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Factors influencing arsenic accumulation by *Pteris vittata*: A comparative field study at two sites

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Nutrient elements influenced the arsenic-accumulation capacity of Pteris vittata in the field.

Abstract

This study compared the factors influencing arsenic (As) accumulation by *Pteris vittata* at two sites, one containing As along with Au mineralization and the other containing Hg/Tl mineralization. The soils above these two sites contained high As concentrations (26.8–2955 mg kg⁻¹). Although the As concentration, pH, soil cation exchange capacity and plant biomass differed significantly between the two sites, no differences were observed in the As concentrations in the fronds and roots, or the translocation factors, of *P. vittata*, suggesting that this species has consistent As hyperaccumulation properties in the field. The As concentration in the fronds was positively related to phosphorus (P) and potassium (K), but negatively related to calcium (Ca), at one site. This suggested that P, K and Ca influenced As accumulation by *P. vittata* in the field.

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1. Introduction

Since the discovery of the first known arsenic (As) hyperaccumulator, *Pteris vittata*, in China and the USA, growing interest has been focused on the mechanism of As uptake and accumulation by this fern. *P. vittata* has high As tolerance and good As-accumulating abilities, and was shown to grow healthily both in tailings containing 23 400 mg kg⁻¹ As on a mining site and in soils containing 1500 mg kg⁻¹ spiked As under greenhouse conditions (Ma et al., 2001; Chen et al., 2002a). Another fern, *Pityrogramma calomelanos*, was also reported to be an As-hyperaccumulator and showed great potential in the phytoremediation of As-contaminated soils in Thailand (Francesconi et al., 2002; Visoottiviseth et al., 2002). Additional screening for As hyperaccumulators among fern

species, especially within the genus *Pteris*, has been carried out using field surveys and/or greenhouse experiments. *Pteris cretica* has been reported as another As-hyperaccumulator (Meharg, 2002; Wei et al., 2002; Zhao et al., 2002).

P. vittata from Florida in the USA (Ma et al., 2001), Hunan in China (Chen et al., 2002a) and Thailand (Visoottiviseth et al., 2002) all show high As concentrations in the fronds. Pteris longifolia, which is the most closely related fern species to P. vittata, is widely distributed throughout northern USA and Europe, and has been proven to be an As-hyperaccumulator (Meharg, 2002). Although field surveys and pot/hydroponic experiments have confirmed the As-hyperaccumulation properties of P. vittata (Chen et al., 2002a,b; Tu et al., 2002; Tu and Ma, 2002; Zhao et al., 2002), no previous studies have compared As accumulation by P. vittata from different localities in the field.

Arsenic is usually present in ores of gold (Au), copper (Cu), lead (Pb), tin (Sn) and zinc (Zn) (O'Neill, 1995). Previous surveys showed that soils in some mineralized sites in southern

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China had elevated As levels from geogenic sources. *P. vittata* growing on different mineralized sites might provide valuable information on As accumulation and its relationship with nutrient elements. The purposes of the present study were as follows: first, to determine the differences in As-accumulation patterns of *P. vittata* from two sites with high As levels from different geogenic sources; and second, to investigate the possible relationship between the accumulation of As and major nutrient elements in *P. vittata*.

2. Materials and methods

2.1. Sites and sampling

Two sites, Zimudang (ZMD) and Lanmuhang (LMC), with high geogenic As levels were chosen for this study. Both sites were located near Huilong town, Xinren City, in the southwest Guizhou province of China, with a north latitude of 25°34′22″ and an east longitude of 105°27′58″ for ZMD, and a north latitude of 25°31′40″ and an east longitude of 105°30′32″ for LMC. The distance between the two sites was 12 km. However, the geology differed markedly between the two sites: ZMD was an Au-mineralized site, while LMC was an unexploited mercury/thallium (Hg/Tl) — mineralized site. As was present at both sites, and neither site had been contaminated by anthropogenic sources of mining or smelting activities. Samples of *P. vittata* and soil from around the roots were collected at both sites. A distance of more than 20 m was kept between the samples to ensure that they were independent and representative.

2.2. Determination of arsenic and soil physical—chemical properties

In the laboratory, the fern samples were separated into fronds (above ground) and roots (below ground), washed thoroughly under tap water, rinsed in deionized water three times and then dried in an oven at 70 °C for 5 h. Plant samples were powdered using an electric miller and mixed thoroughly to ensure that they were homogenized. Soil samples were air dried, separated from stone and plant debris, and passed through a 20 mesh (0.84 mm) before the soil pH, grain size (soil texture) and cation-exchange capacity (CEC) analyses. Sub-samples (~15 g) were ground with a quartz mortar and pestle and passed through a 100 mesh (150 µm) for chemical analysis. The pH was measured using a 1:2.5 ratio of soil to deionized water, the CEC was determined by the neutral ammonium acetate method (Rhoades, 1982) and the total organic matter (TOM) was calculated using the Walkley-Black method (Nelson and Sommers, 1982). The grain size of the soil was determined by laser-diffraction analysis (Mastersizer 2000, Malvern, UK). The soil and plant powders were digested by a mixture of HNO3 and HClO4 on an electric hot plate at 120 °C. Thiourea and ascorbic acid were added as reducing reagents, and the final solution for the determination of total As contained 5% HCl. The As concentration was determined using a hydride-generation atomic-fluorescence spectrometer (AFS830, Beijing Jitian Xiaotiane Company, China). The concentrations of calcium (Ca), Cu, iron (Fe), potassium (K), magnesium (Mg), manganese (Mn), sodium (Na), phosphorous (P), sulphur (S) and Zn were determined by inductively coupled plasma atomic-emission spectroscopy (ICP-AES). Standard soil and plant references (Centre for Standard Reference of China) were used to check the accuracy of the chemical analysis. Milli Q water (18 Ω) was used to dilute the digested residues. All reagents were of analytical grade or better.

2.3. Statistical analysis

Statistical analysis was carried out using SPSS software. Student *t*-tests were used to compare the following data from the two sites: grain size, CEC, As and nutrient concentrations in the fronds and roots of *P. vittata*, and As and nutrient concentrations in the soil around the roots. Correlations were evaluated using the bi-variation method, with two-tailed significance and Pearson correlation coefficients. Principle components analysis (PCA) was used to test the inter-relationships among the different elements in *P. vittata* at each site.

3. Results and discussion

3.1. Ecology of P. vittata at the two sites

Although the soils at both sites contained high levels $(26.8-2955 \text{ mg kg}^{-1})$ of As from geogenic sources, P. vittata grew well, which indicated high As tolerance. The analytical results showed only minor changes in pH, CEC, TOM and soil texture for both sites, suggesting that the soil properties at each were relatively uniform. However, the As concentrations varied greatly and showed marked heterogeneity in the field. The soils at the two sites differed significantly in pH: the mean value was 7.1 (6.5-8.0) at ZMD compared with 5.9 (5.0-6.7) at LMC. The sand and silt contents as well as the CEC at the two sites were also significantly different. The As and P concentrations in the soils of the two sites were significantly different, with higher As and lower P at ZMD compared with those at LMC (Table 1). Thus, the soil properties between the two sites at which P. vittata grew were evidently different.

3.2. Hyperaccumulation of arsenic by P. vittata

All of the plant samples contained high As concentrations, with average values of 1104 and 810 mg kg⁻¹ in the fronds at ZMD and LMC, respectively. The As concentrations in the roots were lower than those in the fronds, with average values of 690 and 536 mg kg⁻¹ at ZMD and LMC, respectively. These results were consistent with previous reports on *P. vittata* grown under controlled pot/hydroponic conditions (Chen et al., 2002b; Zhao et al., 2002; Cao, et al., 2003; Tu and Ma, 2003, 2005). More than one-half of the samples of *P. vittata* had greater As concentrations in the fronds than those detected in the soils (data not shown). The average translocation factors (the ratio of the As concentration in the fronds

Table 1
Arsenic concentration and basic property of soils at the two sites

Sites	pН	CEC	TOM	Texture (%)			As $(mg kg^{-1})$	$P (mg kg^{-1})$	Fe (mg kg ⁻¹)
		$(\text{mmol } 100 \text{ g}^{-1})$	$(g kg^{-1})$	Sand	Silt	clay			
ZMD	7.1 ± 0.4**	28.3 ± 3.8**	6.2 ± 1.2	20.1 ± 7.7	22.8 ± 2.2*	42.7 ± 6.9	1342 ± 653**	973 ± 246	65928 ± 7337
LMC	5.9 ± 0.4	19.7 ± 7.7	8.3 ± 2.3	$41.4 \pm 5.1*$	22.9 ± 2.4	36.7 ± 3.4	509 ± 406	$1547 \pm 365**$	$56750 \pm 11475*$

ZMD: Zimudang (n = 14), LMC: Lanmuchang (n = 13).

Statistical difference of the data between the two sites at the level of *P < 0.05, **P < 0.01. Data are presented as means \pm SD.

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