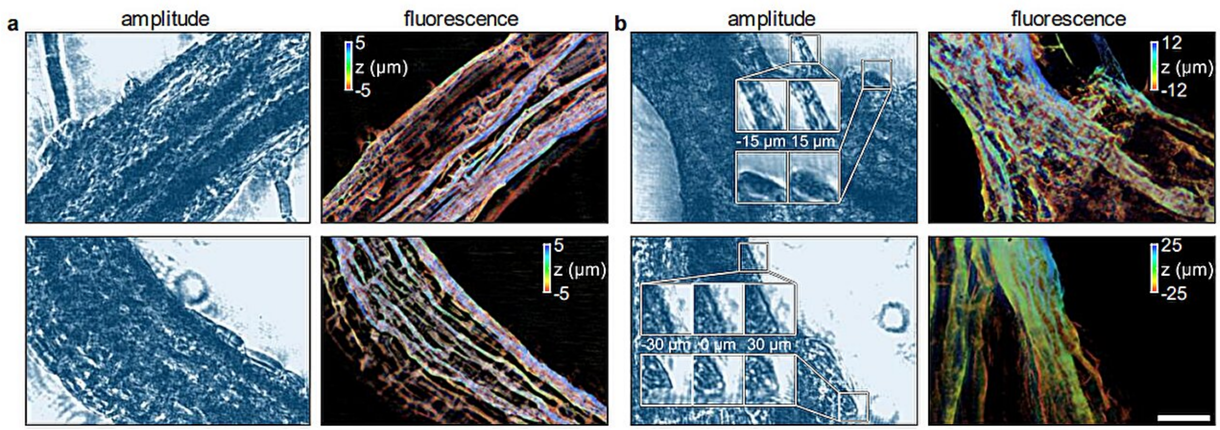
Hybrid imaging approach reveals microbes in 3D

August 26 2024, by Lori Dajose



Amplitude and 3D fluorescence images of additional (a) thin and (b) thick sections on the maize root. Credit: *Proceedings of the National Academy of Sciences* (2024). DOI: 10.1073/pnas.2403122121

Caltech researchers have developed a new method to create three-dimensional images of complex communities of bacteria and plant roots. The technology synthesizes two traditional methods of imaging: visualizing microbes with fluorescence and a noninvasive technique called quantitative phase imaging.

This technology is a step toward understanding the complicated environment of the rhizosphere, the region of soil where a plant's roots interact with microorganisms. Bacteria in the rhizosphere help plants obtain crucial nutrients like phosphorous, but this environment has been difficult to study and image as it is underground.

The research is a collaboration between the laboratory of imaging specialist Changhuei Yang, the Thomas G. Myers Professor of Electrical Engineering, Bioengineering, and Medical Engineering, Heritage Medical Research Institute Investigator, and executive officer for electrical engineering; and the laboratory of biologist Dianne Newman, the Gordon M. Binder/Amgen Professor of Biology and Geobiology, and Merkin Institute Professor.

Their paper describing the study is published in the *Proceedings of the National Academy of Sciences*.

"It's challenging to observe the dynamics in the rhizosphere because it's naturally concealed beneath the opaque layers of soil," says postdoctoral scholar Reinaldo Alcalde, a co-first author of the new study. "This was a motivation to develop better methods to image bacteria in these regions."

Traditionally, researchers who want to study bacterial dynamics have genetically engineered bacteria to fluoresce under laboratory conditions, their artificial green glow visible with a microscope. However, not all microbial species can be engineered in this way. Another method to image bacteria, quantitative phase imaging, is able to detect miniscule differences in transparency without the use of fluorescence.

The new technology is a combination of these two techniques into a single optical setup. Called CFAST (Complex-field and Fluorescence microscopy using the Aperture Scanning Technique), the novel technique can create three-dimensional images of microbial communities much faster and with less damage than commercial microscopes.

"Through the 3D camera setup, the two techniques work simultaneously and seamlessly," says postdoctoral scholar Oumeng Zhang, also a co-first author on the study.

As the work is still a proof of concept, the bacterial communities were imaged outside of soil under simplified conditions. The team aims to continue developing and improving the technology to be able to precisely image roots and bacteria together.

"This is a multidisciplinary collaboration that came out of curiosity and blending two fields of science to create something useful," says Alcalde.

"There are many questions about what really goes on in soil, but there are few efforts aimed at developing good technologies for getting good data from the soil," says Yang. "Hopefully, this project represents the start of an effort here on campus."

More information: Oumeng Zhang et al, Investigating 3D microbial community dynamics of the rhizosphere using quantitative phase and fluorescence microscopy, *Proceedings of the National Academy of Sciences* (2024). DOI: 10.1073/pnas.2403122121

Provided by California Institute of Technology