Arsenic speciation and transport in the rhizosphere of an arsenic-hyperaccumulating fern

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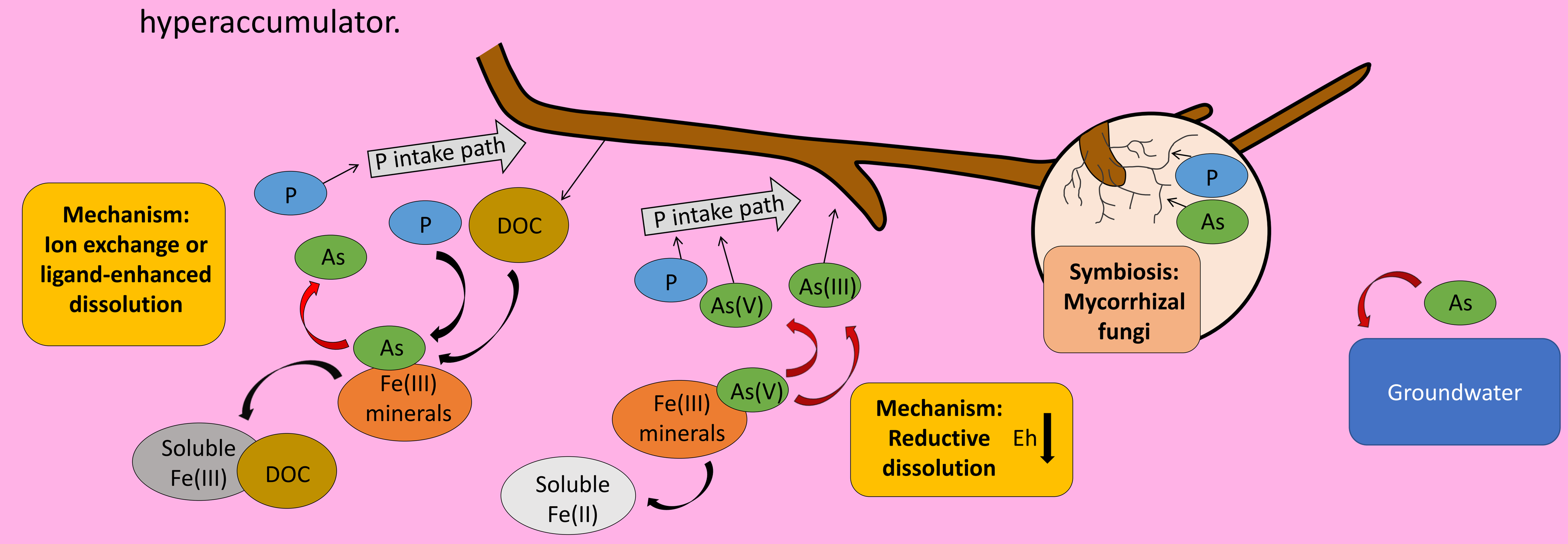
MOTIVATION

- **Arsenic phytoextraction** could be a **sustainable** way to clean up arsenic-contaminated soils, but **remediation rates are slow.**
- **Fertilizer application and mycorrhizal fungi** inoculation could **increase arsenic uptake** in the fern, but could also **increase** arsenic **leaching.**
- Understanding the extent to which **arsenic is mobilized from bulk *vs.* rhizosphere soil** (which affects how much soil is remediated), **and how arsenic is released from soil solids into solution** (which could be affected by nutrient application) is crucial to **optimize remediation** strategies.

Pteris vittata, the brake fern, is an arsenic hyperaccumulator.

In a soil column study, we determined the effects of (i) phosphorus (P) and (ii) mycorrhizal fungi inoculant addition to soil on arsenic uptake and leaching in the *P. vittata* soil-plant system.

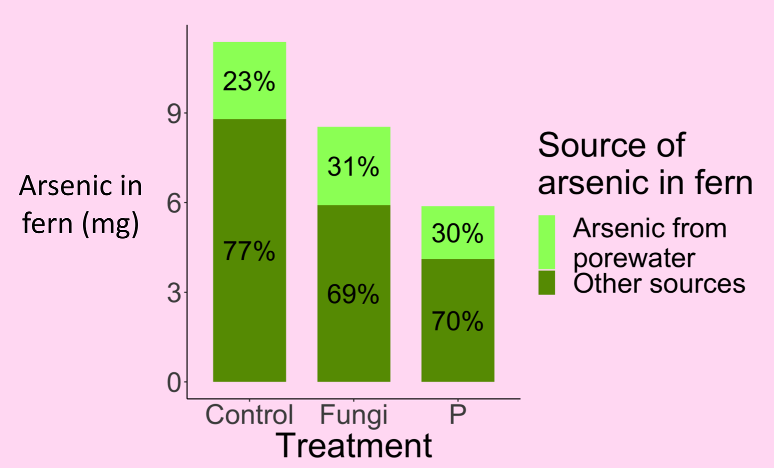


Rhizosphere processes could be important in release of arsenic from soil for uptake into the fern.

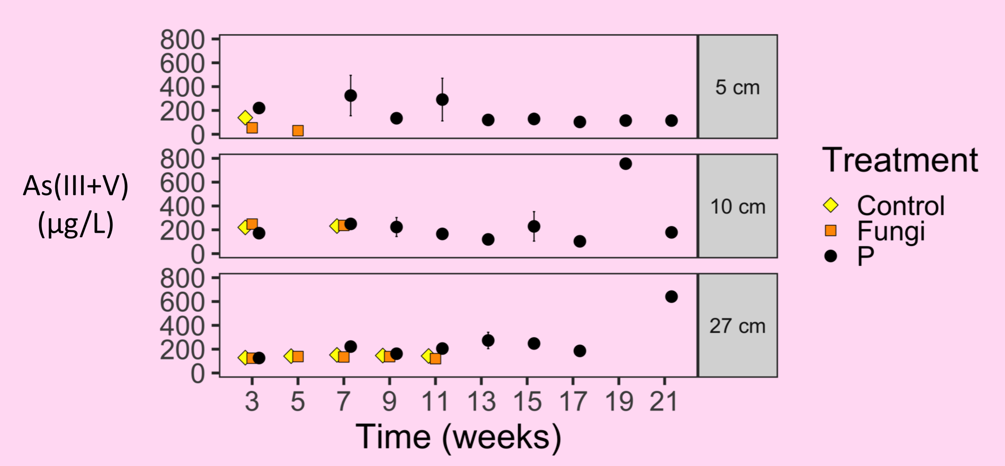
FINDINGS

- Porewater arsenic does not supply the bulk of arsenic in the fern, suggesting **only rhizosphere soil is phytoextracted.**
- **Phosphorus addition decreases arsenic uptake** into the fern, but **increases arsenic leaching** from soil.
- **Arsenic and iron** are primarily **oxidized in rhizosphere and bulk soil**, suggesting **arsenic is mobilized through ion exchange** with phosphorus or dissolved organic carbon, and not through reductive dissolution.
- **Even under oxic conditions leaching occurs**, potentially causing environmental contamination.
- **Recommendation: Avoid excessive phosphorus** to promote healthy fern growth, increase transpiration, increase arsenic uptake, and decrease leaching.

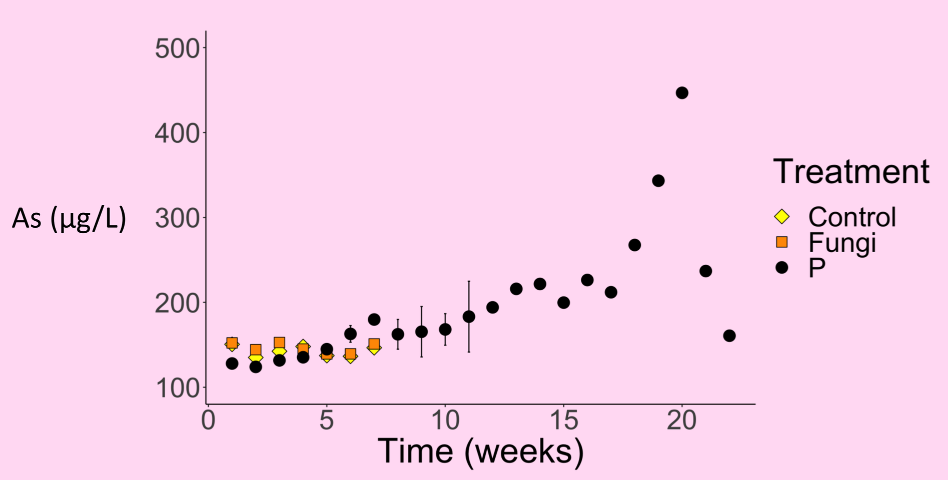
ARSENIC UPTAKE AND LEACHING

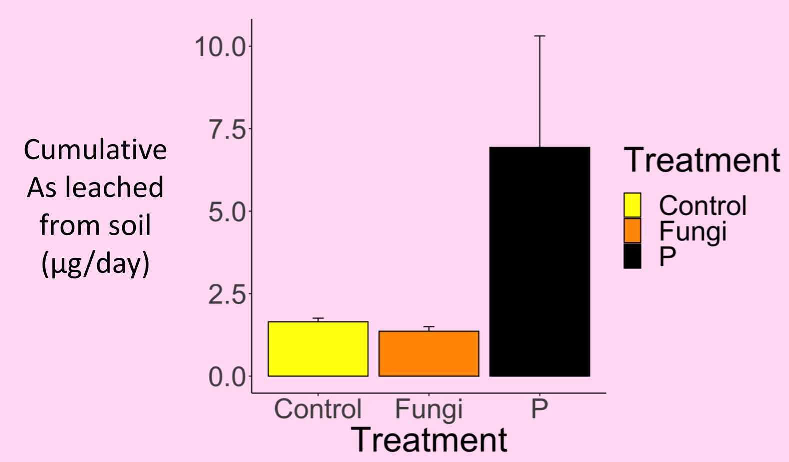


Only about 30% of arsenic in fern is contributed from bulk soil *via* transport in porewater.



Concentrations of arsenic (primarily arsenic(V)) in porewater increase with depth.



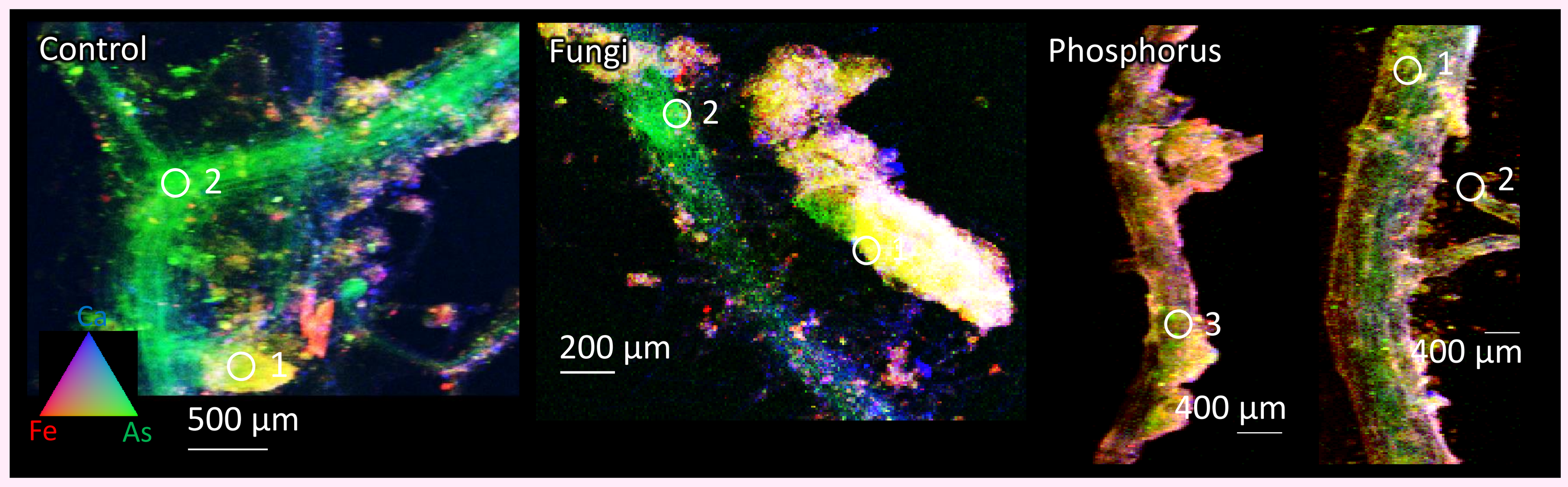


Phosphorus addition increases both effluent arsenic concentrations and volume, leading to increased cumulative leaching. Note: Effluent stops in Control and Fungi-treated columns after 7 weeks when transpiration exceeds water application.

SPECIATION

Linear combination fits of µXANES spectra from **whole roots** and **root longitudinal thin sections** show mainly oxidized arsenic on soil grains even directly adjacent to root surface.
Microprobe work reveals primarily reduced arsenic at the surface of control and fungi-inoculated roots. Less reduction is found on phosphorus-treated roots, or in control and phosphorus bulk root spectra.
Soil aggregates show increased arsenic(III) in the wetter phosphorus-treated column, compared to the drier control column, regardless of distance from aggregate surface (to right side of images).

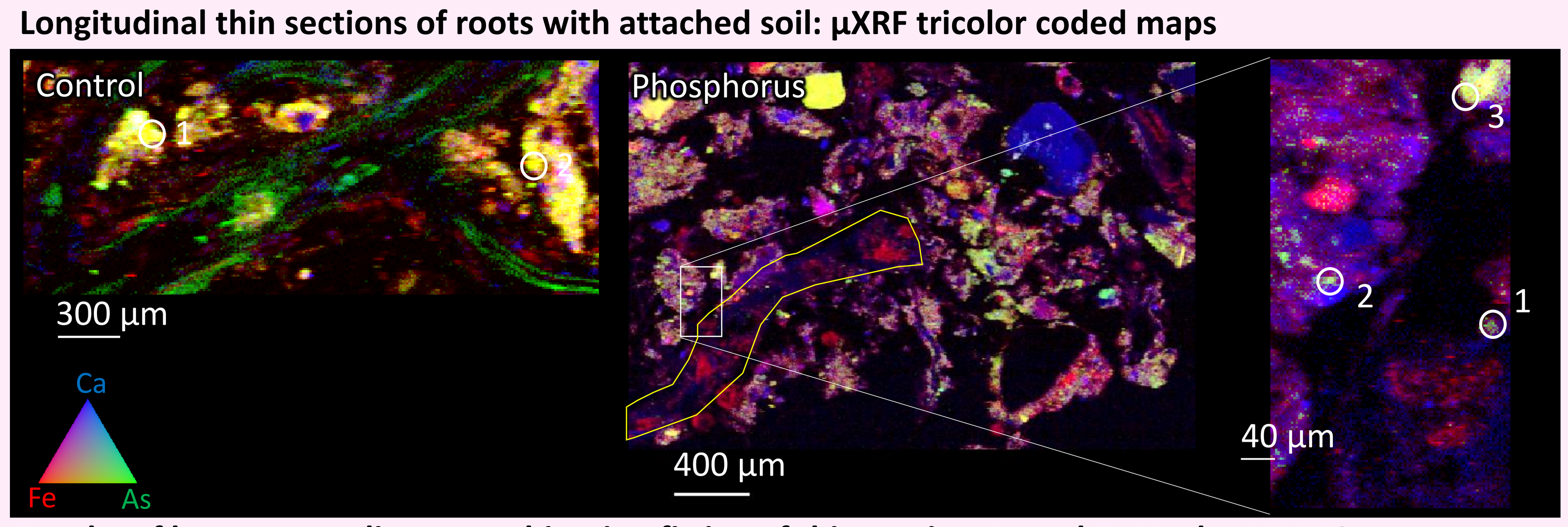
Whole roots: µXRF tricolor coded maps



Results of least-square linear combination fitting of whole root As and Fe K-edge XANES spectra

Spot	Arsenic		Iron					
	(III) %	(V) %	Component 1	%	Component 2	%	Component 3	%
Control								
1	15	88	Iron (III) silicate	43	Iron (II) sulfate	29	Iron (II, III) silicate	23
2	80	20	Iron (II, III) silicate	57	Biogenic iron oxide	43		
Fungi								
1	13	94	Iron (III) silicate	48	Iron (II) silicate	28	Iron oxide	27
2	100	0	Iron (III) silicate	76	Iron (II) sulfate	15	Iron (II,III) silicate	12
Phosphorus								
1	45	55	Iron (III) silicate	73	Iron (II, III) silicate	20	Iron (II) silicate	7
2	21	80	NA					
3	10	90	Iron (III) silicate	68	Iron (II, III) silicate	17	Iron (II) silicate	15

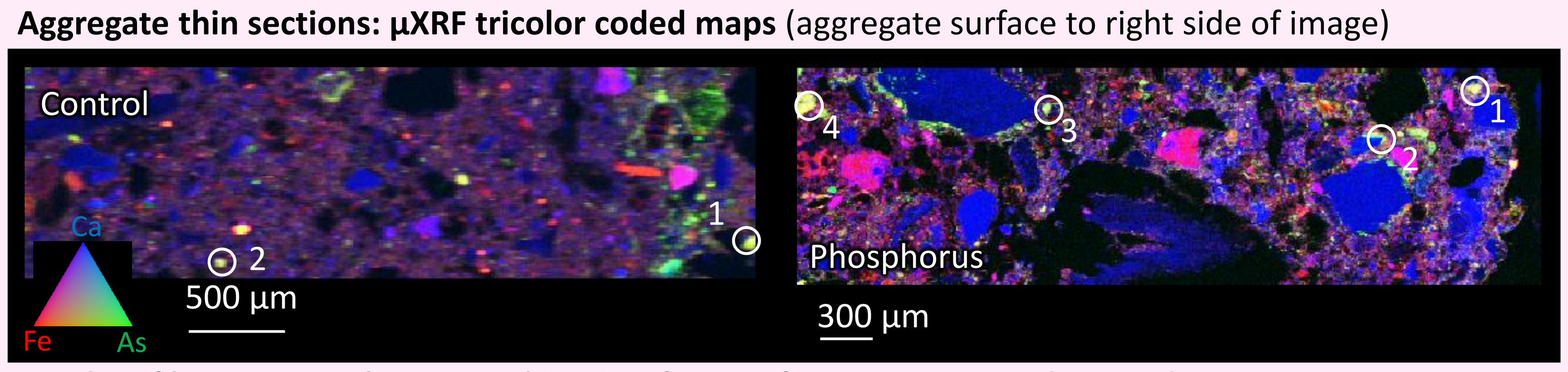
Longitudinal thin sections of roots with attached soil: µXRF tricolor coded maps



Results of least-square linear combination fitting of thin section As and Fe K-edge XANES spectra

Spot	Arsenic		Iron					
	(III) %	(V) %	Component 1	%	Component 2	%	Component 3	%
Control								
1	17	76	Iron(II) silicate	60	Iron (II, III) silicate	39	Iron (III) silicate	8
2	0	100	Iron (III) silicate	73	Iron (II) sulfate	24	Iron (II, III) silicate	9
Phosphorus								
1	9	88	Biogenic iron oxide	65	Iron (II) silicate	40		
2	34	78	NA					
3	21	81	NA					

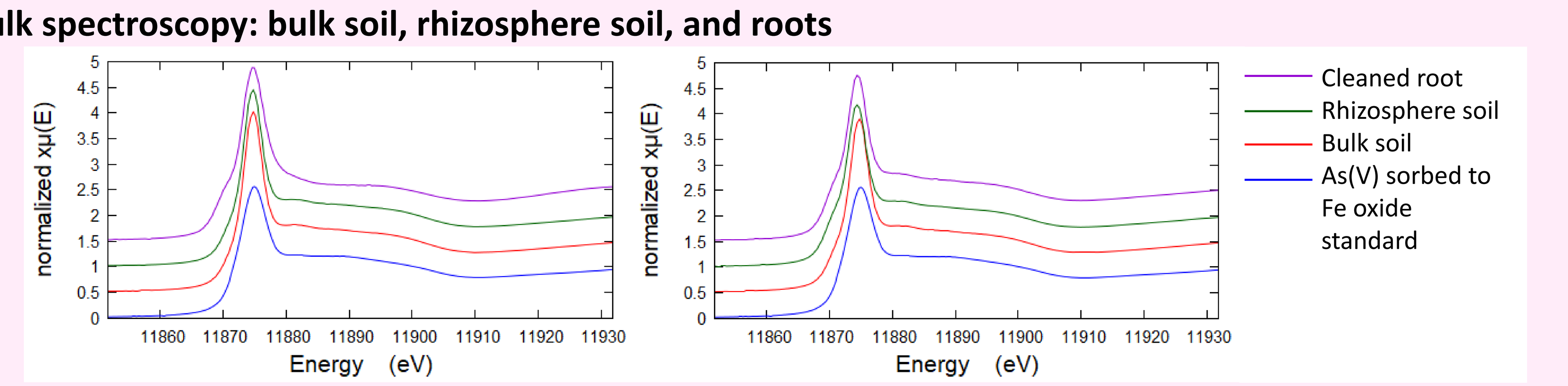
Aggregate thin sections: µXRF tricolor coded maps (aggregate surface to right side of image)



Results of least-square linear combination fitting of aggregate As and Fe K-edge XANES spectra

Spot	Arsenic		Iron					
	(III) %	(V) %	Component 1	%	Component 2	%	Component 3	%
Control								
1	0	100	Iron (III) silicate	56	Iron oxyhydroxide	19	Iron (II) silicate	22
2	0	100	Iron (III) silicate	40	Iron oxide	33	Iron (II) silicate	25
Phosphorus								
1	25	91	Iron oxyhydroxide	58	Iron (II) sulfate	35	Iron (II, III) silicate	11
2	0	100	Biogenic iron oxide	99	Iron (II) silicate	9		
3	60	42	Iron (III) silicate	52	Iron (II) sulfate	40	Iron (II, III) silicate	11
4	14	83	Iron (III) silicate	67	Iron (II) silicate	20		

Bulk spectroscopy: bulk soil, rhizosphere soil, and roots



METHODS

Soil column study: *P. vittata* was planted in columns packed with sandy loam soil historically contaminated with arsenic (114 mg/kg). Soil was treated either with phosphorus (1.2 g/kg) or *Funneliformis mosseae* mycorrhizal fungi inoculant, with 3 replicates per treatment. Over 22 weeks, synthetic rain was eluted through columns. Arsenic (As), iron (Fe), phosphorus (P), and dissolved organic carbon were tracked during growth in soil porewater, column effluent, and plant tissue.
Root and soil samples for microprobe and bulk XAS measurements were collected from soil 10 cm deep and placed immediately on ice. Whole root samples were frozen in liquid nitrogen, and samples for thin sections and bulk spectroscopy were returned to the lab and dried in an anaerobic chamber. Roots (with attached soil) and aggregates for microprobe work were embedded in EPO-TEK 301 epoxy and prepared as thin sections with roots sliced longitudinally. For bulk XAS, rhizosphere soil attached to roots was carefully removed with a small brush. Aggregates with no visible fern roots were considered bulk soil. Bulk soil, rhizosphere soil, and roots were ground, mounted on a filter, and dried.

Microprobe: µXRF elemental mapping and µXANES were performed at ALS beamline 10.3.2. Whole roots and thin sections were mounted with silicon thermal grease onto a Peltier stage. Data were collected in fluorescence mode at -20°C at ambient pressure using a Canberra 7-element Ge solid state detector or Amptek single element fluorescence detector. For low-As spots, XANES were collected at 2 adjacent spots and merged. Spectra were deadtime corrected, calibrated, deglitched, pre-edge background subtracted and post-edge normalized using a suite of LabVIEW software available at the beamline. The spectra were subsequently least-square linear combination fitted (LCF) using a XAS database of As and Fe standards. All spectra were calibrated using the main peak of a Na arsenate standard set at 11875 eV, or Fe foil standard set to 7110.75 eV.

Bulk XAS: Bulk XANES spectra were collected at SSRL beamline 7.3. Filter membranes were sealed and attached to sample holders with Kapton tape. Data were collected in fluorescence mode using a cryostat sample holder at 10 K and a Canberra 30 element Ge solid state detector. Spectra were calibrated to a reference Au foil (E0 = 11919 eV) or Fe foil (E0 = 7110) in SixPak (Webb 2005), and normalized and plotted in Athena.

B. Ravel and M. Newville, *ATHENA, ARTEMIS, HEPHAESTUS: data analysis for X-ray absorption spectroscopy using IFEFFIT*, Journal of Synchrotron Radiation **12**, 537–541 (2005).
S. M. Webb, *SIXpack: A graphical user interface for XAS analysis using IFEFFIT,* Phys. Scr. T115, 1011 (2005).

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