 3275          |                |                 |
| *Pteris fauriei*                | 1365          |                |                 |
| *Pteris oshimensis*             | 3942          |                |                 |
| *Pityrogramma calomelanos*      | 2270–6380     | 20–8800        | Visoottiviseth et al. (2002) |
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by *P. vittata* with frond arsenic up to 7,230 mg kg$^{-1}$ and BF of 74 (Tu et al., 2002). In a separate greenhouse study investigating the effect of repeated harvests on arsenic removal by *P. vittata*, ferns were grown in six arsenic-contaminated soils with total arsenic ranging from 22.7 to 640 mg kg$^{-1}$ (Gonzaga et al., 2008). With the application of extended time-release base fertilizer (N:P:K ratios of 18:6:12) at a rate of 2 g kg$^{-1}$ soil, the ferns were well-established after 4 months of growth and produced a good frond biomass ranging from 24.8 to 33.5 g plant$^{-1}$ with frond arsenic being 66 to 6,151 mg kg$^{-1}$ and BF being 4.7 to 48 from the first harvest (October, 2003, 4 months after transplant). During the harvest, all aboveground biomass was removed, making it difficult for the plants to re-grow under a cooler climate in the second growing period. As a result, 34–75% lower frond arsenic concentrations and 40–84% less frond biomass were obtained upon the second harvest (April, 2004). The results indicate that, though *P. vittata* is effective in arsenic extraction, proper growth timing and harvest method (leaving some fiddleheads for faster regrowth) is important to achieve optimum phytoextraction.

This conclusion is supported by a two-year field study of Kertulis-Tartar et al. (2006) who found that it is necessary to leave the fiddleheads (young fronds) as well as few live fronds at harvest to facilitate the regeneration and survival of ferns in winter season. Furthermore, compared to senesced fronds, live fronds accumulated 25–49% higher arsenic concentrations in field conditions, suggesting the necessity of frond harvest before they senesce to maximize arsenic extraction. In the field trial in North Central Florida, surface soil arsenic was reduced from 190 to 140 mg kg$^{-1}$ by *P. vittata* after 2 years with planting density of 0.09 m$^2$ per fern (Kertulis-Tartar et al., 2006). Based on the projected removal capacity, 7–8 years is needed to remove arsenic in top 15 cm soil below the cleanup level for residential site (2.1 mg kg$^{-1}$) or commercial site (12 mg kg$^{-1}$) established by Florida Department of Environmental Protection. If reasonable remediation time can be achieved using *P. vittata*, in some cases, phytoextraction could be competitive in comparison with other conventional technologies for soil remediation (Table 4.2).

The applicability of phytoextraction using *P. vittata* has been further tested in 21 contaminated-soils from England with different soil types, arsenic contamination sources and concentrations as well as the coexistence of Cu, Cd, Zn and Pb (Shelmerdine et al., 2009). After three sequential growth and harvest of *P. vittata* over 9 months, 0.1–13% of total soil arsenic was removed from the soils where arsenic removal efficiency varied by up to 130-fold. Higher arsenic depletion was found under conditions with relatively low available P and low contamination of Pb, Cd, Cu and Zn. For instance, with comparable total arsenic being in soil-1 (367 mg kg$^{-1}$) and soil-4 (330 mg kg$^{-1}$), 1.7% of soil arsenic was removed after three harvests of fern fronds from soil-1 compared to only 0.26% from soil-4, which contained 3–24 times higher concentrations of Pb, Cd, Cu and Zn than soil-1. This confirmed that *P. vittata* performed better in soils with only arsenic contamination while exhibited apparent phytotoxicity and low arsenic uptake when grown in multiple metal/metalloid co-contaminated soils (Caille et al., 2004).

To predict the performance and success of arsenic phytoextraction with *P. vittata*, a combined solubility-uptake model was established by Shelmerdine et al. (2009). According to the model, arsenic phytoextraction by *P. vittata* is only suitable for marginally contaminated sites with relatively high soil pH (>6.0). For example, for a heavily-contaminated soil (pH 5.6, total As concentration 1250 mg kg$^{-1}$, and labile As fraction of 3%), assuming an annual harvestable yield of 3 t ha$^{-1}$, no significant decrease in total soil arsenic content over 30 years is expected. Therefore, arsenic phytoextraction using *P. vittata* is applicable in lightly contaminated soils and can be used as a soil-polishing tool in combination with other conventional remediating strategies.

### 4.2.2 Arsenic hyperaccumulation mechanisms

Based on a number of studies, the unique mechanisms of As hyperaccumulation have been gradually unraveled, which appears to involve efficient As mobilization in the rhizosphere, rapid root uptake and enhanced frond translocation of As by *P. vittata* (Fig. 4.1).
Table 4.2. Typical greenhouse and field studies of arsenic phytoextraction by *P. vittata*.

<table>
<thead>
<tr>
<th>Study site source of arsenic contamination</th>
<th>Initial soil arsenic [mg kg(^{-1})]</th>
<th>Growth conditions</th>
<th>Soil properties</th>
<th>Study time</th>
<th>Frond arsenic [mg kg(^{-1})dw]</th>
<th>BF</th>
<th>Frond biomass (dw)</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>USA, Chromated copper arsenate (CCA)</td>
<td>190</td>
<td>Field site with subtropical climate</td>
<td>loamy, siliceous; pH 7.4–7.6</td>
<td>2 years</td>
<td>3186–4575</td>
<td>29–45</td>
<td>1.3 t ha(^{-1}) yr(^{-1})</td>
<td>Kertulis-Tartar <em>et al</em>. (2006)</td>
</tr>
<tr>
<td>Australia, Arsenic-based pesticides</td>
<td>393–1903</td>
<td>Field site with subtropical climate</td>
<td>pH 4.8</td>
<td>5 months</td>
<td>775–2569</td>
<td>1–3</td>
<td>39.5 g fern(^{-1})</td>
<td>Niazi <em>et al</em>. (2009)</td>
</tr>
<tr>
<td>England, As-rich parent material, mining, industrial activities, agrochemical application and biosolid disposal</td>
<td>8.8–3580</td>
<td>Pot trial in a greenhouse with day/night temperatures of 25/16(^{\circ})C and a 16 h photoperiod</td>
<td>pH 4.0–8.2, Cu 12.4–3,560 mg kg(^{-1}), Cd 69 mg kg(^{-1}), Pb 19–19,400 mg kg(^{-1}), Zn 57–21,000 mg kg(^{-1})</td>
<td>9 months</td>
<td>9–3150</td>
<td>1.2–229</td>
<td>15.6 g fern(^{-1}) (2.5 t ha(^{-1}) yr(^{-1}) with planting density of 16 plants m(^{-2}))</td>
<td>Shelmerdine <em>et al</em>. (2009)</td>
</tr>
<tr>
<td>USA, Mining activities, arsenical insecticide, pesticide, CCA, and natural soil with high arsenic</td>
<td>23–640</td>
<td>Pot culture in a greenhouse</td>
<td>pH 6.7–7.9</td>
<td>16 months</td>
<td>110–6151</td>
<td>4.4–47.8</td>
<td>25–34 g fern(^{-1})</td>
<td>Gonzaga <em>et al</em>. (2008)</td>
</tr>
<tr>
<td>USA, CCA</td>
<td>98</td>
<td>Pot culture in a greenhouse</td>
<td>pH 7.5</td>
<td>5 months</td>
<td>6000</td>
<td>3.0–87.2</td>
<td>18 g fern(^{-1})</td>
<td>Tu <em>et al</em>. (2002)</td>
</tr>
</tbody>
</table>

*BF* is bioconcentration factor and is the ratio of total arsenic concentration in fronds to that in soil.
4.2.2.1 Arsenic mobilization via root exudates
Both root exudates and bacteria associated with *P. vittata* have been shown to help arsenic solubilization in the rhizosphere. In a greenhouse study, *P. vittata* excreted 2 times more dissolved organic carbon (DOC) than a control non-hyperaccumulating fern *Nephrolepis exaltata* (Tu et al., 2004b). As a result, the organic acids from root exudates of *P. vittata* induced 3–18 times higher mobilization of arsenic from insoluble arsenic minerals (AlAsO$_4$ and FeAsO$_4$) and an arsenic-contaminated soil as compared to *N. exaltata*. Besides root exudates, arsenic-resistant bacteria inhabiting *P. vittata* rhizosphere (*Pseudomonas* sp., *Comamonas* sp. and *Stenotrophomonas* sp.) have been shown to exhibit a remarkable ability to increase arsenic concentration in the uptake solution from <5 µg L$^{-1}$ to 5.04–7.37 mg L$^{-1}$ by solubilizing insoluble FeAsO$_4$ and AlAsO$_4$ (Ghosh et al., 2011). The production of pyochelin-type siderophores by arsenic-resistant bacteria has been suggested to play a role in arsenic solubilization.

To further investigate the rhizosphere characteristics of *P. vittata* relevant for its use in phytoextraction and the effects of root uptake on arsenic redistribution in soils, a sequential extraction procedure has been developed to fractionate arsenic into five operationally-defined fractions with decreasing availability (Fitz and Wenzel, 2002). It includes non-specifically bound, specifically bound, bound to amorphous hydrous Fe/Al oxides, bound to crystalline hydrous Fe/Al oxides, and residual fractions. In comparison with non-hyperaccumulator *N. exaltata*, *P. vittata* was more efficient to access arsenic from all five fractions, leading to greater removal of arsenic (39–64% vs. 5–39%) from rhizosphere soils than *N. exaltata* (Gonzaga et al., 2006). This observation seems related to 9% higher DOC and 0.5 unit higher pH in *P. vittata* rhizosphere, which tend to increase arsenic bioavailability by facilitating arsenic desorption from solid phase via anion competition. In addition, the majority of arsenic removed by *P. vittata* was from the major arsenic sink in soils (i.e., bound to Fe/Al hydrous oxides), accounting for 68% arsenic decrease in the rhizosphere. Fitz et al. (2003) reported that the difference in non-specifically bound arsenic (readily labile) between bulk and rhizosphere soils accounted for only 8.9% of total arsenic accumulated in *P. vittata*, again suggesting arsenic uptake was mainly from less available pools. Compared with
the bulk soils, a 2–2.8-fold greater Fe and DOC concentration was observed in *P. vittata* rhizosphere soil, indicating that arsenic mobilization was mainly resulted from Fe solubilization by organic compounds in *P. vittata* rhizosphere. In the experiment with *P. vittata* growing in an arsenic-contaminated soil (total arsenic 2270 mg kg\(^{-1}\)) for 41 d, arsenic depletion and limited resupply in the root rhizosphere was successfully illustrated by a 19.3% decrease in arsenic flux from solid phase to soil solution using diffusive gradients in thin films (Fitz et al., 2003). It seems DGT technique holds promise as an effective tool to monitor the bioavailability of target contaminant during and after phytoextraction.

### 4.2.2.2 Efficient root uptake system

In addition to being efficient in arsenic solubilization from the soil, *P. vittata* is also effective in arsenic uptake from soils. A comparison study between *P. vittata* and non-hyperaccumulating fern *N. exaltata* demonstrated that a more extensive root system and highly efficient root uptake contribute to the effective arsenic extraction by *P. vittata* (Gonzaga et al., 2007b). Compared to *N. exaltata*, a 2.4–3.8 times greater root system in terms of biomass was developed by *P. vittata* after 8 weeks of growth in both arsenic-contaminated and control soils. Furthermore, arsenic root uptake efficiency, which is defined as arsenic accumulation in fronds or roots per unit root biomass was 8–23 times greater in *P. vittata* than that of *N. exaltata*, indicating a more efficient root uptake system in *P. vittata*. The more extensive root system coupled with much higher root uptake efficiency of *P. vittata* accounts, at least partially, for the 29-fold higher arsenic depletion from soil by *P. vittata* in comparison to *N. exaltata* (2.51 vs. 0.09 mg arsenic per plant). This is consistent with the study of Poynton et al. (2004) who reported a significantly lower Michaelis constant \( K_m \) for arsenate [As(V)] influx into the roots of *P. vittata* than *N. exaltata* (1.1–6.8 µM vs. 9.9–19.9 µM), indicating a higher affinity of transporter protein for As(V) in *P. vittata*.

As the major arsenic species in aerobic soils, As(V) shares P transport system in higher plants including *P. vittata* (Zhao et al., 2009). As(V) uptake by *P. vittata* roots tend to be inhibited by increasing P in a competitive manner (Wang et al., 2002). Nevertheless, in the presence of As(V) at 1–10 mg L\(^{-1}\) in the uptake solution, P concentration in *P. vittata* was increased by 6.3- and 2.2-fold in the roots (from 0.91 to 5.76 mg g\(^{-1}\)) and fronds (from 2.33 to 5.19 mg g\(^{-1}\)), respectively (Luongo and Ma, 2005), which is in direct contrast to other tested ferns with an average of 40% reduction in P level. Therefore, it is suggested that the maintenance of sufficient P in fern tissues via efficient root uptake in the presence of high arsenic may constitute an essential detoxification mechanism in *P. vittata*.

On the other hand, it should be noted that due to the restriction of fern root extension, arsenic phytoextraction using *P. vittata* from soil profiles beyond root zones is much slower (Fitz et al., 2003). Therefore, it is necessary to evaluate fern rooting depth under field conditions, which will help to determine the effective depth of phytoextraction using *P. vittata*.

### 4.2.2.3 Efficient arsenic translocation to fronds

Not only does *P. vittata* have efficient root uptake system but also effective translocation mechanisms, making it the most striking attribute of an arsenic hyperaccumulator. Translocation factor (TF) has been used to characterize the effectiveness of plant arsenic translocation from the roots to fronds. As reported by Ma et al. (2001), arsenic TF in *P. vittata* reached 24 with frond arsenic being up to 7,234 mg kg\(^{-1}\) compared to root arsenic of 303 mg kg\(^{-1}\) after 20-week growth in an arsenic-contaminated soil (98 mg kg\(^{-1}\) As). Under variable soil arsenic concentrations from 6 to 1500 mg kg\(^{-1}\), arsenic accumulation in *P. vittata* fronds increased rapidly to 755–15,861 mg kg\(^{-1}\) after two weeks with frond BF being 10.6–126 (Ma et al., 2001). Following the first report unraveling the highly efficient translocation of arsenic in *P. vittata*, much research using hydroponics, greenhouse and field studies has consistently supported this constitutive trait of *P. vittata* (Natarajan et al., 2009; Singh and Ma, 2006; Su et al., 2008; Tu et al., 2002). For instance, by growing accessions of *P. vittata* from both contaminated and uncontaminated environments in arsenic-contaminated soils (0–500 mg kg\(^{-1}\)), efficient arsenic translocation resulted in an average of TF value of 6.8 with frond BF being 11.7 to 21.6 (Zhao et al., 2002).
The mechanisms responsible for the highly efficient arsenic translocation in *P. vittata* have been gradually unraveled. Arsenite [As(III)] is consistently present as the major species in *P. vittata* fronds with As(V) dominating the roots regardless arsenic species supplied (Kertulis *et al.*, 2005; Mathews *et al.*, 2010; Singh and Ma, 2006; Wang *et al.*, 2002). This suggests that efficient As(V) reduction and As(III) translocation as an important contributing factor to arsenic hyperaccumulation in *P. vittata*.

Su *et al.* (2008) reported that 93–98% of arsenic in the xylem sap of *P. vittata* was As(III) with either As(V) or As(III) being supplied, indicating a significantly higher mobility of As(III) than As(V) in xylem transport. Mathews *et al.* (2010) investigated the location of As(V) reduction by exposing *P. vittata* to 0.10 mM As(V) for 8 d. They found that As(III) concentrations increased significantly from 7% in the roots up to 68% and 71% in the rhizome sap and rhizome tissue. Along with upward translocation to the fronds, 86% As(III) in the frond sap and 90–100% As(III) in the pinnae were recorded, indicating a remarkable reduction capacity of the fronds as well as the rhizome. This is supported by Tu *et al.* (2004c) who has shown that upon exposing excised tissues of *P. vittata* to 0.67 mM As(V) for 2 d, 86% and 24% As(III) was present in the excised pinnate and roots. Taken together, it is conceivable that rhizomes and fronds are primarily responsible for As(V) reduction in *P. vittata* with increasingly more reduction along upward translocation. Besides, little arsenic efflux from the roots to external media and lack of a strong arsenic sequestration in roots both facilitate the highly efficient arsenic translocation in *P. vittata* (Su *et al.*, 2008; Zhao *et al.*, 2009).

Arsenic phytoextraction with *P. vittata* has been demonstrated to be affected by plant age, with young ferns exhibiting higher arsenic translocation than the older ones (Gonzaga *et al.*, 2007a; Santos *et al.*, 2008; Tu *et al.*, 2004a). For example, 36% more arsenic was accumulated in 2-month old *P. vittata* than 4–16 month old plants after 8-week growth in an arsenic-contaminated soil, which is likely associated with the higher metabolic activity in younger plants (Gonzaga *et al.*, 2007a). In addition, arsenic TF was reduced from 3.2 to 1.6 from old to young ferns, indicating decreased arsenic translocating ability of *P. vittata* with increasing plant age. Furthermore, frond biomass after 8 weeks growth increased by 39, 6.9, 2.0 and 1.1 times for *P. vittata* of increasing age of 2-, 4-, 10- and 16-month old, respectively. These results highlight the necessity to use young ferns in phytoextraction and harvest fronds before they senesce to minimize the remediating time and potential arsenic being leached from senesced fronds by rainwater in the field.

### 4.2.3 Potential improvement

#### 4.2.3.1 Phosphorous amendment

To achieve more effective phytoextraction, it is essential to employ proper agronomic techniques and plant management such as fertilizing and rhizosphere manipulation. One key strategy towards efficient phytoextraction is to enhance arsenic availability in soils. P amendment can be used in assisting arsenic uptake by *P. vittata* in the presence of toxic metals (e.g., Pb, Zn, and Cd) through increasing plant biomass and soil arsenic bioavailability via competitive anion exchange together with reduced metal toxicity by immobilization (Cao *et al.*, 2003; Fayiga and Ma, 2006; Tu and Ma, 2003). A maximum of P/As ratio of 1.2 in soil solution or 1.0 in the fronds has been suggested for an improved performance of *P. vittata* in arsenic phytoextraction by enhancing fern biomass and arsenic uptake (Tu and Ma, 2003). For instance, with a range of P added to the tested soil containing 400 mg As(V) kg⁻¹, a maximum of 26% soil arsenic extraction by *P. vittata* was recorded at water soluble P/As molar ratio of 2.3 after 20 weeks growth in a greenhouse experiment. Similarly, for another arsenic hyperaccumulator *Pityrogramma calomelanos* discovered in Thailand, a significant increment of frond arsenic content of 14 mg plant⁻¹ was reported after 8-week growth in the field containing 136–269 mg As kg⁻¹ with the addition of 100 mg P kg⁻¹ soil (Jankong *et al.*, 2007). In hydroponics, split P addition (134 + 66 μM) during *P. vittata* acclimation and after arsenic exposure (145 μg L⁻¹) has been shown to induce 1.5-folder higher efficiency in frond As accumulation in the younger ferns (45-d-old) compared to the older ones (90-d and 180-d-old) (Gonzaga *et al.*, 2008), suggesting the more efficient stimulation of P on As uptake in
the young ferns. However, in addition to increasing As availability in soils, P also competes with As for plant uptake so caution needs to be excised when adding P so plant arsenic uptake is not adversely affected.

4.2.3.2 Mycorrhizal symbiosis

Regarding rhizosphere manipulation via mycorrhizal symbiosis, which has a well-documented role in improving P uptake, it can increase arsenic accumulation and frond biomass in *P. vittata* (Agely et al., 2005; Fitz and Wenzel, 2002). *Glomus microaggregatum, G. mosseae, G. brohultii* and *G. geosporum* represent the most common arbuscular mycorrhizal fungi (AMF) in the rhizosphere of *P. vittata* (Wu et al., 2009). At the exposure of 3.8–75 mg L\(^{-1}\) As(V) in hydroponics, inoculation of an uncontaminated isolate of *G. mosseae* almost doubled arsenic influx into *P. vittata* roots relative to the control (25–105 vs. 14–55 mg As kg\(^{-1}\) dw h\(^{-1}\)) during a 20-min uptake experiment (Wu et al., 2009). In a greenhouse experiment with total soil arsenic of 100 mg kg\(^{-1}\), 2–5 times more arsenic accumulation in the fronds was found in *P. vittata* colonized by a community of AMF, with the enhancement being more evident with increasing soil P level as compared to those without colonization (Agely et al., 2005). Considering the fact that arsenic acts as a chemical analog of P and the well-documented role of AMF in enhancing P nutrition for the host plants, it is not surprising to find enhanced arsenic accumulation by mycorrhizal *P. vittata* with concurrent increase of plant biomass. However, the contribution of AMF to plant growth and arsenic translocation in *P. vittata* has been shown to largely depend on the species of AMF (Trotta et al., 2006), implying arsenic phytoextraction with *P. vittata* could be optimized by selected AMF symbiosis.

4.2.4 Potential environmental risks

4.2.4.1 Invasive risk

As a hardy and perennial fern species, *P. vittata* propagates quickly and is easy to maintain in a humid tropic/subtropic climate, which facilitates arsenic phytoextraction using *P. vittata*. However, from an ecological point of view, there is a concern regarding its invasive potential considering it is classified as a type-II invasive species in Florida. It is important to employ young ferns (e.g., 2 month-old), which exhibits higher capacity in arsenic accumulation than the old ferns (e.g., 16 month-old) (Gonzaga et al., 2007a). Such practice can reduce the potential of fern invasion with little spores being produced during phytoextraction. In addition, to reduce its invasion risk and overcome the geographical limitation of arsenic hyperaccumulators, exploration of indigenous species with potential for arsenic phytoextraction from local sites rich in arsenic, e.g., mining areas, may provide alternative options, which are well-adapted to the local environment (Antosiewicz et al., 2008; Mahmud et al., 2008; Visoottiviseth et al., 2002).

4.2.4.2 Disposal of arsenic-rich biomass

Safe management and economical disposal of arsenic-loaded biomass remains unsolved. Improper treatment of arsenic-rich plant materials may pose threats to ecosystem safety. For *P. vittata*, inorganic As(III) accounts for ~94% of the total arsenic in the fronds after 18 weeks growth in soil containing 50 mg As kg\(^{-1}\) (Tu et al., 2003). Furthermore, when the fronds were air-dried, the amount of leached arsenic substantially increased, with arsenic concentration in the leachate reaching 0.65 mg L\(^{-1}\) after 5 d of drying. The same holds true for arsenic hyperaccumulating fern *P. calomelanos* which grows naturally on arsenic-contaminated sites in southern Thailand, arsenic in the fronds is primarily present as inorganic species with 86–93% being water-extractable with the majority being As(III) (60–72%) (Francesconi et al., 2002). Taken together, these results suggest that arsenic concentrated in the hyperaccumulating ferns has high water solubility and hence the arsenic-rich biomass should be properly managed and kept away from water supply to minimize secondary contamination.

As for the possible disposable options, there is a paucity of data regarding the post-treatment technology and associated cost-benefit analysis. To effectively reduce plant biomass, incineration has been considered and more than 90% of plant biomass can be reduced by this means.
(Sas-Nowosielska et al., 2004). As a volatile metalloid, arsenic tends to be easily released by vaporization during thermal processing. Recent study has revealed that 24% of the accumulated arsenic in *P. vittata* could be emitted during incineration at 800°C with a 94% loss of biomass (Yan et al., 2008). The residual ash provides an enriched and recoverable ore with arsenic concentration being increased by 11-fold. While the development of a highly efficient capture system targeting arsenic-containing flue gas seems essential for further application of this technology.

In addition, water extraction of the harvested biomass may be feasible considering its high water solubility of arsenic. The resulting arsenic-rich extract could be recycled by an industry. Furthermore, decreased arsenic level in plant biomass could make the residue non-hazardous waste, with lower fee in landfill disposal. However, there is still large uncertainty for this option regarding arsenic extraction efficiency, residual arsenic concentration, and economic effectiveness. Alternatively, high-arsenic biomass could be disposed in marine system (Francesconi et al., 2002), which has a high detoxification capacity by transforming inorganic arsenic into essentially non-toxic organic forms by natural biochemical processes. While preliminary test and thorough assessment with regard to the ecological effect of this scenario is needed to minimize potential environmental risks. In addition, recent study has shown polymeric encapsulation technique could efficiently stabilize arsenic-bearing solid residual (e.g., iron-based sorbent) producing more than an order of magnitude lower concentration of leached arsenic than the conventional cement encapsulation (Shaw et al., 2008). However, the applicability of polymeric matrices to encapsulate arsenic-rich biomass needs both standard and landfill simulation leaching tests. In brief, there is a long way to go to establish a technically feasible, economically acceptable and environmentally safe disposal method.

4.3 PHYTOSTABILIZATION

4.3.1 Indigenous tolerant species with low TF

In sites contaminated with high-level of arsenic and other toxic metals (e.g., Cu, Zn, and Cd), phytostabilization is advantageous by immobilizing arsenic and reducing its exposure risks to ecological receptors (Table 4.3). High adaptability of spontaneous species with multiple metal/metalloid tolerance has been widely reported to serve as potential candidates for phytostabilization of the contaminated sites (Antosiewicz et al., 2008; Vamerali et al., 2009; Whiting et al., 2004). For instance, in a recent phytoremediation trial on a contaminated site with 0.15 m layer of gravelly soil over a 0.7 m layer of cinders containing As, Co, Cu, Pb and Zn, 100% survival of indigenous *Populus* and *Salix* were obtained after two-year of experiments with soil amelioration (e.g., mixing with imported soils, tillage, and fertilization) in spite of 16–92% lower tissue biomass than the control (Vamerali et al., 2009). The fact that trace metals were preferentially accumulated in the woody roots (84–89%) with marginal shoot translocation suggests the potential utilization of native *Populus* and *Salix* in phytostabilization of arsenic-contaminated sites.

4.3.2 Substrate improvement by legumes

To overcome the general poor nutritional status in degraded sites with arsenic contamination, legume plants with good N fixation capacity and strong root system serve as promising pioneering colonizer species for substrate improvement and revegetation. Furthermore, most species of legume generally exhibited low capability for shoot translocation of metals and hence pose low risks of exposure to other organisms in the food chain. For example, by growing white lupin in soils contaminated by the spill of acid pyrite sludge with co-occurrence of As and Cd, symbiosis with rhizobia were successfully established by lupin plants with root nodule formation, although the efficiency of N-fixation were reduced by 30–40% due to metal toxicity and inhospitable substrate. Simultaneously, soil acidity was mitigated with a marked increase of soil pH from
<table>
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<tr>
<th>Location and contaminant source</th>
<th>Contaminant concentration [mg kg $^{-1}$]</th>
<th>Soil amelioration</th>
<th>Plant species</th>
<th>Remediating time</th>
<th>Outcome</th>
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<tr>
<td>Torviscosa, Udine, Italy Cinder waste from pyrite ore roasting</td>
<td>As 886, Co 100, Cu 1735, Pb 493, Zn 2404</td>
<td>Metal-rich cinders covered with an unpolluted 0.15 m layer of soil. Both cinders and soil were mixed with sand (1:1 w/w) in pot experiment.</td>
<td><em>Populus and Salix</em></td>
<td>2 years</td>
<td>Coarse and fine roots provided a significant sink for contaminants with both TF and BCF &lt;1</td>
<td>Vamerali et al. (2009)</td>
</tr>
<tr>
<td>Rixton clay pits Kidsgrove Merton Bank, UK Industrial waste Aznalcóllar, Spain; Mine tailing spill</td>
<td>As 60–78, Cu ~508, Cd ~36</td>
<td>Green waste compost (GWC, 30% v/v) Biochar (20% v/v)</td>
<td><em>Miscanthus</em></td>
<td>8 months</td>
<td>Biomass yield increased by 2–4 times with GWC; Both TF and BCF &lt;1</td>
<td>Hartley et al. (2009)</td>
</tr>
<tr>
<td></td>
<td>As 49–339, Pb 73–607, Sb 4.5–37.7</td>
<td>Organic matter Ca-rich amendments</td>
<td><em>Quercus ilex subsp. Olea europaea Populus alba L. Phillyrea angustifolia L. Pistacia lentiscus L. Rosmarinus officinalis L., Retama sphaerocarpa L. Tamarix africana Poir.</em></td>
<td>since 1999</td>
<td>BCF &lt;0.03 for As, Pb, Sb in most species; BCF for Cd in <em>Populus alba</em> approaching 2</td>
<td>Domínguez et al. (2008)</td>
</tr>
<tr>
<td>Sugar Brook, UK Fazakerley, UK Kirby Moss, UK Merton Bank, UK Cromdale Grove, UK; Landfill, industrial waste, sewage sludge</td>
<td>As 4.9–5266, Zn 2.8–1300, Cu 10–880, Ni 10–109, Pb 45–1770</td>
<td>Soil turning to a depth of 30 cm followed by rotovation and weed-control treatment</td>
<td><em>Salix Populus hybrids Alnus Betula Larix</em></td>
<td>3 years</td>
<td>Low mobility and plant transfer for As, Pb, Cu, Ni with BCF &lt;1; Higher transfer of Cd and Zn with BCF up to 4–13 in <em>Salix</em></td>
<td>French et al. (2006)</td>
</tr>
<tr>
<td>Sanlúcar la Mayor, Spain Mine tailing spill</td>
<td>Available Cd 0.49 μM Available As 40 μM</td>
<td></td>
<td><em>Lupinus albus</em> (white lupin)</td>
<td>3 weeks for pot study; 1–6 months for field trial</td>
<td>Both TFs of Cd and As &lt; 1; Decreased soluble Cd and As in planted soil</td>
<td>Vázquez et al. (2006)</td>
</tr>
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</table>
3.9 to 6.5 after two weeks growth of white lupin in a pot experiment. Furthermore, soil soluble concentrations of As and Cd were reduced by 56% and 86% after five-month growth in field conditions with the roots being the major sink of As and Cd (Vazquez et al., 2006).

### 4.3.3 Fe oxides and biochar

In some cases, amendments such as Fe oxides are essential to mitigate arsenic toxicity and hence facilitate plant survival. For example, two highly-contaminated mine tailings in South Korea contained arsenic levels up to 6670 and 56,600 mg kg\(^{-1}\), resulting in high arsenic toxicity to plants. Reduction of \(~70–80\%\) available arsenic was achieved by adding amorphous Fe with the majority of arsenic bond to the stable Fe precipitates (Kim et al., 2003). In a field trial involving four arsenic-contaminated sites from UK containing 748 mg As kg\(^{-1}\), a mean reduction of 22–32% arsenic uptake by tested vegetables was recorded upon the addition of 0.2–0.5% Fe as ferrous sulfate in the top soils (Warren et al., 2003), confirming the high capacity of iron in soil arsenic immobilization. In a field trial of a grassland established on an abandoned chemical waste site, soil amendment with Fe(III) plus lime was the most efficient treatment with the labile arsenic being reduced by 92%. An 8% reduction in leached arsenic upon the application of lime was observed, resulting from the binding of arsenic with Ca\(^{2+}\) and resulting in reduced As mobility (Hartley et al., 2009a).

Biochar, as a promising soil amendment, has important environment implications for the bio-chemical behaviors of metals in soils. Regardless the type of feedstock and pyrolysis conditions, biochar with relatively high cation exchange capacity consistently shows adsorption capacity towards metal cations but little binding ability for arsenic species at typical environmental pH (Mohan et al., 2007). It should be noted that arsenic liability in soils increased to varying extent upon the application of biochar with higher arsenic concentrations in water-soluble and surface-adsorbed pool, probably due to raised soil pH by biochar (Hartley et al., 2009b; Namgay et al., 2010). For example, in a pot experiment with maize, the extractable arsenic in soil increased from 5.16 mg kg\(^{-1}\) in the control soil to 5.96 mg kg\(^{-1}\) in the biochar treatment (15 g biochar kg\(^{-1}\) soil) (Namgay et al., 2010). Similarly, in a 60-d field experiment, arsenic concentration in soil pore water increased by 30 fold with biochar application (soil:biochar = 2:1, v/v). These results highlight the potential risk of arsenic mobilization upon biochar application to arsenic-containing soils.

To effectively immobilize arsenic upon the application of biochar, iron oxides can be employed considering their high affinity to arsenic (Hartley and Lepp, 2008). Therefore, the combination of biochar and iron oxides can be a useful strategy to immobilize arsenic while improving soil fertility.

### 4.3.4 Phosphate

As a chemical analog of As(V), P is an effective competitor of As(V) for binding sites in soils. Due to the competitive anion exchange, increased bioavailability and plant uptake of arsenic has been well-documented upon P application (Cao and Ma, 2004; Hartley et al., 2009a). In a pot experiment with soil arsenic at 0, 15, and 30 mg kg\(^{-1}\), elevated arsenic accumulation in both rice grain and straw with lower grain yield was observed after P application of 50 mg kg\(^{-1}\) P (Hossain et al., 2009). For example, arsenic concentrations in rice grain and straw increased from 0.64 and 5.77 mg kg\(^{-1}\) to 0.71 and 6.21 mg kg\(^{-1}\) upon P addition at 30 mg kg\(^{-1}\) arsenic. As a result, the grain yield (g pot\(^{-1}\)) was reduced by 33–66%. Furthermore, arsenic concentration associated with Fe plaque was reduced by 20% on average, indicating higher arsenic solubility induced by P in rice rhizosphere. This highlights the risk that P fertilization may induce arsenic mobilization in soils and increase its uptake by food crops. Similar case has been reported during phytostabilization of a gold mine tailings with elevated arsenic (>1000 mg kg\(^{-1}\)) in New Zealand, where P has been employed to improve the hostile environment for better plant establishment (Mains et al., 2006). As a result, the leached arsenic increased proportionally to the amount of P applied. With P amendment at 3 g m\(^{-2}\), up to 0.5 and 0.9 mg L\(^{-1}\) arsenic was leached from the bare and
planted lysimeters during the five-month field trial. Furthermore, an interactive effect of plant species and type of P amendment on arsenic mobilization has been observed (Mains et al., 2006). For example, organic bioboostr amendment (dehydrated sewage sludge) induced higher leached arsenic than superphosphate fertilizer from barley (2.4 vs. 1.0 mg L\(^{-1}\)), but not for rye, corn or blue lupin. The fact that soil arsenic mobilization may occur upon plant establishment with P amendment highlights the necessity of pre-assessment to choose proper amendment strategy and plant candidate to minimize arsenic leaching and soil-plant transfer during phytostabilization.

### 4.3.5 Organic matter

As a complex mixture of varying components, organic matter (OM) has inconsistent effects on arsenic mobility, mainly due to the type of compost applied, degree of humification and pH variation (Juwarkar et al., 2008; Lagerkvist et al., 2008; Shiraliipour, 2002). For example, a significant increase in soil soluble arsenic from 5.7 mg L\(^{-1}\) to 7.1 mg L\(^{-1}\) was reported upon application of municipal solid waste and biosolids compost in a greenhouse experiment (soil pH 6.87) (Cao and Ma, 2004). Increased OM under neutral soil pH may improve microbial activity, and hence facilitate As(V) reduction to more mobile As(III). At the end of the experiment, As(III) concentration in soil solution was \(\sim 20–24\%\) in OM treatment compared to \(< 10\%\) in the control. In contrast, a reduction of water-soluble arsenic from 32 mg L\(^{-1}\) to 25 mg L\(^{-1}\) upon OM application was obtained in an arsenic-spiked soil with pH of 5.45 in the same experiment (Cao and Ma, 2004). This is probably due to arsenic adsorption onto OM in acidic conditions. Therefore, it is critical to monitor arsenic availability using OM to reduce the potential environmental risks of arsenic mobilization while improving substrate conditions.

### 4.3.6 Mycorrhiza

Proper inoculates of As-tolerant mycorrhiza can serve as a potential strategy to confer host-enhanced tolerance and facilitate phytostabilization by selectively accumulating P over As(V) (Sharples et al., 2000). For example, short-term As(V) and P uptake kinetics of mycorrhizal fungus *Hymenoscyphus ericae* from the roots of *Calluna vulgaris* were identical for the mine-impacted and control population. However, enhanced efflux of As(III) from mycelia preloaded with 0.1 mM As(V) for 1 h was observed in the resistant *H. ericae* from the mine population (14.4% h\(^{-1}\) vs. 6.6% h\(^{-1}\)), indicating the role of fungus as biofilter to maintain low plant arsenic via efficient As(III) efflux (Sharples et al., 2000). White clover (*Trifolium repens* Linn.) and ryegrass (*Lolium perenne* L.), which represent legumes and grasses that are commonly used in revegetation, exhibit high dependence on the mycorrhizal associations with *G. mosseae* for surviving in soils with severe arsenic contamination (Dong et al., 2008). Using a compartmented cultivation system, mycorrhizal inoculation of *G. mosseae* increased plant P concentration by 50–200%, with shoot arsenic being reduced by 9%–30% in the presence of 1 and 205 mg kg\(^{-1}\) arsenic. The data indicate the beneficial effect and potential use of AMF in phytostabilization via selectively accumulating P over As(V).

### 4.4 PHYTOEXCLUSION

For agriculture soils contaminated by arsenic, it is impractical to employ non-food crops for either phytoextraction or phytostabilization. Rice, which feeds half of the world’s population, is more efficient in arsenic accumulation than other cereals through efficient silicon transport pathway (Ma et al., 2008), particularly under flooding conditions. As a result, elevated arsenic has been widely reported in paddy rice with arsenic TF often approaching unity (Abedin et al., 2002; Stroud et al., 2010; Voegelin et al., 2010; Williams et al., 2007; 2009). To improve agriculture sustainability and food safety, a range of agronomic strategies and biotechnologies have been developed (Zhao et al., 2010), which may provide effective solutions to remediate arsenic-contaminated agriculture soils and reduce arsenic uptake by rice.
4.4.1 Water management

Because arsenic mobility and toxicity in paddy soils is largely controlled by soil redox potential, water management can be effective in reducing arsenic mobilization resulting from reductive dissolution of Fe hydroxides in anaerobic conditions. Compared with the conventional flooding cultivation, arsenic availability and uptake by rice can be remarkably decreased under aerobic conditions (Li et al., 2009; Xu et al., 2008). For example, compared with control, 4–16 times higher soluble arsenic concentrations were reported with As(III) accounting for 81–95% of the arsenic in flooding treatment. With efficient As(III) uptake system in rice, the enhanced arsenic solubility in flooded soils caused 10–15 fold higher grain arsenic concentrations than those under the aerobic treatment (Xu et al., 2008). Similarly, in a field trial with paired plots to compare raised bed and conventional flooding, a significantly higher redox potential was observed in the raised bed as compared with paddy conditions (0 vs. 120 mV in 0–15 cm soil), resulting in 3–6 fold lower arsenic concentrations in rice straw under aerobic condition than those in the flooded treatment (Duxbury et al., 2007). However, in the presence of Cd, aerobic treatment tends to mobilize soil Cd through the oxidation of CdS into CdSO4 (Kawasaki et al., 2009), indicating the potential negative effect of aerobic rice cultivation in soils contaminated with both As and Cd.

4.4.2 Silicon fertilization

Efficient As(III) uptake by rice is via Si transport system (Ma et al., 2008), suggesting enhanced Si availability can mitigate arsenic transfer in soil-rice system as well as improving grain yield (Ma and Yamaji, 2006). In a pot experiment, Si fertilization (20 g SiO2 kg−1 soil) decreased arsenic concentration in rice straw and grain by 78% and 16%, in spite of the 1.5–2 fold higher arsenic concentration in soil solution [with 78–100% As(III)] (Li et al., 2009). However, increased As availability did not translate to higher rice arsenic uptake probably due to enhanced competition of silicic acid for As(III) by plant uptake. Moreover, Si fertilization affected arsenic fraction by reducing inorganic arsenic level while enhancing arsenic methylation in both rice grain and husk. For example, 59% reduction of inorganic arsenic concentration and a concurrent 33% increase in dimethylarsinic acid concentration was found in rice grain with Si application (Li et al., 2009).

Therefore, Si fertilization is a promising strategy to reduce arsenic uptake and phytotoxicity in arsenic-contaminated soil-rice system.

4.4.3 Arsenic sequestration by Fe plaque

For paddy rice as well as other aquatic species, iron plaque formed on the root surface due to oxygenation of rhizosphere exhibits high capacity for retaining As(V), and therefore could effectively diminish arsenic influx into rice roots. In a pot culture experiment with rice growing under flooded condition, arsenic concentrations in the rhizosphere soil solutions were remarkably decreased, being 2.5-fold and 16-fold lower upon the amendment of amorphous iron at 0.1 and 0.5%, resulting from enhanced arsenic sequestration by Fe plaque. In comparison with control, arsenic content binding to Fe-plaque was 3–4 times higher in Fe treatments (0.51 vs. 1.49 and 2.41 mg pot−1). As a result, arsenic concentrations in rice shoots were significantly reduced, accounting for only 1/7–1/2 that in the control (Ultra et al., 2009).

Recent studies have further demonstrated significant variation in Fe plaque formation and arsenic sequestration among rice cultivars (Mei et al., 2009) and genotypes (Liu et al., 2006). Based on the pot experiment with 25 rice cultivars, significant negative correlation (p < 0.001) was observed between grain arsenic and root porosity and the rate of radial O2 loss (Mei et al., 2009). Rice cultivars with higher root porosity and rate of radial O2 loss exhibit higher oxidizing ability by releasing more O2 to the rhizosphere, and hence possess higher capacity in limiting arsenic influx into rice roots via effective arsenic fixation by Fe plaque. Taken together, Fe amendment and breeding provide potential strategy to minimize arsenic transport into rice by enhancing arsenic binding to the rhizosphere Fe plaque.
4.4.4 Pretreatment of arsenic-contaminated irrigating water

In South and Southeast Asia, extensive irrigation with arsenic-contaminated groundwater accounts predominantly for the elevated arsenic in paddy soils. Besides the above strategies to minimize arsenic contamination in soil-rice system, it is equally important to take proper measures to scavenge arsenic from irrigation water. Arsenic phytofiltration, an emerging technology, has been tested in both laboratory- and pilot-scale experiments using arsenic hyperaccumulators such as *P. vittata*. With initial As(V) concentration ranging from 20 to 200 µg L⁻¹, both *P. vittata* and another arsenic hyperaccumulator *P. cretica* exhibited high removal efficiency to reduce arsenic to below the drinking water limit of 10 µg L⁻¹ within 24 h (Huang et al., 2004). Furthermore, rapid arsenic removal has also been observed by repeatedly using the same batch of *P. vittata* after a short recovery (12–36 h), indicating the high sustainability of this technology. While in the presence of 1.6 mg L⁻¹ P with initial arsenic of 200 µg L⁻¹, a short term (60 min) arsenic influx into *P. cretica* roots was decreased by 88% as a result of the strong anion competition for root uptake (Huang et al., 2004). However, for the well-established *P. vittata* (6–7 month old), P treatment (1.2 and 2.4 mg L⁻¹) exerted little effect on arsenic depletion, with arsenic in the contaminated groundwater decreased from initial 130 µg L⁻¹ to less than 10 µg L⁻¹ in 2 d (Natarajan et al., 2009).

In a pilot scale phytofiltration system, arsenic concentration in the outflow was invariably less than 2 µg L⁻¹ through 84 d demonstration with initial arsenic between 6.6 and 14 µg L⁻¹ and flow rate between 255 and 1900 L d⁻¹. Besides, arsenic removal efficiency was unaffected by day length, light intensity and humidity (Elless et al., 2005), demonstrating the high reliability of this technology. Therefore, to reduce arsenic buildup in agriculture soils, phytofiltration is potentially useful for arsenic depletion from irrigation water in the regions where prevalent arsenic contamination occurs in water supply. However, phytofiltration with *P. vittata* is limited to subtropical and tropical regions and more suitable for small-scale water treatment considering the increased cost of fern maintenance in cold areas and huge space need for large-scale treatment system.

4.5 CONCLUSIONS

Arsenic phytoextraction with hyperaccumulators such as *P. vittata* has a potential for soil remediation with low to moderate arsenic contamination. Arsenic phytoextraction technologies based on greenhouse and field studies mainly include candidate plant screening, fertilizing, rhizosphere manipulation, growth timing, and harvest method, which provide essential basis for larger scale application of this technology. Proper measures need to be taken to diminish fern invasive risks particularly in eco-fragile regions and to achieve safe disposal of arsenic-rich biomass.

Arsenic phytostabilization with indigenous tolerant species with low translocation capacity is advantageous for heavily contaminated sites with high levels of arsenic and other toxic metals. Amendments such as iron oxides, phosphate, organic matter, N-fixation legume and mycorrhizal inoculation are important for plant survival in hostile environment and serve as core strategies to facilitate the success of phytostabilization.

To alleviate arsenic contamination in soil-rice system, a range of agronomic strategies and biotechnologies from water management and Si and Fe fertilization to pretreatment of irrigation water provide effective phytoexclusion measures to remediate arsenic-contaminated agriculture soils and reduce arsenic uptake by rice.

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