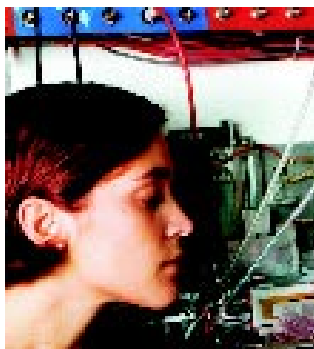


## X-ray Imaging of Zinc Accumulation in Bioremediators

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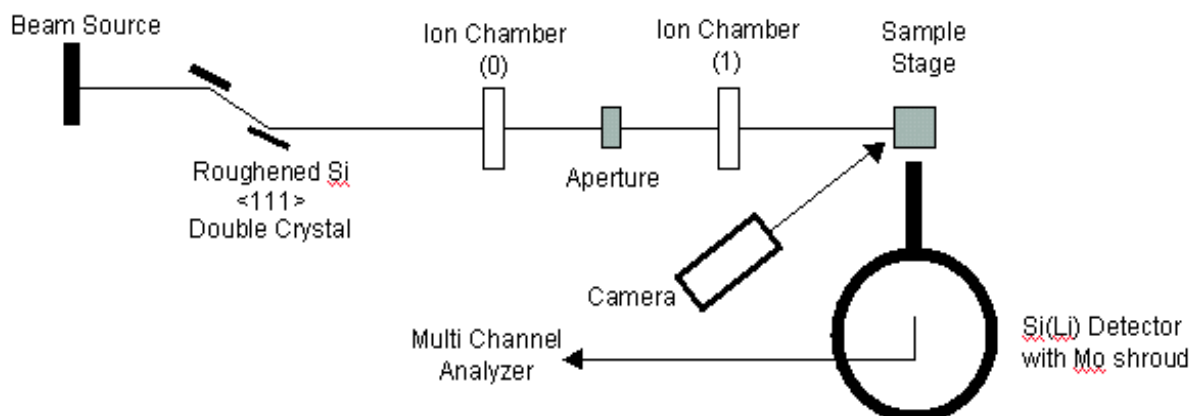


Megan Toaspern

Soils contaminated with toxic metals are a growing and challenging environmental cleanup problem. Traditional methods of stripping away the contaminated soil and trucking it off to a landfill only results in millions of tons of soil being moved without eliminating the contamination. The use of plants to clean up heavy metal and radioisotopes has been focused on as an efficient and affordable form of bioremediation. Plants termed phytoextractors are capable of absorbing large amounts of heavy metals from the soil, and accumulating these metals in plant tissue. The plants can then be harvested and incinerated, minimizing the waste products.

Finding and understanding these plants is a goal of the Cornell USDA Lab, directed by Leon Kochian. One of the best known heavy metal hyperaccumulators is *Thlaspi caerulescens*, a member of the cabbage family which has been shown to accumulate up to 3% Zn on a dry weight basis without showing toxicity symptoms. X-ray fluorescence and absorbance spectroscopy (XANES) measurements at CHESS have been used to investigate Zn hyperaccumulation in *T. caerulescens*.

Using a pinhole aperture on the B-2 station at CHESS (Figure 1), a small beam spot was rastered over the surface of a leaf sample. A 10 KeV X-ray beam excited Zn fluorescence in the sample, and was measured with a Si(Li) detector. Elemental distribution maps were then reconstructed from the point by point data.



**Figure 1:** A schematic diagram of the experimental set-up in the B-2 hutch at the CHESS lab. All images were generated using this equipment.

Shown in the X-ray fluorescence images in Fig 2, the pattern of Zn accumulation in leaves was different in leaves of *T. caerulescens* compared with the related nonaccumulator species, *Thlaspi arvense*. For plants grown in a range of Zn concentrations, very little Zn fluorescence was emitted from leaves of the nonaccumulator species and this signal was associated exclusively with the leaf vascular tissue. In the Zn-hyperaccumulator *T. caerulescens*, the pattern of Zn accumulation was dependent upon the amount of Zn stored in the leaves. After growth on low Zn levels, most of the Zn

was localized in the vascular tissue.

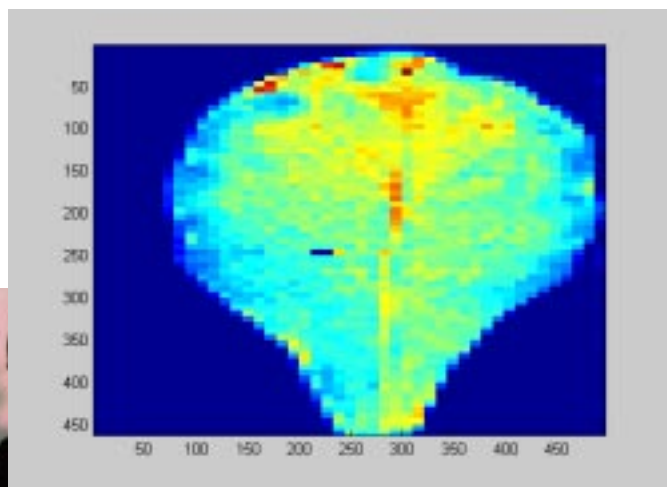
As Zn accumulation in leaves increased, Zn fluorescence increased from leaf tissue between the veins, indicating that the excess Zn is stored inside living leaf cells. These results are important because they suggest that Zn hyperaccumulation in *T. caerulescens* involves alterations in Zn transport systems and chemical speciation in leaves. Studies have been initiated to characterize the chemical environment associated with Zn accumulation in leaf cells

and thereby determine what organic ligands might be complexed with Zn.

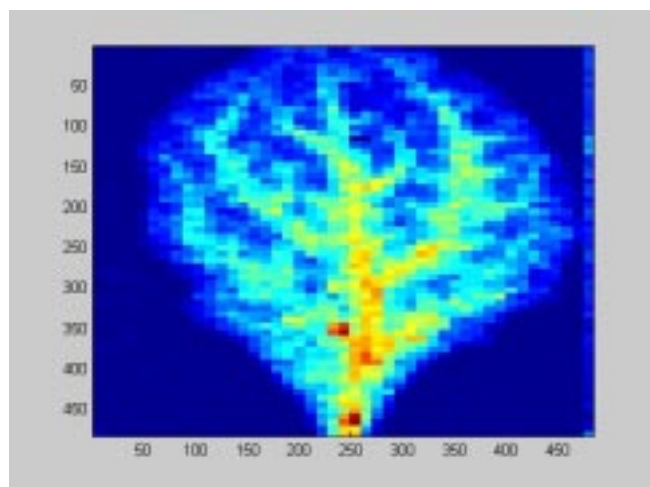
Further areas of study may include studying *Thlaspi caerulescens* grown in a range of zinc concentrations to determine the efficiency of the plant as a phytoremediator in lower zinc levels. Also, since the technique of X-ray fluorescence is non-destructive, it would be possible to look at one plant leaf over a period of time to study time-dependent uptake. One other area of study would be to analyze the roots of the two species of *Thlaspi* vs. the shoots of both species. The nonaccumulator should have a higher concentration of zinc in the roots, while the hyperaccumulator should have a higher concentration of zinc in the shoots.

These questions may be addressed with further use of X-ray imaging. The mechanisms of heavy metal transport, specifically the translocation in the xylem (and possibly phloem), transport across the leaf-cell plasma membrane and storage in the leaf-cell vacuole can be further studied with higher beam resolution. Currently, it is possible to achieve beam sizes as low as 10 microns. With smaller beam sizes to create high resolution scans, it is possible to study zinc in relation to cell physiology as well as complex ligand structures.

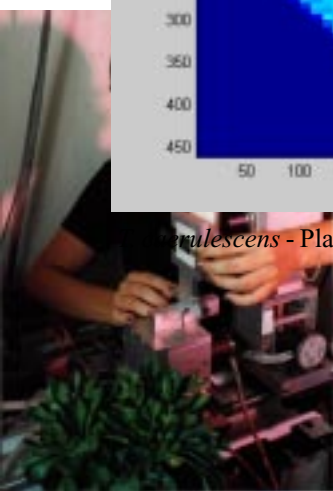
Future research can benefit greatly by integrating the use of X-ray imaging. This innovative tool offers scientists a versatile tool in performing research. The use of synchrotron technology can lead to major advances in the field of biology and a new multi-disciplinary field of study.



*Thlaspi caerulescens* - Plant likes zinc



*Thlaspi arvense* - Plant hates zinc



X-ray fluorescence images where blue = low zinc concentration and yellow = high zinc concentration. Food for thought: how can the *T. caerulescens* plant successfully move zinc through the stem and out to the outer biomass of the leaf? In the case of the *Thlaspi arvense* plant, the zinc becomes clogged in the veins and the plant dies at concentrations that *T. caerulescens* thrives upon?

Megan Toaspern was a Research for Undergraduates (REU) student from Gettysburg College, PA. Her work was featured at the National Science Foundation exhibit at the special 100th Anniversary celebration of the American Society in Atlanta, Georgia in 1999.