

The *Medicago truncatula* nitrate transporter LATD/NIP plays an important role in maintaining root and nodule meristem function and in mediating ABA signaling. In order to identify transcription factors (TFs) that function in the ABA-LATD/NIP signaling pathway, we performed a large-scale gene expression profiling by qRT-PCR on wild-type (WT) and *latd* roots grown with or without ABA. We found that the set of TFs regulated by ABA differs in WT and *latd* mutants with only 42 out of 192 genes (22%) regulated by ABA in both genotypes. Conversely, we found that the genes misexpressed in *latd* mutants change almost completely in the presence of exogenous ABA. The only gene whose expression is altered by ABA in both genotypes and by the *latd* mutation under both ABA treatments is the NODULATION SIGNALING PATHWAY 2 (NSP2) gene. The NSP2 gene encodes a transcription factor required for the formation of nitrogen-fixing symbiotic nodules. We show that *NSP2* and *preMIR171h*, which encodes a miRNA that targets *NSP2*, are both regulated by nitrate, indicating a role for NSP2 in root nitrate responses. We find that *preMIR171h* is also regulated by ABA and *LATD/NIP*, indicating that NSP2 is regulated by ABA at multiple levels, and that LATD/NIP is required for this process. NSP2 is also regulated by cytokinin signaling during nodulation. We found that *Rhizobium* inoculation failed to induce expression of NSP2 as well as that of two NSP2-regulated genes, *NIN* and *CLE13*, in *latd* roots, and that the regulation of these genes by the hormone cytokinin is altered as well, both in timing and in level of induction. Together, these findings indicate a distinct and critical role for LATD/NIP and ABA in the early nodulation signaling pathway, particularly in the regulation of NSP2, and in the crosstalk between cytokinin and Nod factor.