
*Changes in Biochemical Constituents in response to
Arsenic-induced Stress in *Pteris vittata* and
Eichhornia crassipes to determine Stress Tolerance
– A Review

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Abstract

Large areas of West Bengal have to rely on arsenic-contaminated ground-water for irrigation of staple crops such as rice. **Phytoremediation** is the use of plants to remove or render contaminants harmless in the ecosystem while **Phytoextraction** is the use of plants, preferably **hyperaccumulators**, to take up contaminants. In this work, *Pteris vittata*, exhibited remarkable ability to hyperaccumulate arsenic in the fronds as compared to *Eichhornia*. While arsenic toxicity resulted in significantly diminished biochemical constituents in *Eichhornia*, *Pteris* was comparatively less affected. This indicates that *P.vittata* is better equipped to tolerate arsenic stress than *Eichhornia* and is more suitable for arsenic phytoremediation.

Key Words: Arsenic stress, Phytoremediation, Phytoextraction, hyperaccumulator, *Pteris vittata*, *Eichhornia crassipes*, Stress tolerance, Biochemical constituents, Stress Physiology.

1. Introduction

Arsenic is ubiquitous in the environment. Arsenic contamination in soils often leads to groundwater contamination and arsenic toxicity in plants, animals and humans. Remediation of arsenic-contaminated soils has become a major environmental issue. Elevation of arsenic

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levels in soils causes considerable concern with respect to plant uptake and subsequent entry into wildlife and human food chains. Arsenic speciation in the environment is complex, existing in both inorganic and organic forms, with interconversion between species regulated by biotic and abiotic processes.

Arsenic in the environment is often associated with other elements (Au, Ag, Cu and Sn in particular), and mining and processing of these ores has led to extensive arsenic pollution of mining regions throughout the world.¹ Historically, the use of arsenic-based pesticides has led to considerable contamination of domestic and agricultural land, through their use as lawn herbicides, and insecticides for rice, orchards and cotton.²

The environmental fate and behaviour of arsenic has received increasing attention due to a crisis in South-East Asia (West Bengal, Bangladesh and Vietnam). Tens of millions of people have been exposed to high levels of arsenic in ground-water, the region's primary source of drinking-water.³ Large areas of Bangladesh, West Bengal and Vietnam have to rely on arsenic-contaminated ground-water for irrigation of staple crops such as rice.^{3,4} Consequently, in addition to exposure through drinking-water, people are being exposed to arsenic through ingestion of vegetation which has been contaminated by irrigation with arsenic-contaminated water, and from livestock and their products, where livestock have been fed on arsenic-contaminated vegetation.

2. Phytoremediation

Phytoremediation is the use of plants to remove or render contaminants harmless in the ecosystem. Phytoremediation actually includes several methods, such as phytovolatilization, phytostabilization and phytoextraction. **Phytovolatilization** refers to the uptake, translocation and volatilization of contaminants from plants. The contaminants may or may not be transformed during this process. **Phytostabilization** employs plants in order to contain contaminants in the soil, preventing migration of the contaminant off site. **Phytoextraction** is the use of plants, preferably **hyperaccumulators**, to take up contaminants. Subsequently, the plants are harvested, transported and disposed off site. Phytoextraction has become increasingly popular because of its low cost as compared to more traditional remediation technologies. The costs involved in phytoremediation may include planting, maintenance, harvesting and disposal of plant biomass. The volume and mass of the plant disposal are significantly less than the disposal of soil when excavation is required. However, because phytoextraction is dependent on the plant, conditions at the site must be able to maintain plant production, and the contaminant must be accessible to the roots for uptake. In addition, soils with very high contaminant concentrations may inhibit plant growth and/or significantly prolong the amount of time required for remediation.⁵ Much research is still required to ensure proper employment and utilization of phytoextraction.

In 2001, University of Florida researcher Lena Ma discovered that the **Chinese brake fern** (*Pteris vittata*) grows well in arsenic-contaminated soil.⁶ This plant accumulates large amounts of arsenic in its fronds, the portion of the fern that is above ground. This discovery led to the idea that brake fern could help clean up arsenic-contaminated soil. Phytoremediation thus offers an environmentally-friendly and cost-effective method

to remove arsenic from contaminated soil. In addition to brake fern, other plants have been found to be useful in phytoremediation. Sunflowers were very effective at removing radioactive materials from the water near the site of the 1986 nuclear-power-plant disaster at Chernobyl, Ukraine. Poplar trees are also useful in removing a wide range of pollutants from soil and have been used widely for this purpose.

The term "**hyperaccumulator**" describes a number of plants that belong to distantly related families, but share the ability to grow on metalliferous soils and to accumulate extraordinarily high amounts of heavy metals in the aerial organs, far in excess of the levels found in the majority of species, without suffering phytotoxic effects. Three basic hallmarks distinguish hyperaccumulators from related non-hyperaccumulating taxa: a strongly enhanced rate of heavy metal uptake, a faster root-to-shoot translocation and a greater ability to detoxify and sequester heavy metals in leaves.⁷

3. Objective

The existing threat of **arsenic pollution in West Bengal** and the pressing need to look for an **eco-friendly solution** based on green technology i.e. **Phytoremediation** is the origin of this work. The biochemical profile *i.e.* studies on different **Biochemical** parameters *viz.* constituents in *Pteris* and *Eichhornia* will reveal the impact of arsenic stress on the physiology and metabolism of these two plants *pre-* and *post* exposure and their possible use in phytoextraction of arsenic-contaminated soils.

4. Reasons for selecting *Pteris* and *Eichhornia*

Pteris vittata produces a relatively high biomass, it is a fast-growing plant. It is a perennial and is commonly found growing almost everywhere in Kolkata and Howrah. It is also tolerant of full

sun, unlike many other ferns, but it also grows well under shady conditions. *Pteris vittata* prefers an alkaline soil environment. This can contribute to its ability of arsenic-hyperaccumulation because, in general, arsenic is more available at a higher pH. *Pteris vittata* is also able to take up many forms of arsenic.⁸ *Eichhornia crassipes* (Mart.) Solms. is native to Brazil. Plants are thought to have been first introduced into the United States at the 1884 Cotton States Exposition in New Orleans, LA. Water hyacinth is a floating, flowering, perennial weed (requiring a wet habitat), form dense rafts in the water and mud. The large, robust plants of water hyacinth are often referred to a 'bull hyacinths'. It grows faster than any other tested plant. Water hyacinth is an aquatic plant which can flourish and reproduce floating freely on the surface of water or it can also be anchored in mud. It is of common occurrence in water bodies and is known to bioaccumulate heavy metals.

5. Discussion

The Brake fern, *Pteris vittata*, has the remarkable ability to hyperaccumulate arsenic in its shoots, with shoot concentrations reaching levels ~100-fold higher than soil concentrations.⁶ This ratio is held for uncontaminated soils (6 mg kg⁻¹ As) and highly contaminated soils (1500 mg kg⁻¹ As). The fern is capable of taking up a range of inorganic and organic arsenic species including arsenate, arsenite and MMA, with up to 93% of the arsenic concentrated in the fronds.⁶ This capability to hyperaccumulate arsenic is, to date, unique. It has been shown that *P. vittata* is efficient in taking up arsenic from contaminated soils. In a greenhouse study, in 2002 it was reported that *P. vittata* extracted up to 38 mg As/plant after 20 weeks of plant growth, translating to a 25% reduction in soil arsenic.⁸

In another work in 2014 most of the arsenic concentration was reported in the fronds (*Pteris*) or leaves (*Eichhornia*).⁹ Hence further work was carried out with frond and leaf samples. *Pteris vittata* and *Eichhornia crassipes* used for the biochemical experiments were grown under controlled laboratory conditions to completely eliminate the possibility of any previous contamination and were of identical age. The selected concentrations of arsenic were 0, 133 and 267 μ M and the plants were harvested at three intervals, i.e. 1, 5 and 10 days after arsenic treatment. From the results obtained *Pteris vittata* seems to be more effective in arsenic uptake than *Eichhornia*. Arsenic concentration was high in 10 day old samples of *P. vittata* as compared to *Eichhornia* which showed relatively lower uptake. While *Eichhornia* showed significantly diminished concentrations of chlorophyll, carotenoids, protein, amino acid, nitrogen, total carbohydrates and total phenols even in low concentrations of arsenic, *Pteris* appeared to be comparatively less affected. The comparatively higher starch content in *Eichhornia* under arsenic stress is perhaps an adaptation for stress - combating. This indicates that *P.vittata* is better equipped to tolerate arsenic stress than *Eichhornia* and is more suitable for phytoremediation of arsenic.^{9,10}

Arsenic is known to induce oxidative stress in plants by generating various ROS¹¹ resulting in a range of responses in plants, including readjustment of transport and metabolic processes and growth inhibition.¹² Like other environmental stress, arsenic may create conditions in the thylakoids where the energy level exceeds the amount that can be dissipated by the metabolic pathways of the chloroplast.¹³ As a consequence, the electron transport processes in the thylakoid membranes are impeded and toxic symptoms develop. Several toxic intermediates (superoxide anion, hydroxyl radicals, hydrogen peroxide and lipid peroxide) are generated in the cell wall region as well as inside the cell during this process, which affects membrane permeability, enzyme activity, metabolic pool, photosynthetic activity, plant biomass and leaf chlorosis and necrosis.^{14,15}

Plants often contain trace concentrations of many contaminants of concern. At low levels, plants can usually metabolize or dispose of these compounds without any significant injury. Generally, at high contaminant concentrations in soil or water, plants often suffer (like *Eichhornia*, in the earlier study)^{9,10} and/or die because of their inability to metabolize these harmful elements. However, some plants can, (like *Pteris*, in the earlier study)^{9,10} survive and/or thrive when they accumulate high concentrations of toxic elements.

From the results obtained so far, *Pteris vittata* appears to be more suitable than *Eichhornia crassipes* for its possible use in phytoextraction of arsenic from arsenic-contaminated soils. The comparative study reveals that *Pteris vittata* is the more efficient plant in combating and tolerating arsenic stress.

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