

At the Interface: Glycoproteins, Glycolipids, and WAK-mediated Signaling are Required for Plant-Microbial Symbiosis in *Medicago truncatula*

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In nature plants interact with beneficial root associated bacteria and fungi that aid in nutrient uptake and promote plant growth and resilience. Here we use *Medicago truncatula* as a model plant to investigate the role of the plant cell surface in beneficial plant-microbe interactions with arbuscular mycorrhizal fungi (AMF) and nitrogen-fixing *Sinorhizobium*. The plant cell surface plays a key role in symbiosis by forming a specialized plant-microbial interface composed of plant-derived membranes, cell wall polysaccharides, and protein complexes, through which nutrients and information are bi-directionally exchanged^{1,2}. It has previously been reported in the literature through the use of glycan-directed monoclonal antibodies that arabinogalactan proteins (AGPs) aggregate at the symbiotic interface in a variety of plant-microbe mutualisms, however, the identity of the genes encoding these glycoproteins has remained unknown³⁻⁶. Here we report the discovery of several AGP encoding genes in *M. truncatula* that are specifically expressed during symbiosis with either AMF or *Sinorhizobium meliloti*. Functional studies using RNAi-mediated knockdown of specific AGPs result in drastic symbiotic phenotypes, including poor mycorrhizal colonization and impaired nitrogen fixation in *Sinorhizobium* infected root nodules. In parallel, we have identified a wall-associated kinase (WAK)-like receptor and a glycosyltransferase enzyme involved in sphingolipid biosynthesis that are also required for symbioses. Our data indicate that glycopeptides, glycolipids, and WAK mediate signaling at the cell surface is necessary for establishing and maintaining symbiosis in *M. truncatula*.

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References

1. Balestrini, R. & Bonfante, P. The interface compartment in arbuscular mycorrhizae: A special type of plant cell wall? *Plant Biosystems - An International Journal Dealing with all Aspects of Plant Biology* **139**, 8–15 (2005).
2. Brewin, N. J. Plant Cell Wall Remodelling in the Rhizobium–Legume Symbiosis. *Critical Reviews in Plant Sciences* **23**, 293–316 (2004).
3. Jackson, O., Taylor, O., Adams, D. G. & Knox, J. P. Arabinogalactan Proteins Occur in the Free-Living Cyanobacterium Genus *Nostoc* and in Plant–*Nostoc* Symbioses. *Mol. Plant Microbe Interact.* **25**, 1338–1349 (2012).

4. Bonfante-Fasolo, P., Tamagnone, L. & Peretto, R. Immunocytochemical location of hydroxyproline rich glycoproteins at the interface between a mycorrhizal fungus and its host plants. *Protoplasma* (1991).
5. Tsyganova, A. V. *et al.* Distribution of legume arabinogalactan protein-extensin (AGPE) glycoproteins in symbiotically defective pea mutants with abnormal infection threads. *Cell Tiss. Biol.* **3**, 93–102 (2009).
6. Berry, A. M. *et al.* Arabinogalactan proteins are expressed at the symbiotic interface in root nodules of *Alnus* spp. *New Phytologist* **155**, 469–479 (2002).