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Analyzing the soybean transcriptome during autoregulation of mycorrhization identifies the transcription factors GmNF-YA1a/b as positive regulators of arbuscular mycorrhization

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Abstract

Background: Similarly to the legume-rhizobia symbiosis, the arbuscular mycorrhiza interaction is controlled by autoregulation representing a feedback inhibition involving the CLAVATA1-like receptor kinase NARK in shoots. However, little is known about signals and targets down-stream of NARK. To find NARK-related transcriptional changes in mycorrhizal soybean (*Glycine max*) plants, we analyzed wild-type and two *nark* mutant lines interacting with the arbuscular mycorrhiza fungus *Rhizophagus irregularis*.

Results: Affymetrix GeneChip analysis of non-inoculated and partially inoculated plants in a split-root system identified genes with potential regulation by arbuscular mycorrhiza or NARK. Most transcriptional changes occur locally during arbuscular mycorrhiza symbiosis and independently of NARK. RT-qPCR analysis verified nine genes as NARK-dependently regulated. Most of them have lower expression in roots or shoots of wild type compared to *nark* mutants, including genes encoding the receptor kinase GmSIK1, proteins with putative function as ornithine acetyl transferase, and a DEAD box RNA helicase. A predicted *annexin* named *GmAnnx1a* is differentially regulated by NARK and arbuscular mycorrhiza in distinct plant organs. Two putative *CCAAT-binding transcription factor* genes named *GmNF-YA1a* and *GmNF-YA1b* are down-regulated NARK-dependently in non-infected roots of mycorrhizal wild-type plants and functional gene analysis confirmed a positive role for these genes in the development of an arbuscular mycorrhiza symbiosis.

Conclusions: Our results indicate *GmNF-YA1a/b* as positive regulators in arbuscular mycorrhiza establishment, whose expression is down-regulated by NARK in the autoregulated root tissue thereby diminishing subsequent infections. Genes regulated independently of arbuscular mycorrhization by NARK support an additional function of NARK in symbioses-independent mechanisms.

Keywords: Affymetrix annexin, GeneChip, autoregulation, arbuscular mycorrhiza, CCAAT-binding transcription factor NF-Y, *Rhizophagus irregularis*, *Glycine max* (soybean), quantitative RT-PCR, split-root system

Background

Plants have a long success story in hosting microsymbionts in their roots to improve their supply with mineral nutrients, particularly the two important macronutrients phosphate and nitrogen. The arbuscular mycorrhiza (AM) symbiosis, an interaction of plants

with fungi of the phylum *Glomeromycota* [1], probably co-evolved with the early land plants around 450 million years ago (for overview see [2]). Nowadays, the majority of land plants can form an AM symbiosis that is characterized by the exchange of phosphate against monosaccharides [3]. Moreover, mycorrhizal plants can also benefit from improved availability of other minerals and water, and from induced abiotic and biotic stress tolerance, all contributing to higher plant biodiversity and productivity of ecosystems [4]. AM fungi are obligate

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