A Review of Pteridophyta Potential in Phytoremediation of Heavy Metal Contaminated Environments

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Abstract Pteridophyta are vascular plants (plants with xylem and phloem) that reproduce and disperse via spores. In evolution, pteridophyta lie between bryophyta and spermatophyta. Pteridophyta have unique ecological characteristics of high environmental adaptation and barren resistance. Some varieties of pteridophyta have ability of excessive absorption and accumulation of heavy metals such as arsenic (As) and antimony (Sb). Besides, pteridophyta have excellent performance in absorbing such heavy metals as cadmium (Cd), lead (Pb), copper (Cu), and nickel (Ni), and rare earth elements. In this paper, a review was made for application, mechanism, and advantages of pteridophyta in remediation of heavy metal contaminated environments, and prospect and possible research fields of pteridophyta in phytoremediation were discussed.

Key words Pteridophyta, Heavy metal contamination, As hyperaccumulators, Phytoremediation

1 Introduction

Pteridophyta can be traced back to the late Silurian Period as early as 400 million years ago[1]. Pteridophyta are main terrestrial plants during the Paleozoic Era and the Mesozoic Era, lie between bryophyta and spermatophyte in system evolution, and are most primitive and earliest vascular plants. Most existing pteridophyta are herbaceous plants and varieties are numerous[2], and ecological types are rich[3], propagation and adaptation methods are diversified[4], and pteridophyta are essential parts of plant diversity. Some varieties have high adaptation to extreme hostile environment. For example, *Hymenophyllum anguinolentum* has high resistance to drought, and can survive several days of −40 Mpa osmotic stress[5]. *Trichomanes speciosum* is widely distributed in western Europe, and it is highly barren and weak light resistant, and it even can survive in volcanic ashes[6]. There are also varieties that can survive in barren soil[7], metal abandoned mine[8], and seriously polluted areas[9]. Many pteridophyta may be used as indicator of environmental changes due to special adaptation to living environment[10-13]. In recent years, it has proved that Pteris has function of accumulating heavy metals, especially for arsenic (As) and stibium (Sb), and it has huge potential of ecological remediation.

2 Application of plants in ecological remediation of heavy metal contamination

In this paper, ecological remediation refers to phytoremediation. Since its introduction, phytoremediation has been receiving wide concern in control of heavy metal contamination. In recent years, it has been reported that there are more than 450 varieties of hyperaccumulators for heavy metals, belonging to 45 families respectively[14,15], most of them are hyperaccumulators for nickel (Ni), up to 318 varieties[16], and there are several tens of pteridophyta hyperaccumulators, mainly are As, Sb hyperaccumulators and most are *Pteridaceae*.

2.1 Resource and performance of pteridophyta that can accumulate As Arsenic is a chemical metalloid element known to cause cancer[17]. Its physical and chemical properties and environment behavior are much similar to heavy metals, so it is often included when discussing heavy metals[18]. In normal soil, ordinary plants contain As generally not higher than 3 mg/kg[19], while some pteridophyta can absorb excessive As and still can normally grow.

Pteris vittata L. is the first As hyperaccumulator found[20-22]. The As content accumulated in its above-ground part is up to 7526 mg/kg, accounting for 2.3% of biomass (dry weight) of above-ground part, and the content is even higher than phosphorus in the plant. Another As hyperaccumulator Pteris cretica has average As content in the above-ground part up to 418 mg/kg (dry weight) and the maximum As content is up to 694 mg/kg. The average As content in under-ground part (root) is up to 293 mg/kg, the maximum As content is 552 mg/kg, the bioaccumulation factor is 1.3 −4.8, and the translocation factor (ratio of As content in above-ground part to under-ground part) is 1 −2.6[23], and the As absorbed is mainly accumulated in mesophyll tissue of fronds[24,25]. Later, many As hyperaccumulators were found[26-30], and many of them are *Pteridaceae*, and some are screened from *Hemionitedaceae*. Visoottiviseth *et al.*[31] found that fronds of *Pityrogramma calomelanos* can accumulate As as high as 8350 mg /kg (dry weight). Xu Weihong *et al.*[32] measured the As content in above-ground part of *P. calomelanos* var. austroamericana up to 2 438.33 mg/ kg (dry weight). By now, domestic and foreign countries

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have found more than 20 varieties (including variants) of As hyperaccumulators, as listed in Table 1.

Table 1 As hyperaccumulator pteridophyta

No.	Taxa	Plant name (Latin)	Accumulation concentration mg/kg	Locations	Bioaccumulation factors	References
1	Variety	*Pteris aspericaulis* var. cuspigera	2410	Fronds	48.2	[26]
2	Species	*Pteris biaurita* L.	~2100	Fronds	~21	[27]
3	Species	*Pteris cretica*	418 – 694*	Above-ground part	1.3 – 6.6	[23]
4	Cultivation variety	*Pteris cretica* cv. Albo – lineata	6200 – 7600	Fronds	11.7 – 21.6	[28]
5	Variant	*Pteris cretica* var. chilsii	1358	Fronds	13.6	[29]
6	Variant	*Pteris cretica* var. crista	1506	Fronds	15	[29]
7	Variant	*Pteris cretica* var. mayii	1239	Fronds	12.4	[29]
8	Variant	*Pteris cretica* var. nervosa	1670 – 3280	Fronds	16.4 – 33.4	[26]
9	Variant	*Pteris cretica* var. parkerii	2493	Fronds	24.9	[29]
10	Variant	*Pteris cretica* var. rowerii	1425	Fronds	14.3	[29]
11	Variant	*Pteris cretica* var. wimsetti	6200 – 7600	Fronds	11.7 – 21.6	[28]
12	Species	*Pteris fauriei*	3220	Fronds	16.1	[26]
13	Species	*Pteris longifolia*	6200 – 7600	Fronds	11.7 – 21.6	[28]
14	Species	*Pteris longifolia* L.	2361	Fronds	23.6	[29]
15	Variant	*Pteris multifida* f. serrulata Miau	3650	Fronds	18.25	[26]
16	Species	*Pteris multifida* Poir	3900	Fronds	19.5	[26]
17	Species	*Pteris oshimensis*	1340	Fronds	6.7	[26]
18	Species	*Pteris quadriaurita* Retz	~2800	Fronds	~28	[27]
19	Species	*Pteris ryukyuensis* Tagawa	3647	Fronds	36.5	[27]
20	Species	*Pteris umbrosa* R. Br.	6200 – 7600	Fronds	11.7 – 21.6	[28, 30]
21	Species	*Pteris vittata* L.	1442 – 7526	Fronds	14.9 – 77.6	[20 – 22, 26]
22	Variant	*Pityrogramma calomelanos* var. austroamericana	2438.33	Above-ground part	7.5 – 18.6	[32]
23	Species	*Pityrogramma calomelanos*	8350	Fronds	4.35	[31]

Note: data with * were inferred according to block diagram in original document.

2.2 Resource and performance of pteridophyta that can accumulate Sb Antimony (Sb) is a trace element in the Earth's crust[33]. In plant body, 5 – 10 mg/kg Sb can cause toxicity[19]. At present, there are only several potential Sb hyperaccumulators[34]. Because Sb and As have similar chemical properties, while most As hyperaccumulators belong to pteridophyta, researchers screened Sb hyperaccumulators from pteridophyta. Tisarum *et al.* [35] took *Pteris vittata* L. of the United States of America, China, and Brazil as materials, measured accumulation concentration of Sb 4192 – 12000 mg/kg, and found that *Pteris vittata* L. mainly absorbed trivalent Sb. Another As hyperaccumulator *Pteris cretica* has high endurance to high concentration Sb[36]. The cultivation variant *Pteris cretica* cv. Albo-lineata can accumulate as high as 6405 mg/kg Sb[37]. However, Sb is mainly accumulated in roots, especially for *Pteris vittata* L., more than 99% of absorbed total Sb is accumulated in roots[35] (Tisarum *et al.*, 2014), and the transfer factor is extremely low. However, few researches about transfer factor have exact evidences and most are based on inference. Feng *et al.* [38] held that Sb can be transferred in many ways: trivalent Sb can be transferred through the transfer pathway of trivalent As, while the transfer pathway is still not found for pentavalent Sb, the possible transfer pathway is phosphate transfer system[35]. In angiosperm, there are also Sb accumulators, such as *Achillea ageratum*, *Plantago lanceolata*, and *Silene vulgaris* have Sb accumulation concentration up to 1367 mg/kg (basal leaves), 1150 mg/kg (roots), and 1164 mg/kg (stems)[39]. In recent years, Affholder *et al.* [40] also found that *Rosmarinus officinalis* can be cultivated in heavy metal contaminated soil, and the roots can accumulate 309 mg/kg Sb. These plants can effectively accumulate Sb, but the accumulation concentration is lower than pteridophyta. Although pteridophyta have low transfer factor of Sb, the potential accumulation of Sb is high.

2.3 Resource and performance of pteridophyta that can accumulate other heavy metals and rare earth elements Existing data indicate that pteridophyta also have high resistance and accumulation to other heavy metals apart from As. When screening hyperaccumulators and tolerant plants, Koller *et al.* [41] found that *Pteris vittata* L. can absorb much As apart from absorbing heavy metals such as Pb and Zn. Roccotiello[42] also found that *Pteris vittata* L. and *Polypodium cambricum* can absorb Zn. Li Ying *et al.* [43, 44] studied Cu absorption and transfer by *Equisetum ramosissimum* and *Pteris vittata* L., the results indicate that *Equisetum ramosissimum* and *Pteris vittata* L. have high resistance and accumulation to Cu, and the total Cu accumulation is up to 1439.47 mg/kg and 398.62 mg/kg, and the accumulation factor of root system is greater than 1, so they can be used pioneer plants to remediate Cu contaminated soil. Pteridophyta also can tolerate combined pollution of many heavy metals. *Salvinia natans* can accumulate Cr, Ni, Fe and Cd in Cr accumulated waste water[45], but it is relatively sensitive to Cd, and the semi-effect concentration is only

2.41 mg/L[46]. Although it has certain purification effect on Cd, it is extremely vulnerable to damage. In combined pollution of Cd, Pb, Mn, Cu, and Zn, roots have highest accumulation of Cd, and the accumulation factor is up to 2.3[47]. In combined pollution of Cr and Ni, *Pteridium aquilinum* can absorb higher Cr and Ni than single treatment, in other words, showing a cooperative phenomenon[12]. In addition, *Dicranopteris linearis* in rare-earth mining areas has rare earth content as high as 3263.8 mg/kg[48]. *Dicranopteris dichotoma* has high accumulation ability of rare earth elements La, Ce, and Nd, which is 100 - 1000 times higher than *Pinus massoniana* Lamb. living in the same gold mine environment, so this feature can be used for remediation of rare earth contaminated areas.

3 Accumulation and tolerance mechanism of pteridophyta to heavy metals

3.1 Excretion mechanism Some pteridophyta can excrete heavy metals or accumulate heavy metals in old leaves and discharge excessive heavy metals through falling leaves. Tu[50] (2002) found that when *P. vittata* L. grows in soil with As content higher than 0.5 mg/kg, As will accumulate in old leaves and then be discharged with leaf falling.

3.2 Binding deactivation and compartmentalization Cell wall or cell membrane or vacuole contains binding holder for binding toxic matters such as heavy metals. For example, polysaccharide aldehydic acid in pectin substance of cell wall and carboxylic acid and aldehyde group in cellulose molecules can bind heavy metals, to reduce transfer of heavy metals to cytoplasm, so as to detoxify. Besides, heavy metals entered into cytoplasm can bind organic acids (such as citric acid and malic acid), amino acid (like histidine), or metallothionein, and phytochelatin. Nishizono *et al.*[51] analyzed function of root cell wall of *Athyrium yokoscense* in detoxication of heavy metals, and results indicate that 70% - 90% of total Cu, Zn, and Cd entering into the plant body remain on cell wall, larger portion remains in the form of ion or bind to structural substances of cell wall, such as cellulose and lignin. Webb *et al.*[52] studied distribution of As in *P. vittata* L. through X ray absorption spectrometry, and results indicate that phytochelatin may play an active role in accumulation of As. Chen Tongbin *et al.*[24] found that 78% As in fronds is distributed in cell sap, and such compartmentalization[25] is an essential reason for detoxication of *Pteris vittata* L.

3.3 Anti-oxidant function After metals and metalloid elements enter into plant body, they may generate some reactive oxygen species (ROS). Excessive ROS may poison plants. To remove ROS, plants will synthesize some enzymes and non-enzyme anti-oxidant substances. The anti-oxidant substances will promote normal plant electron to smoothly transfer. According to studies of Feng Renwei *et al.*[53], under Se stress, the GSH and GR enzyme content in leaves of *P. vittata* L. have significant higher activity because GSH and GR enzyme play the role of adjusting O^{2-}; in comparison, POD, APX, and CAT remove H_2O_2 only in low concentration Se treatment. Through comparing anti-oxidant system of *P. multifida* and *P. vittata* L., Zhang *et al.*[54] found that with rise of As and Pb concentration, the activity of SOD and POD rises, GSH content increases, and thus mitigating metal toxicity.

3.4 Combined action of microorganisms Rhizospheric microorganisms can promote plants to absorb nutritional elements and heavy metals in soil, or strengthen adaptation ability to contaminated environment through secreting growth regulator and antibiotic, bacteriostatic agent or phytochelatins. In pteridophyta, researches about microorganisms strengthening absorption of contaminants focus on As, and experimental microorganisms include bacteria, fungi, and actinomyces. Rhizospheric microorganism *Pseudomonas aeruginosa* can increase absorption of *Pteridium aquilinum* to As[55], possibly because it can secrete siderophores to soil in iron deficient environment, and combine with As to form Fe and As compound, which is easily be absorbed by root system and then transferred to leaves and branches for accumulation. Other studies[56] found that with addition of allogenic bacteria Ts37, the As content of above-ground part of *P. vittata* L. is up to 837 mg/kg; with addition of actinomyce shf2, the As content of under-ground part is 427 mg/kg, which are 206% and 88% higher than the control group respectively. Liu *et al.*[57] found that after inoculation of *Glomus mosseae*, the As content of *P. vittata* L. is up to 300 mg/kg, with As accumulation increase about 43%. In the long term of evolution, pteridophyta constantly adapt to external environment, including contaminated environment. In this process, it may develop various adaptation strategies. Once under stress of contaminated environment, generally several mechanisms act jointly.

4 Strengths of pteridophyta in phytoremediation

4.1 Ancient origin and high adaptation Pteridophyta are ancient plants. With long time of natural selection and evolution, pteridophyta have developed wide adaptation and strong vitality[6]. *P. vittata* L. has strong As tolerance and can normally grow in slags with As content of 23400 mg/kg[21]; in vegetation remediation of tropical and subtropical areas, pteridophyta often become dominant species of herbaceous layer[58,59]. Thus, they can play an important role in remediation of contaminated land.

4.2 Having unique metalloid As resisting genes Pteridophyta are characterized by barren resistance, rapid growth, high breeding ability, and high resistance. They have high resistance to stressed environment and can make up for certain drawbacks and weaknesses of existing remediation plants. In recent years, there are extensive researches about As resisting genes of As hyperaccumulator pteridophyta[60-62], to find out reasons for hyperaccumulation of As from molecular mechanism. Indriolo *et al.*[63] found that *Pteris vittata* L. has gene ACR3 for encoding arsenious acid transferring protein, the expression of sporophyte root system and gametophyte is subject to adjustment of As, and it is positioned on vacuole membrane. It is interesting that this gene is lost in angiosperm. Therefore, in remediation of As contaminated soil, pteridophyta have inborn strength.

4.3 Various and poor selection of organic pollutants accumulated by pteridophyta Pteridophyta can accumulate various organic pollutants and the selection is poor[64]. Some pteridophyta have high ability of pollution tolerance and can accumulate many kinds of heavy metals. *Azolla pinnata* var. *imbricata* can grow in sewage, and its ability of resisting acid and alkali, salt and fertilizer, pH tolerance limit is 2.8 and 12.6, and the acid and alkali resistant range is pH 3.5 – 11.7, and the N and P tolerance limit is 175 mg/L and 800 mg/L[65]. *A. filiculoides* has obvious accumulation function of Cd, As, Zn[66], Pb, and Hg[67] in polluted river, can obsorb much N and P[68,69], and the removal rate of ammonium nitrogen, nitrate nitrogen and phosphorus is up to 94.04%, 95.46%, and 99.0%[70]. At the same time of accumulating Cd, *P. vittata* L. can absorb Pb, Mn, Cu, and Zn[47]; *A. imbircata* can absorb radioactive uranium, in hydroponic condition, the removal rate of uranium is up to 92% – 97%[71]; *Pteridium aquilinum* can accumulate Cr and Ni[12]. Such poor selection feature may be correlated with its evolution degree, but from the perspective of phytoremediation, such broad spectrum activity has high application value.

5 Prospects

In the process of formation and evolution of land ecosystem, pteridophyte is the pioneer population conquering land ecological environment. In modern plants, pteridophyta are diminishing in both the variety and individual quantity. Compared with spermatophyte, pteridophyte is still a small population, but as an essential member of plant kindom, pteridophyta still undertake a great role in keeping diversity of land ecosystem and ecological balance. Pteridophyta have unique biological and ecological characteristics, and have huge potential in remediation of contaminated ecosystem. In recent years, there have been extensive researches about phytoremediation mechanism of pteridophyta. In future, it may be further developed in following aspects: (i) the interaction and influencing rules of absorption and accumulation of As and Sb by pteridophyta in combined pollution condition; (ii) cultivation of new varieties easy for cultivation and remediation of heavy metal contaminated environment based on clone and characterization of existing genes; (iii) biological degradation of pteridophyta to organic pesticide; Nesterenko-Malkovskaya[72] once reported that *Eichhornia crassipes* can remove naphthalene in waste water without assistance of rhizosphere microorganisms, indicating that plant makes great contribution to degradation of organic pollutants. However, it is still not clear whether pteridophyta have such ability. (iv) study of rapid breeding technology for pteridophyta with remediation potential. Pteridophyta are not reproduced through seeds, thus, high efficient breeding of pteridophyte seedlings may be a bottleneck in remediation. Although there have been some breeding methods[73,74], the survival rate is limited and it is difficult to satisfy demands of large field planting. (v) research and development of final risk control and recovery and treatment technologies for accumulators. After pteridophyte accumulating heavy metals, if control is not taken, it may pose ecological risks. Mathews *et al.*[75] and Jeong *et al.*[76] found that in planting areas of As accumulating pteridophyta, herbivores contain higher As in body than the control group, even up to wounding effect. If effective control is not taken, it probably leads to ecological imbalance. If treating by traditional methods such as landfill or burning, it may lead to secondary pollution. Therefore, research and development of high efficient and safe resource based utilization will have broad prospects.

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mental management requirements, business requirements, and macro control requirements of all levels of environmental departments, we developed a new data acquisition and transmission system. It solves the problem of non-standardized communication protocols and difficult interaction of communication information for existing pollution source automatic monitoring equipment, and integrates a variety of on-line monitoring system communication protocols. Besides, it can automatically convert various protocols to information format required by the national standard, allow access of many types of environmental element monitoring equipment, support a variety of data transmission modes, support multi-protocol multi-target parallel data communication, support reverse control, display operating conditions and processes of equipment, support data reissue, and conform to national environmental protection standard.

Our research can provide high performance intelligent data acquisition and transmission equipment for data acquisition and monitoring of environmental protection automatic monitoring equipment, play a great role in accurate and reliable grasp of various monitoring parameters, greatly reduce cost of monitoring system, assist enterprises and levels of environmental protection departments in realizing total amount control and emission reduction target, satisfy increasing requirements of levels of environmental management and competent authorities for monitoring data, and it will greatly promote building of conservation, environmental protection and harmonious society.

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