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# Effect of phosphate minerals on phytoremediation of arsenic contaminated groundwater using an arsenic-hyperaccumulator

Guang-Mei Yang [a,c], Ling-Jia Zhu [b], Jorge A.G. Santos [d], Yanshan Chen [a], Gang Li [e], Dong-Xing Guan [f,*]

[a] State Key Laboratory of Pollution Control and Resource Reuse, School of the Environment, Nanjing University, Nanjing 210023, China
[b] Key Laboratory of Soil Environment and Pollution Remediation, Institute of Soil Science, Chinese Academy of Sciences, Nanjing 210008, China
[c] Department of Agronomy and Horticulture, Jiangsu Vocational College of Agriculture and Forestry, Jurong 212400, China
[d] Soil and Water Science Department, University of Florida, Gainesville, Florida, 32611, United States
[e] Key Laboratory of Urban Environment and Health, Institute of Urban Environment, Chinese Academy of Sciences, Xiamen 361021, China
[f] Key Laboratory of Surficial Geochemistry, Ministry of Education, School of Earth Sciences and Engineering, Nanjing University, Nanjing 210023, China

## HIGHLIGHTS

- *Pteris vittata* was used to remediate arsenic-contaminated groundwater.
- Phosphate rock (PR) and hydroxyapatite (HA) were applied as sparingly-soluble phosphate sources.
- Nitrogen and P concentrations in the solutions were monitored to check eutrophication pollution.
- PR was an ideal P source to facilitate As removal from contaminated groundwater by *P. vittata*.

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## ABSTRACT

To investigate the effect of phosphate sources on phytoremediation of arsenic (As) contaminated groundwater using *Pteris vittata*, a hydroponic experiment was conducted. Sparingly-soluble phosphate minerals (phosphate rock, PR, and hydroxyapatite, HA) and soluble Hoagland solution (HS, containing ammonium phosphate) were chosen as phosphate sources. Arsenic content in the residual nutrient solutions was analyzed to acquire the phytoextraction effect. The results showed that As concentrations in the nutrient solutions treated with sparingly-soluble P decreased more rapidly than that treated with soluble P. For treatment with sparingly-soluble PR, *P. vittata* reduced As concentration more rapidly than that in HA treatment and HS control. Nitrate and P concentrations in the residual solutions were also monitored to check eutrophication pollution. Phosphorus in the residual solutions in PR or HA treatment was not detected and 0.5 $\mu g\ L^{-1}$, respectively, much lower than HS control. These results indicated that sparingly-soluble PR could be an ideal P source to facilitate As removal from contaminated groundwater by *P. vittata* and ensure the nutrient quality of the residual water after phytoremediation.



* Corresponding author.
*E-mail address:* dxguan@nju.edu.cn (D.-X. Guan).

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## 0. Introduction

Arsenic (As) is a carcinogenic metalloid, which can be released into the environment by human activities like mining, industry and agriculture, causing global environmental pollution and threatening human health (Nordstrom, 2002; Zhu et al., 2014). Drinking groundwater with elevated As concentrations is one of the main As exposure pathways (Ng et al., 2003; Yu et al., 2007; Phan et al., 2010). Till now, geogenic As contamination of groundwater occurred in various regions, particularly in south & southeast Asian countries, such as Bangladesh, India and China (Fendorf et al., 2010; Rodriguez-Lado et al., 2013). In some places of these areas, contaminated groundwater may contain up to 3000 $\mu g\ L^{-1}$ As, hundred times higher than the World Health Organization (WHO) threshold value (10 $\mu g\ L^{-1}$) (Smedley and Kinniburgh, 2002; Chakraborti et al., 2008; Rahman et al., 2009). Chakraborti et al. (2004) analyzed more than 50,000 groundwater samples from 64 districts of Bangladesh and found that about 43% samples have As concentration above 10 $\mu g\ L^{-1}$, and 28% have As concentration even above 50 $\mu g\ L^{-1}$. As a result, As exposure has reached epidemic proportions in Bangladesh and more than 200,000 would die of cancer from drinking As contaminated water in Bangladesh (Chakraborti et al., 2010). In addition, As contaminated groundwater has also been widely used for irrigation, leading to high As in food crop like rice and threatens livelihoods of local people (Williams et al., 2006; Stone, 2008; Senanayake and Mukherji, 2014). Since the available resources of freshwater are limited, more efforts should be devoted to reducing As concentration in groundwater.

The former maximum contaminant limit (MCL) for As in drinking water is 50 $\mu g\ L^{-1}$ and chronic exposure via drinking water with >50 $\mu g\ L^{-1}$ As may lead to diseases (Smith et al., 2002). The current MCL set for As in drinking water is 10 $\mu g\ L^{-1}$, which was recommended by WHO in 1993 (WHO, 1993). In 1998 European Union adopted the 10 $\mu g\ L^{-1}$ limit as a mandatory MCL for all its member countries (EU, 1998). Later in 2001, the USEPA also announced to adopt the MCL of 10 $\mu g\ L^{-1}$ for As in drinking water (USEPA, 2001). After the new As standard took effect, groundwater aquifers exceeding the MCL were required for cleanup to protect the ecosystem and human health.

The traditional treatment technologies for As-contaminated water include adsorption, precipitation, ion exchange, etc. Choong et al. (2007). However, these technologies often require pre-oxidation of arsenite ($As^{III}$; occurring predominantly as the neutral molecule $H_3AsO_3$ in groundwater) to arsenate ($As^{V}$; existing as anionic species $H_2AsO_4^-$ or $HAsO_4^{2-}$) and also generate spent media or sludge that need to be managed (Bissen and Frimmel, 2003). Phytoremediation is an effective and environmental friendly technology to remove contaminants from the environment using hyperaccumulating plants (Ali et al., 2013; Zhao et al., 2015a; Mahar et al., 2016). Arsenic hyperaccumulator *Pteris vittata* (Chinese brake fern) is highly efficient in As phytoextraction from environment and in As translocation to the fronds (Ma et al., 2001). Previous studies have also demonstrated the potential of *P. vittata* in phytoremediation of As-contaminated groundwater (Tu et al., 2004; Natarajan et al., 2008, 2011; Guo et al., 2012; Zhao et al., 2015b).

For optimal growth during phytoremediation, *P. vittata* needs to be applied with fertilizer, since groundwater is normally characterized by lacking sufficient nutrients (e.g. macroelements N and P) to sustain plant growth. However, the fertilizer containing inorganic N and P may introduce new contaminants to groundwater and the addition of P may also decrease $As^{V}$ uptake due to the competitive effect (Tu and Ma, 2003). Therefore, controlling the extraneous N and P to low levels is a new and challenging objective while using *P. vittata* for the remediation of As contaminated groundwater.

Arsenic hyperaccumulator *P. vittata* has been proved to be able to efficiently and sustainably dissolve phosphate minerals, as well as promoting plant growth and As uptake (Lessl and Ma, 2013). We hypothesized that the exertion of sparingly-soluble phosphate minerals, rather than soluble phosphate, was effective in cleaning up As-contaminated groundwater without causing water eutrophication. The main aim of this study was to explore the potential of sparingly-soluble P for the phytoremediation of As-contaminated groundwater using *P. vittata*. A hydroponic system was adopted to simulate As-contaminated groundwater and As concentrations in the plants and nutrient solutions were analyzed to determine the phytoextraction effect. In addition, N and P levels in the residual solutions were also monitored in our study.

## 1. Materials and methods

### 1.1. Experimental design

Spores of *P. vittata* were collected from Florida, USA, and they were surface-sterilized and sown on 1/2 Murashige and Skoog (MS) agar medium containing 0.8% agar at pH 5.8 (Chen et al., 2016). Gametophytes were germinated after 20 d, and after ~2 months of cultivation, sporophytes appeared. At 2–3 fronds stage, the sporophytes were transferred to soil. After cultivated for 3–5 months, *P. vittata* sporophytes with 6–8 fronds (with average length of ~15 cm) were acclimated in a hydroponics system containing 0.2-strength Hoagland solution (HS) for two weeks as described by Liu et al. (2015). Then *P. vittata* sporophytes were transferred to 2-L vessels (one plant per vessel) containing a modified 0.2-strength Hoagland solutions (pH 7.8) with different phosphorus sources: ammonium phosphate (HS treatment, control), phosphate rock (PR; containing 8% P and 24% Ca, $Ca_{10}(PO_4)_6F_2(CaCO_3)_X$; PCS Phosphate, White Springs, Florida) or hydroxyapatite (HA, $Ca_{10}(PO_4)_6(OH)_2$). All the solutions were spiked with 200 $\mu g\ L^{-1}$ As, which may represent the average As concentration in As contaminated groundwater in the Southeast Asia region (Rahman et al., 2009). The plants were grown for 40 d and aliquots of 1 mL nutrient solution were taken at 0, 10, 20, 30, and 40 d for analysis of As concentration.

In this experiment, all the plants were grown in a greenhouse under a 14 h photoperiod with a daily light intensity of 180 $\mu mol\ m^{-2}\ s^{-1}$, 20–25 °C temperature and 60%–70% humidity. After 40 d of growth, *P. vittata* sporophytes were harvested for As determination in roots and fronds.