



## **PHYTOREMEDIATION OF ARSENIC BY *Pteris vittata* L.**

### **- A REVIEW**

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### **Abstract**

In current decades arsenic contamination in soil and water has become a serious global concern. Phytoremediation is a promising green method for alleviating the arsenic contamination in soil. *Pteris vittata* is the first identified arsenic hyperaccumulator. This review presents the biochemical and molecular mechanisms involved in hyperaccumulation and phytoremediation of arsenic in *P. vittata*.

### **1. Introduction**

Environmental pollution by toxic heavy metals and metalloids has become a serious problem in the twenty-first century. Increased industrialization and urbanization are making this problem more serious. Among toxic heavy metal(loid), arsenic (As) is widely distributed in nature with low crustal abundance (0.0001%). Anthropogenic point sources such as slag, mine tailings, and wood treatment can further contribute to As found in the environment. Naturally occurring As is broadly distributed among subsurface areas used for drinking water around the globe [1]. As can be bioaccumulated and biomagnified through the food chain/web, and ultimately causes carcinogenic and neurotoxic effects on humans creating serious health hazards [2, 3]. There are several physical and chemical remediation methods for As removal from contaminated areas but most of them have limitations i.e. higher cost, intensive labor, irreversible in physical and chemical properties of soil, and hampering the life processes of soil microflora [4]. Phytoremediation is one such novel remediation method, considered as green alternative solution to the problem of toxic metal(loid)

pollution. The term “phytoremediation” can be defined as “the use of green plants and their associated microorganisms, soil amendments, and agronomic techniques to remove, contain, or render harmless environmental contaminants”[5]. Mainly hyperaccumulator plants are used as a phytoremediating agent which can be grown in toxic heavy metal(loid) contaminated sites to clean up the toxicants [6]. Typically the hyperaccumulator plants have the TF (translocation factor) and BCF (bio-concentration factor) values of more than 1, which sometimes reach 50-100 [7, 8]. Chinese brake fern (*Pteris vittata*) can effectively hyper-accumulate the As present in the environment in its above-ground biomass [9]. *P. vittata* has drawn extensive attention from researchers since the discovery of its bioremediation potential in 2001 [9]. This review is focused on recent molecular and biochemical studies related to the hyperaccumulation of As by *P. vittata*.

## 2. Arsenic hyperaccumulation by *P. vittata*

*P. vittata* has earned recognition in the scientific world as an As hyperaccumulator in 2001[9]. This plant is an excellent phytoextractor of As from soil, and can efficiently transport As through xylem to its above ground parts [9, 10]. Oldfrond of this plant can accumulate up to 13.8 g As Kg<sup>-1</sup> dry weight, which is 142 and 25 times higher than As in soil [11,] and its root [12]; respectively. Normal frond contains up to 93% of the total As content in the plant [9, 10]. Recent studies indicate that the basal and stalk cells of trichomes on the frond contain the highest levels of As compared to other frond tissues[13]. As plays a dual role in influencing the growth of *P. vittata* depending on its concentrations. Low levels of As in soil can stimulate the growth of *P. vittata*. For example, 100 ppm of As in the growth medium can increase by 40% in biomass compared to the control [9]. Beyond the threshold concentration of As in the growth medium growth is inhibited [14, 15].

## 3. Arsenic uptake, transport, detoxification and tolerance in *P. vittata*

Arsenate [As(V)] is the predominant form of As in aerobic soil over arsenite [As(III)], although they are interconvertible depending on the redox regime of the environment. As is taken up by the root system of *P. vittata* as form a of As(V) via phosphate transporter [16, 17, 18] because arsenate is a chemical analog of phosphate [16], whereas arsenite is mostly found in the frond suggesting that arsenate is reduced to arsenite in the plant body. Glutathione-dependent arsenate reductase is responsible for the reduction of arsenate to arsenite is only detected in the root of *P. vittata* indicating that arsenate reduction occurs mainly in the root [19]. Arsenite is translocated from root to shoots through the xylem in both hyperaccumulators and non-hyperaccumulators [20, 21]. Recent studies show three possibilities deal with arsenite transport in *P. vittata*: (1) Efficient Arsenite loading in the xylem is a key factor. (2) A higher degree of Arsenite complexation by thiol-containing compounds [17, 21]. (3) Lack of arsenite efflux from cells to the external environment of *P. vittata* helps inefficient translocation of arsenite from the roots to fronds. But for As non-hyperaccumulators rapid arsenite extrusion from root cells is observed [18]. Plant cells exhibit different strategies for As detoxification, like chelation by glutathione (GSH), vacuolar sequestration, synthesis of heat shock proteins (HSPs), and secondary metabolites productions. As in plant cells also enhances the production of GSH and phytochelatin (PC) biosyntheses. The key process in As detoxification is the formation of As-PC complexes [22]. Bleeker et al., [23] reported

an ABC-transporter involved in As-PC and As-GSH complexes to get sequestered in vacuoles. However, the plant produces stress proteins (SPs) and metallothioneins (MTs), constitutively expressed SPs have participated in protein folding. MTs act as a toxicant chelating agent and ROS scavenger [24]. The oxidative stress induced in plants by As accumulation within the cells causes rapid increase in ROS production. In As hyperaccumulators and tolerant plants, excessive ROS is scavenged by different antioxidants (e.g. lignin, flavonoids, glutathione, guaiacol, ascorbate, etc.). Cellular antioxidative pathways play a key role in maintaining ROS titer within As hyperaccumulators and tolerant plants [25].

#### 4. Molecular mechanisms of As hyperaccumulation in *P. vittata*

Identification of genes and molecular mechanisms of stress tolerance has been studied for last two decades mainly on the model plant *Arabidopsis thaliana* [26], or crop models like rice [27]. But *A. thaliana* and rice plants are not natural As hyperaccumulators, thus they are not suitable for molecular genetics study of As phytoremediation. Therefore, to unveil the molecular genetics behind the As hyperaccumulation functional cloning and analysis methods using *P. vittata* are profoundly required. *P. vittata* has a large genome size approximately 4834 Mb [28], which is 11 and 40 times those of rice and *A. thaliana*; respectively.

Recent molecular studies reveal that phosphate transporter in *A. thaliana* consists of nine members (PHT1;1 through PHT1;9). PHT1 a phosphate-H<sup>+</sup> symporter forms 12 transmembrane helices [29]. PvPht1;3 and PvPht1;5 show a similar affinity for phosphate but a higher affinity for As(V) in hyperaccumulator *P. vittata*. A low level of phosphate enhances the rate of arsenate uptake by root upregulating the transcript's expression of PvPht1;3 [30]. Arsenite is taken up by nodulin-26-like intrinsic proteins (NIPs), tonoplast intrinsic protein (TIP), inositol transporters (INT), and Si transporters [31, 32, 33, 34, 35]. However, NIP3;1 is involved in both arsenite translocation and uptake in *A. thaliana*. He et al. reported that PvTIP4;1 helps in arsenite transport in *P. vittata* [34].

#### Conclusion

Phytoremediation is an eco-friendly, solar-driven technology with moderate public acceptance. It is a relatively recent technology and is mostly in the research stage. In last decades, intensive research has been conducted by scientists to identify contaminants hyperaccumulators and characterization mechanisms of hyperaccumulation. Further studies should explore the role of rhizospheric microbes in promoting the phytoremediation efficiency of the hyperaccumulators. Studies on *P. vittata*-microbe symbiosis will provide insight into the mechanisms of microbe-assisted phytoremediation. Proper disposal of hyperaccumulators within the bio-ore after extraction of contaminants is urgently required. Finally, integrative approaches need to be adopted for the promotion of this green technology in environmental clean-up and enhancement of its social acceptability.

#### Acknowledgements

The authors acknowledge the comments and suggestions of the learned referee which have enriched the paper.

## References

- [1] A. H. Welch, D. B. Westjohn, D. R. Helsel, & R. B. Wanty, “Arsenic in ground water of the United States: occurrence and geochemistry” *Groundwater.*, 38(4), 589-604 (2000).
- [2] M. H. Puckett, Y. Zhang, B. Lu, Y. Lu, H. Sun, C. Zheng, & W. Wei, “Application of fractional differential equation to interpret the dynamics of dissolved heavy-metal uptake in streams at a wide range of scales”, *Eur. Phys. J. Plus.*, 134(8), 377 (2019).
- [3] A. Grzegórska, P. Rybarczyk, A. Rogala, & D. Zabrocki, “Phytoremediation—From environment cleaning to energy generation—Current status and future perspectives”, *Energies.*, 13(11), 2905 (2020).
- [4] S. P. McGrath, F. J. Zhao, & E. Lombi, “Plant and rhizosphere processes involved in phytoremediation of metal-contaminated soils”, *Plant Soil.*, 232(1) 207-214 (2001).
- [5] S. D. Cunningham, & D. W. Ow, “Promises and prospects of phytoremediation”, *Plant Physiol.*, 110(3), 715 (1996).
- [6] N. Gupta, K. K. Yadav, V. Kumar, S. Kumar, R. P. Chadd, & A. Kumar, “Trace elements in soil-vegetables interface: translocation, bioaccumulation, toxicity and amelioration-a review”, *Sci. Total Environ.*, 651, 2927-2942 (2019).
- [7] C. Cluis, “Junk-greedy greens: phytoremediation as a new option for soil decontamination”, *BioTeach J.*, 2(6), 1-67 (2004).
- [8] H. M. Liang, T. H. Lin, J. M. Chiou, & K. C. Yeh, “Model evaluation of the phytoextraction potential of heavy metal hyperaccumulators and non-hyperaccumulators”, *Environ. Pollut.*, 157(6), 1945-1952 (2009).
- [9] L. Q. Ma, K. M. Komar, C. Tu, W. Zhang, Y. Cai, & E. D. Kennelley, “A fern that hyperaccumulates arsenic”, *Nature.*, 409(6820), 579-579 (2001).
- [10] P. Visoottiviseth, K. Francesconi, & W. Sridokchan, “The potential of Thai indigenous plant species for the phytoremediation of arsenic contaminated land”, *Environ. Pollut.*, 118(3), 453-461 (2002).
- [11] C. Tu, L. Q. Ma, & B. Bondada, “Arsenic accumulation in the hyperaccumulator Chinese brake and its utilization potential for phytoremediation”, *J. Environ. Qual.*, 31(5), 1671-1675 (2002).
- [12] C. Tu, & L. Q. Ma, “Effects of arsenic concentrations and forms on arsenic uptake by the hyperaccumulator ladder brake”, *J. Environ. Qual.*, 31(2), 641-647 (2002).

- [13] W. Li, T. Chen, Y. Chen, & M Lei, “Role of trichome of *Pteris vittata* L. in arsenic hyperaccumulation”, *SCI CHINA SER C.*, 48(2), 148-154 (2005).
- [14] J. Wang, F. J. Zhao, A. A. Meharg, A. Raab, J. Feldmann, & S. P McGrath, “Mechanisms of arsenic hyperaccumulation in *Pteris vittata*. Uptake kinetics, interactions with phosphate, and arsenic speciation”, *Plant Physiol.*, 130(3), 1552-1561 (2002).
- [15] W.Zhang, Y. Cai, K. R. Downum, & L. Q Ma, “Thiol synthesis and arsenic hyperaccumulation in *Pteris vittata* (Chinese brake fern)”, *Environ. Pollut.*, 131(3), 337-345 (2004).
- [16] C. Y. Poynton, J. W. Huang, M. J. Blaylock, L. V. Kochian, & M. P. Elless, “Mechanisms of arsenic hyperaccumulation in *Pteris* species: root As influx and translocation”, *Planta.*, 219(6), 1080-1088 (2004).
- [17] F. J. Zhao, J. R. Wang, J. H. A. Barker, H. Schat, , P. M. Bleeker& S. P McGrath, “The role of phytochelatins in arsenic tolerance in the hyperaccumulator *Pteris vittata*”, *New Phytol.*, 159(2), 403-410 (2003).
- [18] Y. H. Su, S. P. McGrath, Y. G. Zhu, & F. J. Zhao, “Highly efficient xylem transport of arsenite in the arsenic hyperaccumulator *Pteris vittata*”, *New Phytol.*, 180(2), 434-441 (2008).
- [19] G. L. Duan, Y. G. Zhu, Y. P. Tong, C. Cai, & R Kneer, “Characterization of arsenate reductase in the extract of roots and fronds of Chinese brake fern, an arsenic hyperaccumulator”, *Plant Physiol.*, 138(1), 461-469 (2005).
- [20] V. G. Mihucz, E. Tatár, I. Virág, E. Cseh, F. Fodor, & G Záray, “Arsenic speciation in xylem sap of cucumber (*Cucumissativus* L.)”, *Anal. Bioanal. Chem.*, 383(3), 461-466 (2005).
- [21] A. Raab, H. Schat, A. A. Meharg, & J. Feldmann, “Uptake, translocation and transformation of arsenate and arsenite in sunflower (*Helianthus annuus*): formation of arsenic–phytochelatin complexes during exposure to high arsenic concentrations”, *New Phytol.*, 168(3), 551-558 (2005).
- [22] J. Hartley-Whitaker, G. Ainsworth, R. Vooijs, W. T. Bookum, H. Schat, , & A. A Meharg, “Phytochelatin are involved in differential arsenate tolerance in *Holcuslanatus*”, *Plant Physiol.*, 126(1), 299-306 (2001).
- [23] P. M. Bleeker, H. W. Hakvoort, M. Blik, E. Souer, & H. Schat, “Enhanced arsenate reduction by a CDC25like tyrosine phosphatase explains increased phytochelatin accumulation in arsenate tolerant *Holcuslanatus*”, *The Plant J.*, 45(6), 917-929 (2006).
- [24] W. Wang, B. Vinocur, O. Shoseyov, & A. Altman, “Role of plant heat-shock proteins and molecular chaperones in the abiotic stress response” *Trends Plant Sci.*, 9(5), 244-252 (2004).

- [25] Y. Sakihama, M. F. Cohen, S. C. Grace, & H. Yamasaki, “Plant phenolic antioxidant and prooxidant activities: phenolics-induced oxidative damage mediated by metals in plants” *Toxicology.*, 177(1), 67-80 (2002).
- [26] H. B. Shao, Q. J. Guo, L. Y. Chu, X. N. Zhao, Z. L. Su, Y. C. Hu, & J. F. Cheng, “Understanding molecular mechanism of higher plant plasticity under abiotic stress” *Colloids Surf. B: Biointerfaces* 54(1), 37-45 (2007).
- [27] V. Stolec, L. Li, X. Wang, X. Li, N. Su, W. Tongprasit, B. Han, Y. Xue, J. Li, M. Snyder, M. Gerstein, J. Wang, & X. W. Deng, “A pilot study of transcription unit analysis in rice using oligonucleotide tiling-path microarray” *Plant Mol. Biol.*, 59(1), 137-149 (2005).
- [28] L. Gumaelius, B. Lahner, D. E. Salt, & J. A. Banks, “Arsenic hyperaccumulation in gametophytes of *Pteris vittata*. A new model system for analysis of arsenic hyperaccumulation”, *Plant Physiol.* 136(2), 3198-3208 (2004).
- [29] L. Nussaume, S. Kanno, H. Javot, E. Marin, N. Pochon, A. Ayadi, T. M. Nakanishi, & M. C. Thibaud, “Phosphate import in plants: focus on the PHT1 transporters”, *Front. Plant Sci.*, 2, 83 (2011).
- [30] S. F. DiTusa E. B., Fontenot, R. W. Wallace, M. A. Silvers, T. N. Steele, A. H. Elnagar, K. M. Dearman, & A. P Smith, “A member of the Phosphate transporter 1 (Pht1) family from the arsenic hyperaccumulating fern *Pteris vittata* is a high affinity arsenate transporter”, *New Phytol.*, 209(2), 762-772 (2016).
- [31] W. Ali, S. V. Isayenkov, F. J. Zhao, & F. J. Maathuis, “Arsenite transport in plants”, *Cell. Mol. Life Sci.*, 66(14), 2329-2339 (2009).
- [32] G. P. Bienert, M. Thorsen, M. D. Schüssler, H. R. Nilsson, A. Wagner, , M. J. Tamás & T. P Jahn, “A subgroup of plant aquaporins facilitate the bi-directional diffusion of As (OH)<sub>3</sub> and Sb (OH)<sub>3</sub> across membranes”, *BMC Biol.*, 6(1), 1-15 (2008).
- [33] G. L. Duan, , Y. Hu, S. Schneider, J. McDermott, J. Chen, N. Sauer, B. P. Rosen, B. Daus, Z. Liu, & Y. G Zhu, “Inositol transporters AtINT2 and AtINT4 regulate arsenic accumulation in *Arabidopsis* seeds”, *Nat. Plants.*, 2(1), 1-6 (2015).
- [34] Z. He, H. Yan, Y. Chen, H. Shen, W. Xu, H. Zhang, L. Shi, , Y. G. Zhu, & M. Ma, “An aquaporin Pv TIP 4; 1 from *Pteris vittata* may mediate arsenite uptake”, *New Phytol.*, 209(2), 746-761 (2016).
- [35] J. F. Ma, N. Yamaji, N. Mitani, X. Y. Xu, Y. H. Su, S. P. McGrath, & F. J. Zhao, “Transporters of arsenite in rice and their role in arsenic accumulation in rice grain”, *Proc. Natl. Acad. Sci.*, 105(29), 9931-9935 (2008).