

Novel use of laser technology reveals interactions between roots, soil organisms

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A good example of cross-sectional images yielded by laser ablation tomography. These show a corn root with a western corn rootworm and damage resulting from this herbivore. Using RGB spectra measured from these images, researchers can



separate the rootworm from root tissue, and assess damaged regions from intact tissue. Credit: Christopher Strock/Penn State

A novel use of a custom laser system—developed in Penn State's College of Agricultural Sciences eight years ago—allows researchers to see how soil organisms affect plant roots. The discovery has implications for future breeding of more resilient and productive crops, according to an international team of scientists.

"This research shows how we can use laser ablation tomography—often referred to as LAT—to visualize the anatomy of roots from several crop species, and see how soil organisms such as fungi, herbivorous nematodes and insects interact with these roots in three dimensions," said Jonathan Lynch, distinguished professor of plant science.

Lynch's research group developed the unique technology in 2011 for other root-analysis applications in collaboration with then-undergraduate student Ted Reutzel, now with Penn State's Applied Research Laboratory, and current LAT entrepreneur Ben Hall of Lasers for Innovative Solutions (L4IS). Lynch explained that researchers using LAT can measure the light spectra given off by different cells cut by the laser to differentiate between various tissues. This differentiation is based on the cells' chemical composition.

"LAT not only can provide us with a novel perspective of interactions among roots and soil organisms, but we also are able to process many root samples in a short period of time with this technology," he said. "That high throughput rate addresses a major limitation for other researchers who are interested in conducting <u>genetic studies</u> and running breeding programs to develop crops that are more resistant to soil pathogens."





Schematic of the laser ablation tomography system: The pulsed ultraviolet laser is directed into beam-shaping optics to transform the beam into an oscillating beam, creating a cutting sheet. The root sample is moved into the beam path on a motorized linear stage as a camera with a macro lens simultaneously images the exposed anatomy sliced by the beam. Credit: Penn State

Soil biota have important effects on crop productivity, but they are difficult to study, noted researcher Christopher Strock, a postdoctoral scholar in the Lynch lab who spearheaded the project. He explained that laser ablation tomography allows for rapid, three-dimensional quantitative and qualitative analysis of root anatomy, providing new opportunities to investigate interactions between roots and soil organisms.



In the study, recently published in the *Journal of Experimental Botany*, researchers used LAT for analysis of corn roots colonized by arbuscular mycorrhizal fungi, corn roots fed on by western corn rootworm, barley roots parasitized by cereal cyst nematode, and common bean roots damaged by Fusarium fungi.

Ultraviolet excitation of root tissues affected by soil organisms resulted in differing natural fluorescence signals that allow the researchers to classify tissues and anatomical features, Strock pointed out. The system displayed samples in three dimensions, enabling quantification of the volume and distribution of fungal colonization, western corn rootworm damage, nematode feeding sites, and tissue compromised by Fusarium.





The camera with a macro lens on the LAT system captures amazing photos. In the inset, a root segment has been inserted into the machine. It is moved into the beam path on a motorized stage where the camera images the exposed anatomy sliced by the laser. Credit: Christopher Strock/Penn State





Cross-sectional images of corn root segments taken using laser ablation tomography. Arbuscular mycorrhizae (red) and root cortical aerenchyma (green) were defined and segmented from root cross sections. Credit: Christopher Strock/Penn State





This image is a three-dimensional reconstruction from laser ablation tomography showing the western corn rootworm feeding on a corn crown root. The larva is displayed as white and longitudinal (c) and transverse (d) cross sections highlight the anatomical features of the larva. Credit: Christopher Strock/Penn State

"Because of its capability to process many root samples quickly, LAT serves as an excellent tool to conduct large, quantitative screenings to characterize genetic control of root anatomy and interactions with soil organisms," Strock said. "This technology improves interpretation of root-organism interactions in relatively large, opaque root segments, providing opportunities for research investigating the effects of root



anatomical traits on associations with soil organisms."

Laser ablation tomography is conducted at only two other locations in the world: at Lasers for Innovative Solutions (L4IS) in State College, Pennsylvania, run by Hall—an alumnus of the Department of Plant Science who co-invented the technology with Lynch and Reutzel; and at the U.K.'s University of Nottingham, which is partnering with Penn State on plant science initiatives. The Lynch research group is helping to get a new LAT system up and running at Nottingham.

"The ultimate goal of my lab is to conduct research that leads to the development of crops with better growth under limited water and nutrient availability, and LAT is an important tool we have to accomplish that," Lynch said. "By demonstrating the versatility of the technology, we pave the way for other scientists to use it to address questions about roots and <u>soil</u> interaction at the anatomical level."

Provided by Pennsylvania State University

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