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Effects of compost and phosphate amendments on arsenic mobility in soils and arsenic uptake by the hyperaccumulator, *Pteris vittata* L.

Xinde Cao[a], Lena Q. Ma[a,*], Aziz Shiralipour[b]

[a]*Department of Soil and Water Science, University of Florida, Gainesville, FL 32611, USA*
[b]*Center for Natural Resources, University of Florida, Gainesville, FL 32611, USA*

"Capsule": *Phosphate amendment increases the effectiveness of Chinese brake fern to remediate As-contaminated soils, by increasing As uptake and decreasing As leaching.*

Abstract

Chinese brake fern (*Pteris vittata* L.), an arsenic (As) hyperaccumulator, has shown the potential to remediate As-contaminated soils. This study investigated the effects of soil amendments on the leachability of As from soils and As uptake by Chinese brake fern. The ferns were grown for 12 weeks in a chromated–copper–arsenate (CCA) contaminated soil or in As spiked contaminated (ASC) soil. Soils were treated with phosphate rock, municipal solid waste, or biosolid compost. Phosphate amendments significantly enhanced plant As uptake from the two tested soils with frond As concentrations increasing up to 265% relative to the control. After 12 weeks, plants grown in phosphate-amended soil removed >8% of soil As. Replacement of As by P from the soil binding sites was responsible for the enhanced mobility of As and subsequent increased plant uptake. Compost additions facilitated As uptake from the CCA soil, but decreased As uptake from the ASC soil. Elevated As uptake in the compost-treated CCA soil was related to the increase of soil water-soluble As and As(V) transformation into As(III). Reduced As uptake in the ASC soil may be attributed to As adsorption to the compost. Chinese brake fern took up As mainly from the iron-bound fraction in the CCA soil and from the water-soluble/exchangeable As in the ASC soil. Without ferns for As adsorption, compost and phosphate amendments increased As leaching from the CCA soil, but had decreased leaching with ferns when compared to the control. For the ASC soil, treatments reduced As leaching regardless of fern presence. This study suggest that growing Chinese brake fern in conjunction with phosphate amendments increases the effectiveness of remediating As-contaminated soils, by increasing As uptake and decreasing As leaching.
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Keywords: Phosphate; Biosolid compost; Municipal solid waste; Arsenic mobility; Arsenic uptake; Chinese brake fern; Remediation

1. Introduction

Arsenic (As) has long identified as a carcinogen. Elevated concentrations in the ecosystem is of great concern for public health and the environment (Hingston et al., 2001). Arsenic contamination in soils results from various human activities including milling, combustion, wood preservation, and pesticide application (Carbonell-Barrachina et al., 1998). There are tens of thousands of arsenic contaminated sites worldwide, with the

arsenic concentration as high as 26.5g kg^{-1} soil (Hingston et al., 2001).

Inorganic waterborne preservatives, such as chromated copper arsenate (CCA), are effective in protecting wood from bacterial, fungal, and insect attacks (Hingston et al., 2001). However, broad use of CCA in treating wood has increased concerns about possible environmental contamination from the leaching losses of wood preservatives. As arsenic accumulates in soils, there may be an increase in health risks resulting from As leaching into ground and surface water and subsequent consumption by animal and human populations. A recent report by the National Research Council concluded that the former arsenic standard of 50 μg l^{-1}

* Corresponding author. Tel.: +1-352-392-1951 ext. 208; fax: +1-352-392-3902.
E-mail address: lqma@ufl.edu (L.Q. Ma).

in drinking water does not achieve USEPA's (United States, Environmental Protection Agency) goal of protecting the public health (Christen, 1999). In response to this research, USEPA has lowered the drinking water standard from 50 to 10 μg l^{-1}, effective nationally by 2006.

Arsenic-contaminated soil is one of the major sources of arsenic in drinking water (Hingston et al., 2001). Therefore, to protect animal and human health, remediation of the contaminated sites has become an urgent issue. Phytoremediation, a plant-based green technology, has been successfully used to remove a number of metals from contaminated soils (Lombi et al., 2001). Chinese brake fern (*Pteris vittata* L.) has been recently discovered to be an arsenic hyper-accumulator (Ma et al., 2001). The plant has accu-mulated up to 2.3% As of dry plant weight from contaminated soils. Phytoremediation is feasible since 90% of the arsenic absobed was in the above-ground biomass, and could be removed by frond harvest (Tu et al., 2002).

A key to effective phytoremediation, especially phy-toextraction, is to enhance pollutant phyto-availability and to sustain adequate pollutant concentrations in the soil solution for plant uptake (Lombi et al., 2001). Var-ious soil amendments have been used to aid plant uptake and accumulation of contaminants (Heeraman et al., 2001; Peryea, 1998; Zhou and Wong, 2001). Incorporation of carbon-rich composts into soils has been shown to increase metal solubility through forma-tion of soluble metal–organic complexes (Zhou and Wong, 2001). Thanabalasingam and Pickering (1986) found that As adsorption by humic materials depends on soil pH with a maximum sorption at pH of 5.5. But arsenic can also be transformed to the reduced As(III) species or organic forms through biomethylation by microbes over a wide range of pHs (Turpeinen et al., 1999). Reduced compounds have higher mobility than As(V) forms, possibly enhancing their plant availability. Phosphate addition to arsenic-contaminated soils has been shown to enhance arsenic release from the soil through competitive anion exchange (Peryea and Kam-mereck, 1997). Peryea (1998) reported that phosphate fertilizer increased soil As availability to apple trees grown in As- contaminated soils.

With the appeal of increased arsenic availability from the application of compost and phosphte for phytor-emediation, there is also the concern for ground water quality. As arsenic availability is increased by soil amendments, it is hoped that the Chinese brake fern will proportionally absorb the available arsenic and minimize arsenic leaching. However, whether the increased plant will balance leaching from the top soil remains unclear.

The overall objective of this study was to determine whether soil amendments could increase arsenic uptake by the Chinese brake fern while avoiding leaching los-ses. The special tasks were: (1) to evaluate the effects of composts and phosphate rock applications on arsenic uptake by Chinese brake fern growing in arsenic con-taminated soils; (2) to determine the effects of composts and phosphate rock on arsenic leachability in arsenic contaminated soils; and (3) to identify possible mechanisms responsible for As mobility in soil after compost and phosphate treatments.

2. Materials and methods

2.1. Soil, compost and phosphate rock samples

The As-contaminated soil was collected from the sur-face (0–20 cm) at an abandoned CCA wood preserva-tion site, located in north central Florida. A non-contaminated soil was taken from the surface (0–20 cm) on the University of Florida campus. After air-drying, the non-contaminated soil was spiked with a Na$_2$HAsO$_4$ solution and incubated for one week to produce an As spiked contaminated soil (ASC) that contained 125 mg As kg^{-1} dry soil. Two composts used in this study were municipal solid wastes (MSW) and Biosolids (BS) which were supplied by the Sumter County Composting Facility and the Palm Beach Authority Composting Facility in Florida, respectively. Phosphate rock [PR, Ca$_{10}$(PO$_4$)$_6$F$_2$ (CaCO$_3$)$_x$, < 60 μm] was obtained from the PCS Phosphate company (White Springs, FL). Phosphate rock was chosen as the P source for the treatment since it would provide a long-term supply of P with a low risk of P leaching due to its low solubility. Selected properties of soils, composts, and phosphate rock are provided in Table 1.

2.2. Soil treatments

Dried MSW and BS composts were sieved to a < 2-mm diameter and were mixed with the CCA and ASC soils at a ratio of 50g kg^{-1} soil. PR was fully with the soils at a ratio of 15g kg^{-1} soil. In addition, Osmocote$^{®}$ extended time release fertilizer (Scotts-Sierra Horti-cultural Products Co., Marysville, OH) was mixed in as a base fertilizer at 1g kg^{-1} soil (Tu and Ma, 2002). 1.5 kg of soil containing different amendments was placed into each pot (2.5 l, d = 15 cm). The three replicates of each amendment were done in a completely randomized factorial design. The soils without PR or compost amendments were used as the control.

2.3. Greenhouse experiment

Fern seedlings were propagated in the lab and trans-ferred, one to a pot, at the 5–6 frond stage (Tu and Ma, 2002). Soil moisture content was maintained at field

Table 1
Selected physicochemical properties of the soil, composts, and phosphate rock used in this study

	pH	CEC[a] (cmol kg^{-1})	OC[b] (%)	Sand (%)	Silt (%)	Clay (%)	Total As (mg kg^{-1})	WS–As[c] (mg kg^{-1})
CCA soil[d]	6.87	7.80	0.91	89.6	7.90	2.50	135	5.72
ASC soil	5.45	6.21	1.85	87.2	9.49	3.21	126	27.8
MSW	6.71	ND[e]	51.2	ND	ND	ND	5.01	0.05
BS	7.18	ND	69.3	ND	ND	ND	7.57	0.07
PR	7.10	ND	<0.01	ND	ND	ND	0.23	<0.01

[a] Cation exchange capacity.
[b] Organic carbon.
[c] Water–soluble As, extracted with deionized water for 1 h at a ratio of liquid/soil = 10.
[d] CCA, chromated–copper–arsenate; ASC, As spiked contaminated; MSW, municipal solid waste; BS, biosolid; PR, phosphate rock.
[e] Not determined.

capacity by periodically weighing the pots and adding water to compensate for any weight loss. The experiment was conducted in a greenhouse at 23–25 °C with an average photosynthetically active radiation at 825 μmol m^{-2} s^{-1}. Pots were randomized on the greenhouse bench and their positions were changed every 4 weeks to minimize variations in the micro environments.

Soil samples were collected at 0, 2, 5, and 12 weeks by using a small core made from 10-ml polypropylene syringe. The collected soils were air-dried and passed through a 2-mm sieve. Ferns were harvested at the end of the experiment (12 weeks). After being washed thoroughly with tap water and then with deionized water, the ferns were separated into above ground (fronds) and below ground (roots). Biomass was measured on a dry-weight basis after being dried at 65 °C for 96 h. The dry plant samples were ground into fine powder by using a tissue mill before acid digestion.

2.4. Speciation of soluble As in soil solution

Speciation of As in the soil solution was performed at the time of plant harvest (12 weeks). Approximately 150 g of each soil at field capacity was centrifuged in a Teflon cup at 27 500 g and 25 °C for 20 min to extract the soil solutions (Dahlgren et al., 1997). These solutions were then filtered through a 0.20-μm acetate membrane for total As, As(V), and As(III) analysis. Triplicates were run for each treatment.

2.5. Sequential extraction of As in soils

Soil samples were extracted using the sequential extraction procedure of arsenic (Onken and Adriano, 1997). The procedure separated As into five operationally defined fractions: water-soluble and exchangeable As (WE–As), aluminum bound As (Al–As), iron bound As (Fe–As), calcium bound As (Ca–As), and residue As (RS–As). Extractants used in the five fractions were 1 mol l^{-1} NH$_4$Cl, 0.5 mol l^{-1} NH$_4$F, 0.1 mol l^{-1} NaOH, 0.25 mol l^{-1} H$_2$SO$_4$, and 1:1 HNO$_3$/H$_2$O$_2$, respectively.

One gram of soil was sequentially extracted with 20 ml of each extraction solution. Between each extraction the soil was washed twice with 25 ml of saturated NaCl. Each treatment was run in triplicate. The arsenic recovery was determined by summing the As present in all extracts and comparing that to the total As. The results showed satisfactory recoveries of 91–121%. The accuracy of the sequential extraction was evaluated by analyzing Standard Reference Material of 2710 (NIST, Gaithersburg, MD)

2.6. Column leaching experiments

At the end of the greenhouse experiment (week 12), soil samples were collected from all treatments, both with and without ferns. After being air-dried, the soils were packed into 60 ml columns (d = 2.5 cm), and the soil bulk density was determined to be 1.17–1.32 g ml^{-1}. Columns were run in triplicate for each treatment. Deionized water was introduced according to the upward filling/downward leaching procedure (Peryea and Kammereck, 1997). The leachates were then collected for both dissolved organic carbon (DOC) and As analyses.

2.7. Chemical analysis

Soil pH was determined using a 1:1 ratio of soil to deionized water after 24 h of equilibration. DOC was determined by using total organic carbon analyzer (TOC-5050A, Shimadzu). Plants and soils were digested using HNO$_3$/H$_2$O$_2$ Hot Block Digestion System (USEPA Method 3050). Arsenic was determined using a graphite furnace atomic absorption spectrometer (GFAAS, Perkin-Elmer SIMMA 6000, Norwalk, CT). Elemental analysis followed an EPA approved QA/QC plan which included a blank, duplicate, and spiked sample in addition to a SRM per 20 samples. Quality control samples included Standard Reference Materials 1547 (Peach Leaves) and 2710 Montana Soil (US NIST, MD). Phosphorus analysis was carried out using a

modified molybdenum blue method (Carvalho et al., 1998). This method eliminated the interference of arsenate with P determination by reducing arsenate to arsenite with L-cysteine. As(V) and As(III) determinations were performed using the method of Carvalho et al. (1998), coupled with the method of Johnson and Pilson (1972). The molybdenum blue method was performed by both with and without the cysteine reduction of As(V). The difference between these two results yields the As(V) concentration. As(III) was found by subtracting As(V) concentration from the total As result obtained by GFAAS.

2.8. Data analysis

All results are expressed as an average of three replicates, and treatment effects were determined by analysis of variance according to the general linear model procedure of the statistical analysis system (SAS Institute Inc.). Differences among the treatment means were separated by least significant difference (LSD). Significance was tested at the 0.01 and 0.05 probability levels.

3. Results and discussion

3.1. Properties of the soil and amendment materials

The two soils used showed a significant difference of ~ 1.5 pH units, with the CCA soil being neutral (pH ~ 7.0) and the ASC soil being acidic (pH ~ 5.5) (Table 1). Total As concentrations in the test soils were significantly higher (125–136 mg kg^{-1}) than the average background of 0.4 mg kg^{-1} for Florida soils (Chen et al., 1999). Although the two soils contained a similar amount of total As, water-soluble As (WS–As) in the ASC soil was five times greater than that in the CCA soil (Table 1). The high level of water soluble As in the ASC soil was attributed to the spiking of As into the soil, in which As was predominantly associated with the labile exchangeable and aluminum oxide fractions ($\sim 80\%$ of total As). On the other hand, in the CCA, soil As was mainly present in the stable Fe–As and Ca–As fractions ($\sim 80\%$ of the total As) (Fig. 1). Phosphate rock and the two composts had neutral pH levels (~ 7). The two composts contained $> 50\%$ organic carbon with more organic carbon in the biosolids. Arsenic concentrations in both composts and PR were negligible, compared with that in the CCA and ASC soils (Table 1).

3.2. Effects of soil amendments on soil pH, DOC, and water soluble As

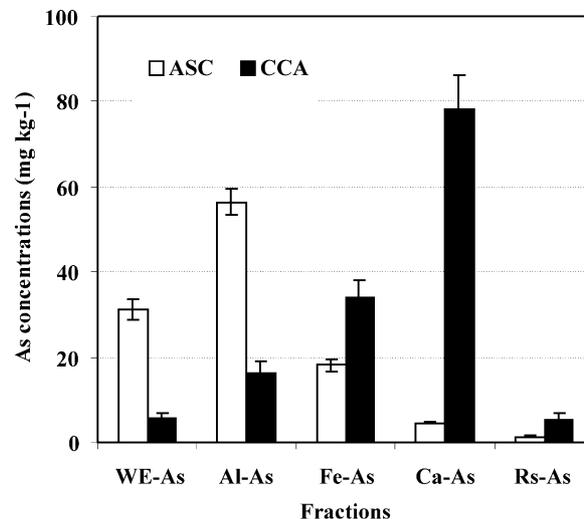Addition of MSW, BS composts or PR had no significant effects on the soil pH ($P < 0.01$) (Fig. 2a and b).

Fig. 1. Arsenic distribution in the CCA and ASC soils. WE–As, water-soluble and exchangeable; Al–As, As associated with Al; Fe–As, As associated with Fe; Ca–As, As associated with Ca; Rs–As, residual As.

No significant change in pH of the CCA soil was probably due to the similar pH of the soil and each of the amendments. The application of the neutral pH amendments increased pH of the acidic ASC soil at first. However, there was no significant difference of pH between the control and amended soils after the 2-week equilibration (Fig. 2a and b). As expected, amending the soil with PR had no effect on DOC in both the CCA and ASC soils. However, both composts increased the DOC in both soils with more pronounced increase observed in the BS treatment (Fig. 2c and d). In addition, the CCA soil contained more DOC than the ASC soil although the original OC in the ASC soil was double that found in the CCA soil (Table 1). This is possibly due to high pH in the CCA soil tending to dissolve more organic matter from the composts (Zhou and Wong, 2001). With time, DOC decreased due to the mineralization, adsorption, and volatilization of the organic matter in the soils.

The water-soluble arsenic (WS–As) was significantly elevated in the CCA soil after soil amendments ($P < 0.05$) (Fig. 3a). Phosphate and arsenate exhibit similar physicochemical behavior and compete directly for sorption sites on soil particles (Davenport and Peryea, 1991). Addition of phosphate to the As-contaminated soils induced arsenate replacement through competitive anion exchange (Peryea, 1998), thereby enhancing As release into the soil solutions. Also, the increased DOC may compete for anion adsorption sites. The increased organic matter coupled with neutral pH may favor microbial activity which may lower the soil redox potential (Turpeinen et al., 1999). This situation is favorable for the reduction of As(V) to As(III), and a subsequent increase in As mobility (Turpeinen et al., 1999). At the end of this experiment (12 weeks), the

CCA–compost soils had up to 24.2% of soluble As in the soil solution present as As(III), as compared to <10% in the control and the phosphate-amended soils (Table 2). Pongratz (1998) reported that the reduction of As(V) to As(III) occurred as a biotic process in anaerobic environments. Organic material from composts could have provided favorable conditions for As reduction. Also, it could have provided an energy source for the micro-organisms which are potentially involved in arsenic transformation (Balasoiu et al., 2001).

Similarly, phosphate amendment significantly increased WS–As ($P < 0.05$) in the ASC soil at 12 weeks although WS–As was less than in the control within first 4 weeks (Fig. 3b). However, compost treatments reduced the WS-As compared with the control (Fig. 3b). It is possible because As may be adsorbed on the organic matter of the composts in acidic ASC soil (pH = 5.45). It has been reported that oxyanion adsorption was enhanced in the presence of organic matter as pH decreases (Sposito, 1984). Xu et al. (1991) reported

that acidification and organic matter addition reduced arsenic mobility with arsenic adsorption reaching a maximum at around pH 5 for As (V). No net transformation of As from As (V) to As (III) occurred in the compost-treated ASC soil (Table 2). It is possible that such a high amount of water-soluble arsenic (~30 mg kg^{-1}) in the ASC soil could have inhibited the microbial metabolism (Turpeinen et al., 1999), showing less possibility of As(V) transformation into more available As(III). Therefore, the reduction of As mobility in the ASC soil may be attributed to arsenic adsorption.

3.3. Soil As redistribution

Arsenic in the CCA soil was mainly associated with Ca (56.0%), while Al–As (50.5%) was the predominant form of As in the ASC soil (Table 3). At planting (0 week), soil amendments decreased non-labile As fractions of Fe–As and Ca–As, but increased water-soluble and exchangeable As (WE–As) and Al–As in the CCA soil. For the ASC soil, however, treatments decreased
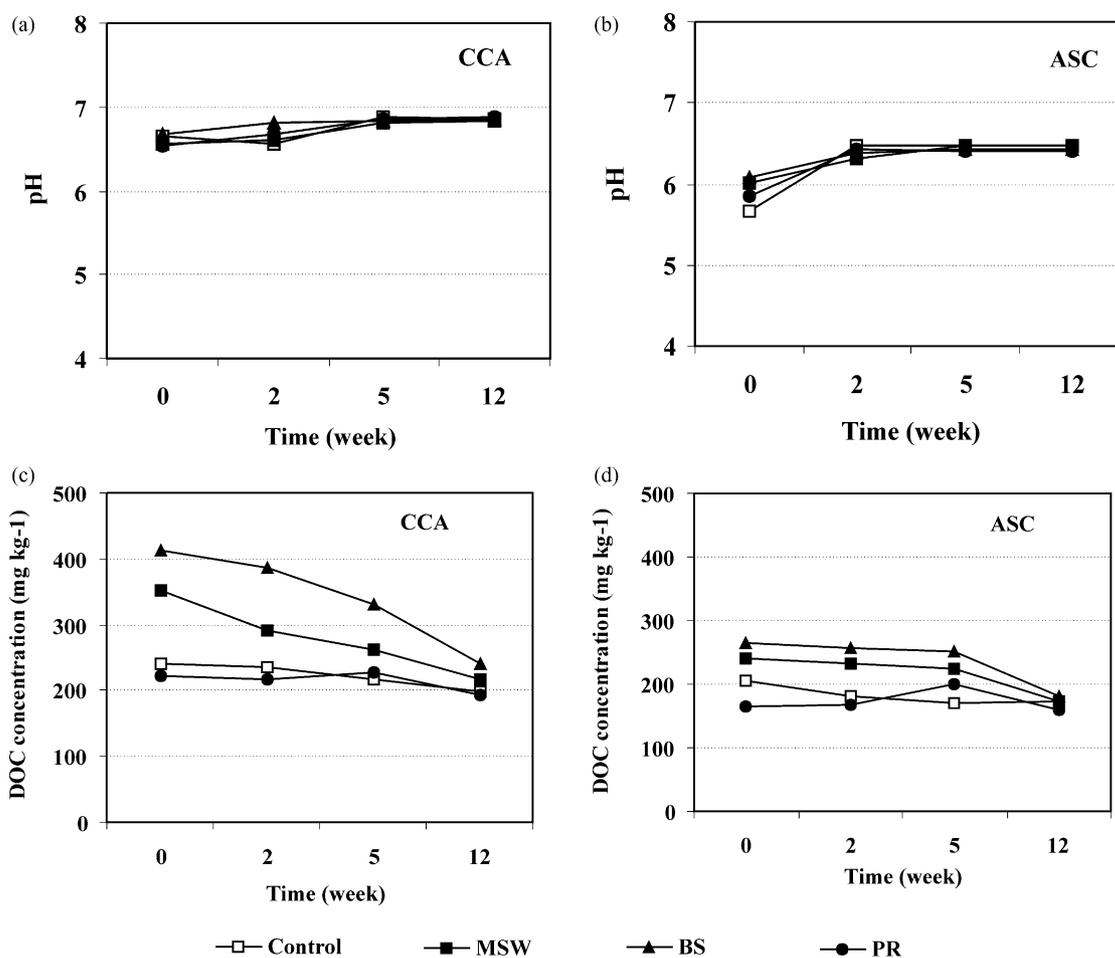

Fig. 2. Soil pH (a and b) and DOC (c and d) in the CCA (a, c) soil and ASC (b, d) soil samples after compost and phosphate treatments as a function of time. CCA, chromated–copper–arsenate, ASC, As spiked contaminated, DOC, dissolved organic carbon; MSW, municipal solid waste; BS, biosolid; PR, phosphate rock.

WE-As, while Fe–As Ca–As and RS–As were significantly elevated.

When the ferns were harvested (week 12), the CCA soil As concentrations in each fraction of the control and treated soils had decreased with time, especially in
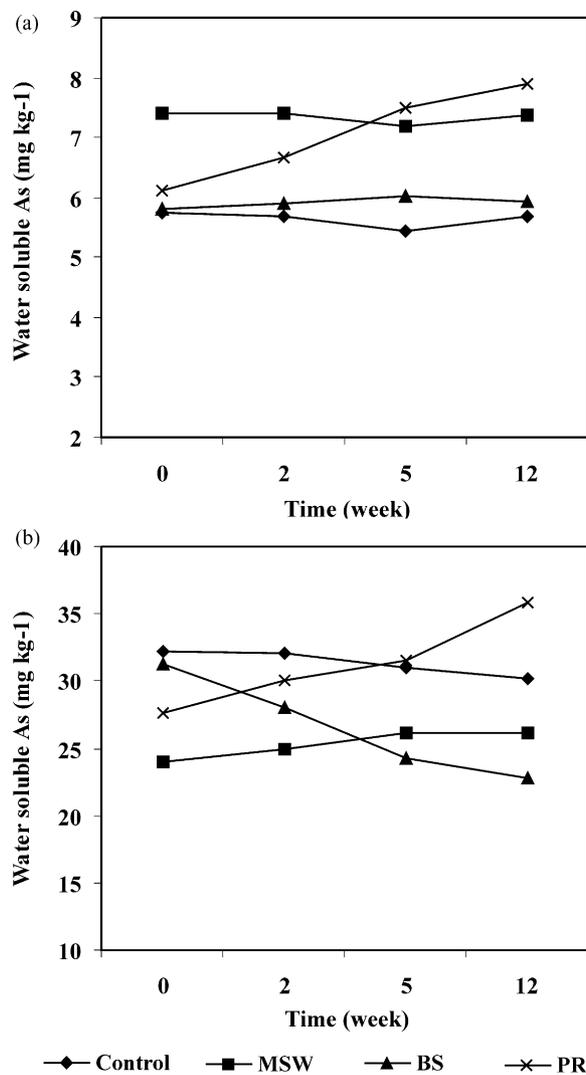

Fig. 3. Water-soluble As in the CCA soil (a) and ASC soil (b) after compost and phosphate treatments as a function of time. CCA, chromated–copper–arsenate, ASC, As spiked contaminated, DOC, dissolved organic carbon; MSW, municipal solid waste; BS, biosolid; PR, phosphate rock.

Al–As and Fe–As. These two fractions had As decreases of 11.7 to 34% and 8 to 40%, respectively, when compared with the concentrations at planting (Table 3). This probably indicates that As uptake by the fern originated mainly from these two fractions, with the greatest contribution coming from the Fe–As. It can be assumed that the displacement of As by P readily occurred on the surface of the Fe particles and that Fe was readily reduced in the anaerobic soil condition induced by compost addition, thus releasing As for the fern uptake. The ASC soil showed a significant decrease ($P < 0.05$) in As concentration in the first two fractions of WE–As and Al–As with time in both the control and the treated soils. As expected, the greatest reduction of WE–As occurred in the compost treatments by up to 20.2% after 12 weeks. However, Ca–As and RS–As increased with time in the ASC soil. This may be partially explained by As aging immobilization in the As-spiked ASC soil. Nevertheless, this large reduction of WE–As may imply that the fern plant took up As mainly from the WE–As fraction of the ASC soil. It should be pointed out that the sequential extraction procedure was only used here to represent successively more recalcitrant forms of arsenic since these fractions do not necessarily represent specific discrete forms.

3.4. Arsenic uptake and accumulation in the fern plants

Tu and Ma (2002) reported that Chinese brake ferns are highly tolerant of arsenic and can survive in a soil containing up to 500 mg As kg^{-1}, which was spiked in the soil as Na_2HAsO_4. For this study, the ferns grew well in the ASC soil with 125 mg As kg^{-1} soil. Fronds accumulated up to 5600 mg As kg^{-1} dry plant weight after 12 weeks (Fig. 4), further demonstrating the As-hyperaccumulating capability of Chinese brake fern reported by Ma et al. (2001).

The PR treatment enhanced As uptake by the Chinese brake fern, with frond concentrations increasing by 256% and 15.4% in the CCA and ASC soils, respectively (Fig. 4), when compared to the control. Otte et al. (1990) reported that *U. dioica* grown in a soil containing 75 mg As kg^{-1} soil took up more arsenic in the presence of P, most likely via competitive desorption where both

Table 2
Speciation of soluble arsenic in the soil solutions after the fern plants were harvested at 12 weeks

	Soluble As (mg l^{-1}) in the CCA soil solution				Soluble As (mg l^{-1}) in the ASC soil solution			
	Total As	As (V)	As (III)	As (III)%	Total As	As (V)	As (III)	As (III)%
Control	5.69±0.11b[a]	5.13±0.16b	0.56±0.13b	9.68b	30.2±1.17b	28.6±2.31a	1.61±0.27a	5.33a
MSW[b]	7.28±0.33a	5.82±0.28b	1.46±0.27a	20.0a	26.2±4.70c	24.7±2.31b	1.52±0.72a	5.80a
BS	6.94±0.32a	5.26±0.21b	1.68±0.31a	24.2a	22.8±2.69c	21.3±1.74b	1.51±0.21a	6.62a
PR	7.40±0.48a	6.72±0.62a	0.68±0.17b	8.61b	35.8±2.92a	23.8±2.17b	1.95±0.45a	5.45a

[a] Mean±standard deviation ($n = 3$), values ending in the same letter within each column are not significantly different ($P < 0.05$).
[b] CCA, chromated–copper–arsenate; ASC, artificially As contaminated; MSW, municipal solid waste; BS, biosolid; PR, phosphate rock.

Table 3
Arsenic concentrations (mg kg^{-1}soil) in each soil fraction at planting and harvest

	CCA Soil					ASC Soil				
	WE–As[a]	Al–As	Fe–As	Ca–As	RS–As	WE–As	Al–As	Fe–As	Ca–As	RS–As
At planting (week 0)										
Control	5.76b[b]	16.2ab	34.1a	80.2a	5.28a	31.2a	56.4b	18.1c	4.62d	1.37c
MSW[c]	7.40a	18.2a	33.7a	71.4bc	6.20a	27.0bc	59.4a	16.9cd	4.87d	1.42c
BS	6.33ab	18.8a	31.0b	75.0b	5.28a	31.2a	56.8b	18.2c	5.04cd	1.65c
PR	6.22ab	19.1a	31.0b	78.1ab	5.32a	28.6b	57.8ab	18.0c	6.77ab	1.86c
At harvest (week 12)										
Control	5.30b	12.0c	27.8c	78.8ab	5.38a	27.5bc	40.3c	18.5c	5.72c	2.17b
MSW	6.16ab	12.3c	24.9d	73.2bc	5.16a	24.5c	38.9c	20.7b	6.32b	2.56b
BS	5.68b	15.6b	19.1e	76.6b	5.58a	24.9c	41.9c	22.3a	6.20b	3.81a
PR	6.56ab	14.7bc	18.6e	75.3b	5.30a	30.9a	37.8c	19.0bc	7.17a	3.47a

[c] CCA, chromated–copper–arsenate; ASC, artificially As contaminated; MSW, municipal solid waste; BS, biosolid; PR, phosphate rock.

[a] WE–As, water-soluble and exchangeable; Al–As, aluminum bound As; Fe–As, iron bound As; Ca–As, calcium bound As; RS–As, residue As.

[b] Mean ($n = 3$), values ending in the same letter within each column are not significantly different ($P < 0.05$).
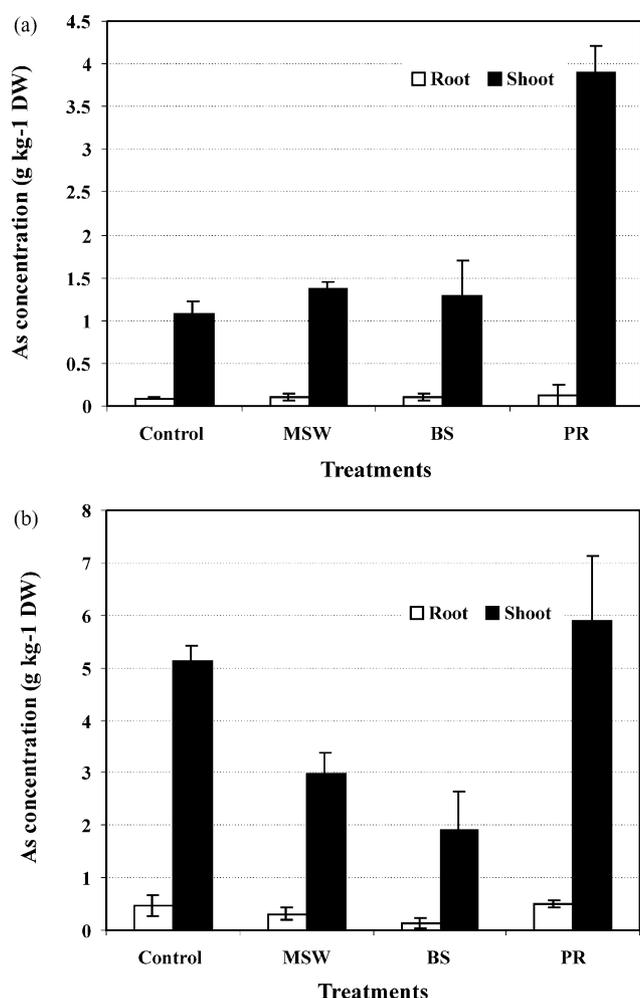

Fig. 4. Root and frond arsenic concentrations in the Chinese brake fern grown in the CCA soil (a) and ASC soil (b) at 12 weeks. CCA, chromated–copper–arsenate, ASC, As spiked contaminated, MSW, municipal solid waste; BS, biosolid; PR, phosphate rock.

elements compete for the same adsorption sites in the soil and root surfaces, which is expected due to their chemical similarity. Peryea (1998) demonstrated that the application of phosphorus fertilizer to arsenic-contaminated soils resulted in the displacement of about 77% of the total arsenic in the soil. Previous studies showed that any effect P has on As uptake by plants is liked directly with the growth media (Jacobs and Keeney, 1970; Meharg et al., 1994; Woolson et al., 1973). In a hydroponic system, 15 mg l^{-1} can reduce As uptake by 75% in both tolerant and non-tolerant plant genotypes of *Holcus lanatus L.* (Meharg and Macnair, 1991). Similarly, Rumburg et al. (1960) reported that increasing concentrations of P decreased the amount of arsenate removed by oats from a nutrient solution. For Indian mustard (*Brassica juncea*), phosphorus addition resulted in a reduction of As uptake by 55–72% over the control (Pickering et al., 2000). However, in the soil system, phosphate addition increases available arsenic by replacing adsorbed arsenic, thus resulting in elevated arsenic uptake (Jacobs and Keeney, 1970; Turpeinen et al., 1999). It is not surprising that the differences of As availability occurred between soil and hydroponic systems since other soil parameters (Eh, pH) also influence As solubility or mobility (Meharg and Hartley-Whitaker, 2002). A positive correlation was found between P and As in the fern plants ($r^2 = 0.83$, $P < 0.05$). Similar results were reported by Komar (1999) with a positive correlation between plant P and As concentration in Chinese brake fern. Increasing cell phosphorus levels reduced formation of the arsenate-substituted ATP analogue and therefore increased overall arsenic tolerance (Meharg et al., 1994). The phosphate uptake system, by which arsenic is taken up (Meharg et al., 1994), is induced under a low phosphate status like in

some angiosperms and fungi. Synergistic effects of P addition to As contaminated soils may be another explanation for the enhancement of As uptake by Chinese brake fern. The reason for this synergistic effect is unclear, but may be related to P nutrition. Since As can replace P in plants, but is unable to carry out the role of P in energy transfer, the plants reacts as if there is a P deficiency. Thus, as plant As increases, the plants reacts by increasing P uptake (Burlo et al., 1999; Carbonell-Barrachina et al., 1998).

The effects of composts varied in the two soils. Both composts increased As uptake from the CCA soil, but decreased As uptake from the ASC soil. Enhanced As uptake from the CCA soil is related to the WS–As increase (Fig. 3a) and the transformation of As(V) to As(III) (Table 2). As(III) increased from 9.7 to 20% and 24.2% of the As in soil solution for the MSW and BS treatments, respectively. Heeraman et al. (2001) suggested that WS–As is a good predictor for plant uptake of As. Sadiq (1986) also indicated a positive relation between water extractable As and plant uptake in corn. It has been observed that As(III) has a higher availability to the plants than As(V) (Carbonell-Barrachina et al., 1998; Marin et al., 1992). Contrary to the CCA soil, compost amendments reduced WS–As in the ASC soil (Fig. 3b), resulting in the reduction of As uptake by the fern (Fig. 4b).

The Chinese brake ferns accumulated much more As from ASC soil than from CCA soil in all treatments (Table 4) because the ASC soil contained more As in the bioavailable fractions (WS–As, Al–As) than the CCA soil (Fig. 1). For the CCA soil, compost amendments enhanced but were not significantly ($P < 0.05$) different from As accumulations in the controls (Table 4), whereas phosphate treatment had the greatest plant arsenic accumulation, at more than three times that of the control. After 12 weeks, PR treatment significantly increased arsenic removal from 2.56 up to 8.27%. Con-

trary to the CCA soil, composts significantly reduced As accumulation from ASC soil. The PR treatment increased As accumulation (Table 4). After 12 weeks, plants with compost amendments removed < 8%As from ASC soil, less than the 11.9% removed from the control. PR showed a significant amount of arsenic removal from the ASC soil (14.4%). Most of the arsenic (> 90%) taken up by the fern was accumulated in the fronds (Table 4).

It should be pointed out that arsenic volatilization may have occurred in the compost amended CCA soils, as less than 84% of As was recovered (Table 5). Up to 16% As loss from the soils may be attributable to microbially-mediated arsenic volatilization in the CCA soil. Loss of As from solution in reduced soils has long been attributed to arsenic volatilization as arsine gas (Onken and Adriano, 1997). It has been proven that application of organic composts reduces soil redox potential, especially when the soil pH is higher (Onken and Adriano, 1997). In such an environment, arsenate is easily reduced to arsenite and then methylated to form methylarsonic acid. These As compounds may further be reduced to methylarsines that volatilize to the atmosphere (Sadiq, 1997). Soil microbes have been shown to produce volatile arsenicals by a reductive pathway from inorganic and methylated forms of As (Onken and Adriano, 1997). Akins and Lewis (1976) added DSMA-[74]As to a soil system and measured a loss of [74]As. The loss of [74]As from the reduced soil system was attributed to the gaseous evolution of arsine, though no arsine was detected. In our experiments, up to 12% arsenic loss was also observed in compost-amended CCA soil even without a fern present (data not shown). No significant change in arsenic was observed in the PR-treated soils. This further supported the ideas that compost induced transformation of arsenate to arsenite, which was further transformed into volatile arsenic.

Table 4
Arsenic accumulation and distribution in Chinese brake ferns grown in the CCA and ASC soils

	As accumulation (mg plant^{-1})			As distribution (%)		As removed % of soil As
	Shoot	Root	Sum	Shoot	Root	
CCA soil[a]						
Control	5.21 ± 1.11b[b]	0.44 ± 0.02b	5.65 ± 1.23b	92.2a	7.78a	2.56b
MSW	5.99 ± 1.07b	0.40 ± 0.05b	6.39 ± 1.85b	93.7a	6.26a	2.90b
BS	6.16 ± 1.34b	0.47 ± 0.04b	6.63 ± 1.07b	92.9a	7.09a	3.01b
PR	17.6 ± 3.25a	0.58 ± 0.06a	18.2 ± 2.59a	96.8a	3.18b	8.27a
ASC soil						
Control	20.7 ± 4.21a	1.73 ± 0.54a	22.5 ± 2.62a	92.3a	7.69a	11.9b
MSW	13.9 ± 1.24b	1.46 ± 0.33a	15.4 ± 1.74b	90.5a	9.48a	8.15c
BS	7.33 ± 0.87c	0.48 ± 0.14b	7.81 ± 0.27c	93.8a	6.15a	4.13d
PR	21.6 ± 3.15a	1.92 ± 0.81a	23.5 ± 3.11a	91.8a	8.17a	14.4a

[a] CCA, chromated-copper-arsenate; ASC, As spiked contaminated; MSW, municipal solid waste; BS, biosolid; PR, phosphate rock.
[b] Mean ± standard deviation ($n = 3$), values ending in the same letter within each column are not significantly different ($P < 0.05$).

Table 5
A mass balance of As in the CCA soil an AAC soil (mg pot^{-1})

	Soil As (week 12)	Fern As (week 12)	Soil + Fern (week 12)	Original (week 0)	% (Sum/total)
CCA soil[a]					
Control	210±21.3a[b]	5.65±1.23b	215±19.3a	225a	95.5a
MSW	182±23.5a	6.39±1.85b	188±21.4b	227a	82.8b
BS	186±19.2a	6.63±1.07b	192±18.7b	230a	83.5b
PR	191±17.4a	18.2±2.59a	209±23.5a	221a	94.6a
AAC soil					
Control	136±15.9a	23.5±3.11a	159±16.1a	171a	93.0a
MSW	137±10.9a	15.4±1.74b	153±12.3a	170a	90.0a
BS	156±19.8a	7.81±0.27c	164±13.1a	168a	97.6a
PR	144±6.52a	22.5±2.62a	166±9.56a	181a	91.7a

[a] CCA, chromated-copper-arsenate; ASC, As spiked contaminated; MSW, municipal solid waste; BS, biosolid; PR, phosphate rock.
[b] Mean±standard deviation ($n=3$ values ending in the same letter within each column are not significantly different ($P<0.05$).

3.5. Arsenic leaching in soils

The effects of soil amendments on As leaching in soils were studied using column experiments. Effluent As concentrations reached steady-state levels within 10 pore volumes (data not shown). The ASC soil had greater leaching of As than the CCA soil at the end of 12 weeks (Table 6). It is because ASC soil As was predominantly associated with exchangeable and aluminum oxide fractions (~80% of total As), while the CCA soil contained approximately 16% of the total As in these fractions (Fig. 1).

Without a fern to absorb soil solution As, soil amendments significantly increased As leaching in the CCA soil. The greatest effect was with the compost treatments that had > 50% increase when compared with the controls. As a result of arsenic transformation to As(III) and competitive desorption with DOC, arsenic leaching from the CCA soil was enhanced in the compost amended soils with the absence of a fern plant (Table 6). In the experiment of Turpeinen et al. (1999) there was clear evidence that microbes enhance As leaching in soil. In formaldehyde-treated soil samples

there was no growth in microbial plate counts and arsenic leaching was greatly reduced. Also, Ahmann et al. (1997) reported that in autoclaved or formaldehyde-treated samples arsenic mobilization (or release) was much lower than in the control samples. Organic matter from composts provides a carbon source for microbes to enhance bioleaching in addition to promoting their growth. Chirenje et al. (2002) noted that arsenic was related to the DOC in the effluents of a column leaching experiment. Similar to this study, arsenic concentrations were positively correlated to DOC in the effluents (data not shown) as the DOC increased after compost addition. Compost amendments in this study increased arsenic leaching, corresponding to significantly higher DOC leaching. As hypothesized, uptake by ferns reduced As leaching with the biggest decline in the PR treatment (Table 6). The PR treatment with ferns decreased 58.5% of As leaching in the CCA soil, respectively, compared with that without ferns.

Displacement of As by P from the sorption sites increased arsenic mobility in the CCA soil when there is no fern root to absorb the As from the soil solution (Table 6). This is consistent with the mechanism of P-enhanced release of As in the soil and subsequent promotion of As movement through the soil by competition of dissolved As and P for ion adsorption sites. (Davenport and Peryea, 1991) reported that phosphate addition significantly increased the amount of As leached from the soil. Nevertheless, in the presence of Chinese brake ferns, phosphate showed the biggest decrease in As leaching as compared to the other treatments (Table 6). This is most likely attributable to the high As uptake.

For the ASC soil, soil amendments reduced As leaching regardless of fern presence with the exception of phosphate application which actually increased slightly As leaching without planting. Arsenic aging immobilization may be responsible for this reduction in the ASC soil since arsenic spiked was equilibrated with soil for only 1 week prior to treatment with compost and phosphate rock.

Table 6
Cumulative mass of soluble As leached from soil columns (10 pore volumes of leaching elution) constructed from treatments after the end of the 12 week study

	CCA soil mg As kg^{-1}		ASC soil mg As kg^{-1}	
	With fern	Without fern	With fern	Without fern
Control[a]	14.6±1.23a[b]	20.7±3.27c	16.5±1.83a	40.5±4.27a
MSW	12.4±0.98b	31.5±2.43a	12.4±0.31b	31.5±3.21b
BS	14.4±2.31a	33.2±3.21a	11.5±0.69b	29.6±2.33b
PR	11.6±1.93b	28.0±1.72b	10.1±1.21b	41.7±3.11a

[a] CCA, chromated-copper-arsenate; ASC, As spiked contaminated; MSW, municipal solid waste; BS, biosolid; PR, phosphate rock.
[b] Mean±standard deviation ($n=3$), values ending in the same letter within each column are not significantly different ($P<0.05$).

With time, As moves to mineral forms which are in equilibrium with the present soil environment. However, the greatest reduction (75.7%) of As leaching was observed in the phosphate amendment in the presence of the fern.

4. Conclusions

Phosphate addition significantly enhanced As uptake by Chinese brake fern, with frond As concentrations increasing up to 265% as compared with the control. After 12 weeks, plants grown in phosphate-amended soil removed up to 8.27% of the As from the CCA soil and 14.4% from the ASC soil. The enhanced uptake of As in the phosphate treatment was attributable to the displacement of soil As by P from adsorption sites into the soil solution. The effect of compost on As uptake depended on soil properties (e.g. pH). In the CCA soil with a neutral pH, compost treatments may have induced an anaerobic environment in the soil, which was favorable for the conversion of As (V) to the mobile As (III), thereby facilitating As uptake by the fern. In contrast, As adsorption onto organic matter applied in acidic soil may be responsible for the decrease of As uptake in the ASC soil after treatment with compost. The Chinese brake fern took up As mainly from Fe–As and Ca–As fractions in CCA soil, and from WE–As fraction in ASC soil. Both compost and phosphate amendments increased As leaching from CCA soil in the absence of the fern, but decreased in the presence of the fern. For the ASC soil, both treatments reduced As leaching regardless of the presence of the fern. The results indicate that growing Chinese brake fern with the application of phosphate rock is more effective for remediating As-contaminated soils.

Acknowledgements

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