

Arbuscular Mycorrhizal Fungi Transport Water to Host Plants

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Project Goals: Our project explores the potential of arbuscular mycorrhizal fungi (AMF) to enhance the tolerance of an annual grass to soil desiccation. We specifically ask whether AMF can access water in soil unavailable to plants and transport that water across air gaps to host plants, thus improving plant-water relations. We used stable isotopes to track and quantify water flow from the soil, through fungal hyphae, to plants. In addition, we used fluorescent dyes to elucidate the location and mechanisms of water transport in fungal hyphae. This research substantially deepens our understanding of AMF functioning and AMF-plant interactions. Our findings have possible ramifications for the management of plant drought tolerance in the context of climate change.

Arbuscular mycorrhizal fungi (AMF) form symbiotic associations with 80% of surveyed land plant species and are well-recognized for mobilizing and transferring nutrients to plants. Yet AMF also perform other essential functions, notably improving plant-water relations. Some research attributes this solely to improved plant nutrition and better ability to osmoregulate when plants are partnered with AMF. However, we hypothesize that AMF can also directly transport a significant amount of water to their plant hosts. We used stable isotopes and fluorescent dyes to track and measure water transport by AMF to plants in a greenhouse experiment. *Avena barbata* inoculated with *Rhizophagus intraradices* was planted in one compartment (the ‘plant compartment’) of two-compartment microcosms. The second compartment (the ‘no-plant’ or ‘soil’ compartment) was separated from the first by an air-gap and either an 18 µm mesh, thus excluding roots but allowing AMF hyphae in, or a 0.45 µm mesh, thus excluding both roots and hyphae. ¹⁸O-labeled water and fluorescent dyes (one membrane permeable, the other membrane impermeable) were injected into the soil in the no-plant compartment. Transpired water from *A. barbata* shoots was collected, measured, and tested for ¹⁸O. Hyphae and *A. barbata* roots were collected and imaged with fluorescence microscopy to identify the presence and location of the dyes.

Plants with AMF able to access the soil compartment transpired twice as much water, and the transpired water had a $\delta^{18}\text{O}$ value three times higher, compared to plants with AMF excluded from the soil compartment. We detected the presence of both fluorescent dyes in the roots of plants with AMF accessing the soil compartment, but not in the roots of plants with AMF excluded from the soil compartment. The membrane permeable dye was broadly visible throughout the roots and nearby soil, suggesting hyphal transport and diffusion out. The membrane impermeable dye was visible in/on hyphae within roots only, indicating that it travelled between the hyphal cell membrane and cell wall, since it cannot cross cell membranes. Based on these findings, we conclude that *R. intraradices* transported a significant amount of water to *A. barbata* and likely carried the water more efficiently through hyphae apoplast than symplast. We believe our results are the first to show that AMF can directly transport a

significant amount of water to host plants. We suggest that AMF can play an important role in improving plant-water relations, not only indirectly, but also by acting as direct extensions of root systems, bridging air-gaps, penetrating micropores, and accessing water in soil not easily accessible to roots, and thereby increasing the amount of soil water available to plants.

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