

## **m-CAFEs: The use of EcoFABs to study beneficial plant-fungus interactions**

Jens Heller<sup>1</sup>, Peter Kim<sup>2</sup>, Trenton K. Owens<sup>1</sup>, Vincent Kam<sup>1</sup>, Peter F. Andeer<sup>1</sup>, Lauren K. Jabusch<sup>1</sup>, Nameera Baig<sup>1</sup>, Dawn M. Chiniquy<sup>1</sup>, Spencer Diamond<sup>3</sup>, Benjamin M. Rubin<sup>3</sup>, Claudio Hidalgo<sup>4</sup>, Matthew Nethery<sup>4</sup>, Joelle Schlapfer<sup>1</sup>, Kateryna Zhalnina<sup>1</sup>, Mary K. Firestone<sup>3</sup>, Jennifer A. Doudna<sup>3</sup>, Jill Banfield<sup>3</sup>, Rodolphe Barrangou<sup>4</sup>, Romy Chakraborty<sup>1</sup>, Adam M. Deutschbauer<sup>1</sup>, Anup K. Singh<sup>2</sup>, Trent R. Northen<sup>1</sup>, and **N. Louise Glass<sup>1</sup>**

<sup>1</sup>Lawrence Berkeley National Laboratory, Berkeley; <sup>2</sup>Sandia National Laboratory, Livermore; <sup>3</sup>University of California, Berkeley; <sup>4</sup>North Carolina State University, Raleigh

<http://eco-fab.org>

Project Goals: To derive mechanistic understanding of fungal contributions to mineral accessibility in marginal soils

The m-CAFEs program is a collaborative, integrated, and mission-driven program to interrogate the function of soil microbiomes with critical implications for carbon cycling and sequestration, nutrient availability and plant productivity in natural and managed ecosystems. We use precisely controlled ecosystem fabrications (EcoFABs) for creating and manipulating plant-microbe interactions under specific environmental conditions. The EcoFABs design consists of a sterile plant growth chambers attached to a microscope slide. This set-up considerably enhances studying the impact of microbes on plant growth phenotypes.

In plants, phosphorus drives biological reactions and is essential for growth. Phosphorus facilitates root formation, reproductive development and synthesis of proteins. Phosphorus is one of the most limited nutrients for plants in the environment because the form that is preferentially assimilated, inorganic phosphate, is unevenly distributed in soils and >80% is not readily available to roots. In the environment, phosphorus is primarily present as insoluble iron and aluminum phosphates in acidic soils or calcium phosphates in alkaline soils.

Numerous microorganisms - especially those associated with roots - have the ability to increase plant growth and productivity. Filamentous fungi of the genus *Trichoderma* are among those microbes and the most commonly studied natural bio-control agents. Some *Trichoderma* species have been shown to protect plants against biotic and abiotic stresses and to promote plant growth by increasing nutrient uptake. Their potential in solubilization of otherwise unavailable mineral nutrients is under investigation. Using EcoFABs, we are able to spatially separate phosphorus sources and plant roots. We demonstrate that *Trichoderma harzianum* promotes plant growth in the presence of insoluble AlPO<sub>4</sub>. Importantly, this effect was only evident when *T. harzianum* was inoculated close to the AlPO<sub>4</sub> source, whereas no growth promotion was observed when the fungus was inoculated close to the seedling. These data suggest that *T. harzianum* helps to solubilize phosphate and makes it available for plant uptake and utilization.

Another efficient strategy of plants to overcome phosphate limitation is the association of roots with arbuscular mycorrhizal (AM) fungi. Even though these microorganisms form symbiotic associations with nearly 80% of terrestrial plants, research on AM fungi is limited due to the challenging nature of these systems. New methods that facilitate the handling of AM fungi are essential to enhance and enable the research on these beneficial organisms. Here we present a new EcoFAB design to study the mycorrhizal plant-fungal interactions. This EcoFAB design consists of a plant growth chamber and compartments for nutrient sources that are separated by a mesh sheet; this set-up effectively prevents roots from accessing nutrients if not associated with AM fungi. Using such a design will enable us to measure and image direct impact of AM fungi on nutrient access, plant growth and rhizosphere microbiome associations.

*Funding statement.*

*This material by m-CAFEs Microbial Community Analysis & Functional Evaluation in Soils, (m-CAFEs@lbl.gov) a Project led by Lawrence Berkeley National Laboratory is based upon work supported by the U.S. Department of Energy, Office of Science, Office of Biological & Environmental Research under contract number DE-AC02-05CH11231*