



# Phytoremediation of As-contaminated soils by As-hyperaccumulator Pteris vittata: long-term efficiency and biomass disposal

Evandro B. da Silva, Jay T. Lessl, Ann C. Wilkie and Lena Q. Ma

Soil and Water Sciences Department, University of Florida

#### Abstract

- Arsenic (As) is toxic to plants, animals and humans. As-hyperaccumulator *Pteris vittata* (PV) can be used in As-contaminated soil remediation. However, disposal of the Asladen biomass might present a drawback of the phytoremediation process.
- The ability of *P. vittata* in taking up As from three As-contaminated soils was assessed over 6 years using pots containing 162 kg soil, each supporting 9 plants. Besides, As extraction from PV biomass was assessed using water and 30% ethanol solution.
- P. vittata reduced soil As concentrations by 45-47%, from 129, 26.6 and 29.8 mg kg<sup>-1</sup> to 69, 16 and 14 mg kg<sup>-1</sup>. However, efficiency decreased over time.
- Arsenic in *P. vittata* biomass was ~60% water soluble and 85% ethanol soluble. Addition of MgCl<sub>2</sub> helped formation of magnesium arsenate precipitate [Mg<sub>3</sub>(AsO<sub>4</sub>)<sub>2</sub>], reducing solution As concentration to  $\leq 2 \text{ mg L}^{-1}$ .



Figure 1. Remediation experiment site with shading

Introduction

Due to its mutagenic and carcinogenic potential and the increase of contaminated areas, Arsenic is a major environmental concern. Phytoremediation is a low-cost green technology that can remedy contaminated soils. Pteris vittata (PV) is an As hyperaccumulator (Ma et al., 2001). Hyperaccumulator plants have the ability to absorb and accumulate more than 1000 mg kg<sup>-1</sup> of metal by dry mass. Besides, PV roots are reported to release large amounts of root exudates that can solubilize As from insoluble forms such as Al-As and Fe-As in soils.

Proper husbandry practices can increase the potential of PV to remediate Ascontaminated sites. However, As-laden biomass disposal might present a drawback of the phytoremediation process. Usually the biomass is either disposed at regulated landfills or incinerated. Besides, As in dried-fronds can be mobile, which can be leached out leading to secondary contamination. Therefore, it is essential to develop an effective pre-treatment strategy prior to biomass disposal.

#### Materials and Methods

- Three soils were collected, one from an abandoned wood treatment facility (CCA) and two from an abandoned cattle dipping vat site (DVA and DVB).
- Soil was air-dried, sieved through a 2 mm screen, and mixed with 15 g kg<sup>-1</sup> phosphate rock [PR, Ca<sub>10</sub>(PO<sub>4</sub>)<sub>6</sub>F<sub>2</sub> (CaCO<sub>3</sub>)<sub>x</sub>, <1 mm]. Each soil (CCA-PR, DVA-PR, and DVB-PR) was placed into a pot containing 162 kg and 30 cm deep.
- Similarly-sized *P. vittata* plants were spaced 15 cm apart 9 per container with 4 replicates. For comparative analysis, *P. vittata* ferns were set up in un-amended soil in addition to pots with no PR and plants (CCA-P and DVA-P).
- Frond harvests and soil samples were taken every 6 months over the past 6 years. Fronds were clipped to ~20 cm from the rhizome (leaving new fiddleheads). Soil cores were taken from four points in each container of two depths 0-15 cm and 15-30 cm. Sequential extraction was performed to analyze the nature of As depletion.
- Arsenic from biomass was extracted using water, 2.1% nitric acid, 2.1% H<sub>3</sub>PO<sub>4</sub>, 1 M NaOH and 30% ethanol. Liquid-solid ratio was 25:1. After As extraction, MgCl<sub>2</sub> was added at Mg:As molar ratio of 4:1.

**Table 1.** Characterization of soils collected from a wood treatment facility (CCA) and two soils from a cattle dipping vat site (DVA and DVB) after 6 years (n=3).

|             | CCA                 | DVA            | DVB            |  |  |
|-------------|---------------------|----------------|----------------|--|--|
|             | mg kg <sup>-1</sup> |                |                |  |  |
| As initial* | $129 \pm 5.6$       | $29.9 \pm 1.1$ | $25.5 \pm 0.9$ |  |  |
| As          | $69 \pm 8.7$        | $16 \pm 1.6$   | $14 \pm 2.6$   |  |  |
| Cr          | $170 \pm 12$        | $7.9 \pm 1.2$  | $31.2 \pm 7.0$ |  |  |
| Mn          | $141 \pm 15$        | $37 \pm 2.9$   | $25 \pm 2.9$   |  |  |
| Fe          | $4003 \pm 127$      | $580 \pm 31$   | $2657 \pm 329$ |  |  |
| Cu          | $80 \pm 4.0$        | $1.2 \pm 0.8$  | $1.2 \pm 0.3$  |  |  |
| Zn          | 306 ± 18            | 14 ± 3.9       | 27 ± 6.0       |  |  |

<sup>\*</sup> Initial soil As concentration

± standard deviation

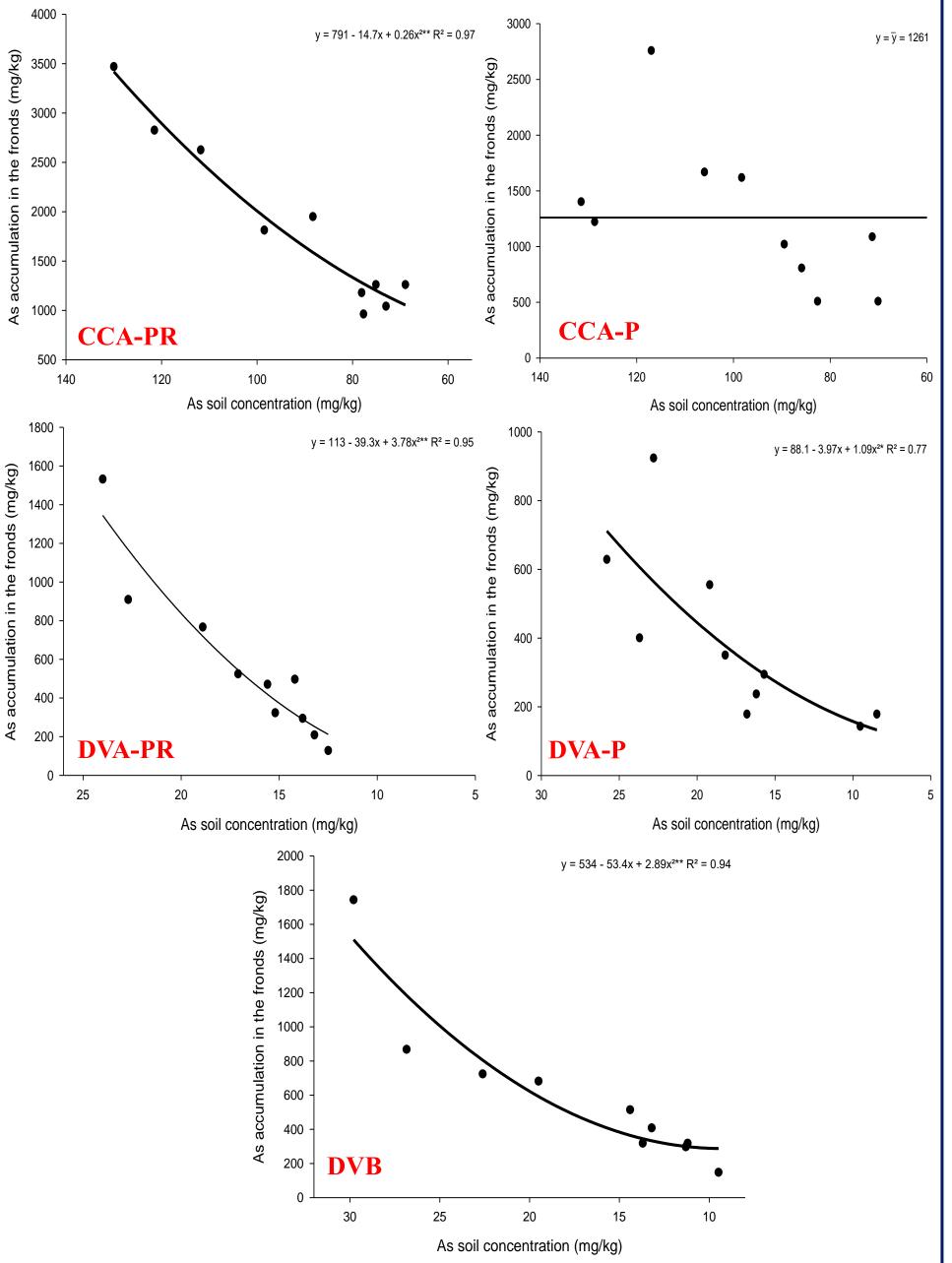
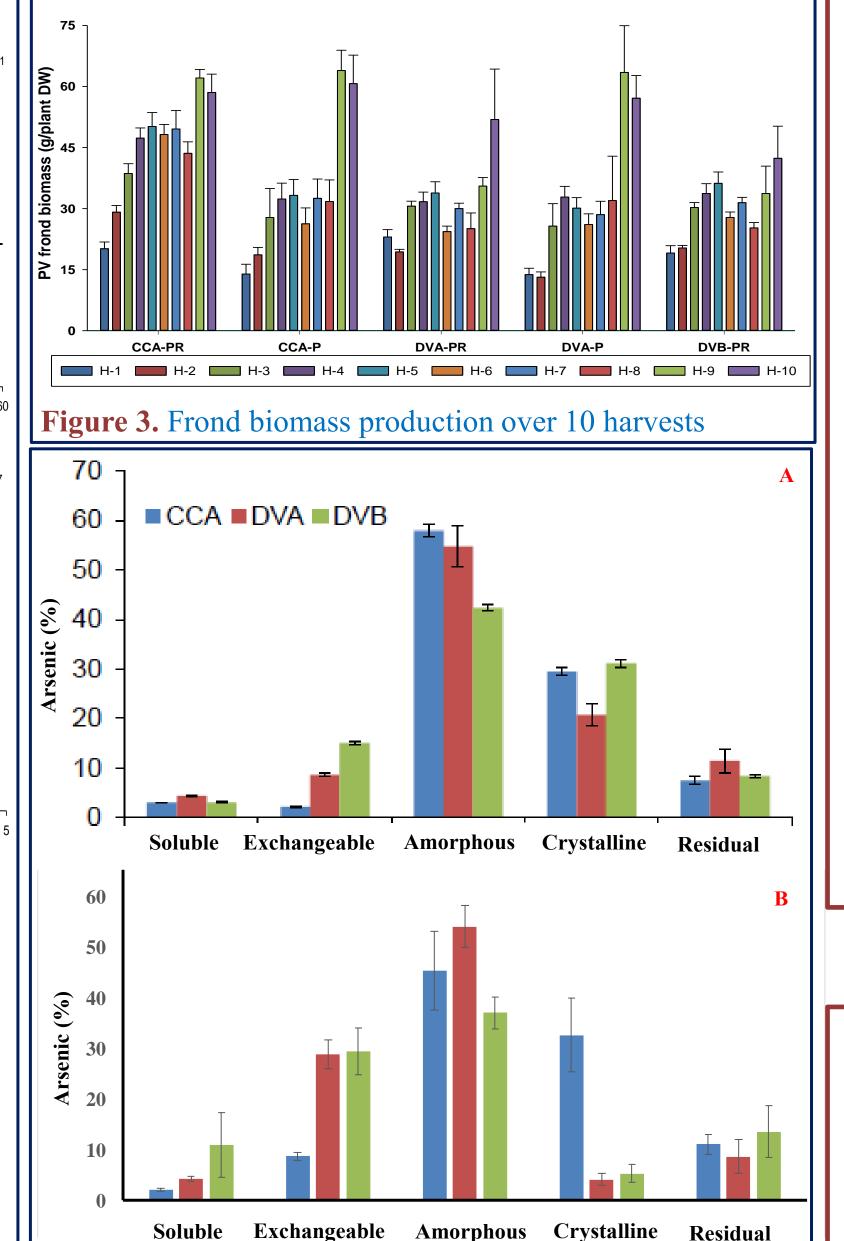


Figure 2. Correlation of As concentration in PV fronds to total As concentration in soil.



**Figure 4.** Soil arsenic fractionation (%) of the PR treatments soils before (A) and after (B) 6 years of remediation with *P. vittata* 

Results

- P. vittata reduced soil As concentrations in CCA, DVA and DVB soils by 45-47%, from 129, 26.6 and 29.8 mg kg<sup>-1</sup> to 69, 16 and 14 mg kg<sup>-1</sup>, respectively. However, the ability of *P. vittata* to remediate arsenic-contaminated soils was decreasing over time.
- Arsenic concentrations of frond biomass in PR amended CCA and DVA were higher than nonamended treatments.
- Arsenic removal was the greatest from the amorphous hydrous oxide-bound and crystalline fractions, with no difference between treatments (data not shown).
- Arsenic in fronds biomass was 67% water soluble. while ethanol extraction removed 85%.
- Addition of MgCl<sub>2</sub> helped formation of magnesium arsenate precipitate  $[Mg_3(AsO_4)_2]$ , reducing solution As concentration to  $< 2 \text{ mg L}^{-1}$ . In ethanol extraction, this formation was spontaneous due to Mg removal from chlorophyll.

### Conclusions

- P. vittata was effective to remediate arseniccontaminated soils. However, efficiency decreased over time.
- P. vittata was able to extract non-labile As in soil, a unique ability of *P. vittata*.
- Ethanol presented an advantage to extract As from PV frond biomass due to spontaneous formation of magnesium arsenate precipitate  $[Mg_3(AsO_4)_2]$ .

## Acknowledgments

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#### Table 2 Arsenic extraction from *Pteris vittata* frond biomass (n=3)

| Table 2. Arsenic extraction from <i>Pierts vittata</i> frond biomass (n–3). |       |                 |                                     |                 |             |  |  |
|---|-------|-----------------|-------------------------------------|-----------------|-------------|--|--|
|   | Water | 2.1% HCl        | 2.1% H <sub>3</sub> PO <sub>4</sub> | 1 M NaOH        | Ethanol 30% |  |  |
| 1 <sup>st</sup> extraction (g)  | 0.67  | $3.2 \pm 0.2$   | $3.1 \pm 0.2$                       | $1.9 \pm 0.1$   | 2.44        |  |  |
| 2 <sup>nd</sup> extraction (2.1% HCl) (g)                                   | -     | $1.3 \pm 0.2$   | $1.06 \pm 0.1$                      | $0.67 \pm 0.2$  | 0.44        |  |  |
| Remaining in biomass (g)  | 0.52  | $0.41 \pm 0.18$ | $\textbf{0.44} \pm \textbf{0.18}$   | $0.31 \pm 0.01$ | 0.51        |  |  |
| Recovery (%)  | 108   | 132             | 125                                 | 75              | 85          |  |  |
| Total (g)   | 1.10  | 3.99            | 3.99                                | 3.99            | 3.99        |  |  |

± standard deviation