

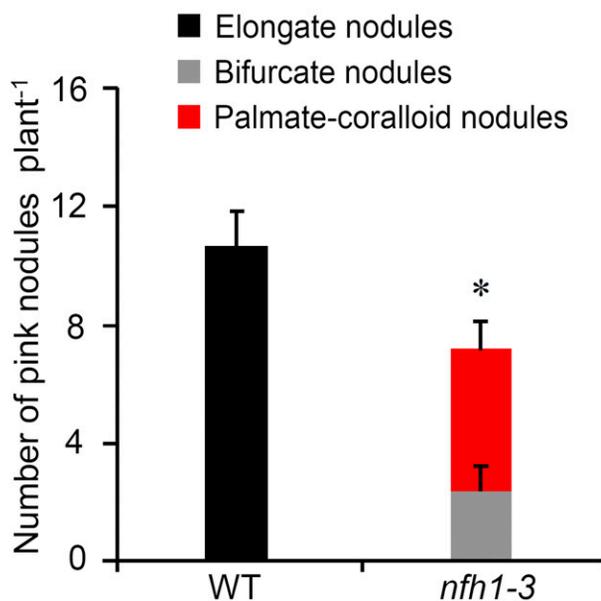
## IN BRIEF

## Goldilocks Principle: MtNFBH1 Ensures Optimal Nod Factor Activity <sup>OPEN</sup>

Partner selection is a critical step that must occur early during establishment of root nodule symbiosis (RNS). RNS refers to the mutualistic interaction between legumes and some nonlegumes with soil bacteria that help convert atmospheric nitrogen into plant usable ammonia. In legumes such as *Medicago truncatula*, a successful interaction initiates two genetically separable but interdependent developmental programs: infection thread (IT) formation and nodule organogenesis. Only once the IT reaches the nodule cortex and the bacteria are released does nitrogen fixation commence (Oldroyd et al., 2011). However, for these processes to occur, legumes must first carefully choose their partner by exchanging chemical signals.

Plants secrete chemoattractants called flavonoids that in turn stimulate production of rhizobial strain-specific signaling molecules called Nod (nodulation) factors (NFs). NFs comprise a chitin-based backbone of variable length that is acylated at the N-terminal end and can have additional functional group modifications resulting in unique chemical signatures (Wang et al., 2012). If rhizobia fail to produce the correct NF or produce too little, RNS is impaired (Roche et al., 1996). However, overproduction of NFs can also be detrimental to symbiosis. **Cai et al. (2018)** characterize the *M. truncatula* NOD FACTOR HYDROLASE1 to demonstrate that not only the presence of specific NFs but also their concentration controls the establishment of RNS.

The *M. truncatula* NOD FACTOR HYDROLASE (MtNFBH1) enzyme can hydrolyze the *Sinorhizobium meliloti* (*Sm*) NFs into lipodisaccharides in vitro and in the rhizosphere (Tian et al., 2013). In this study, the authors show that both the activity and the expression of MtNFBH1 increase with increasing concentrations of the NFs but are strongly reduced in the *dmi1*, *dmi2*, and *dmi3* mutants, indicating that MtNFBH1 lies downstream of the common symbiosis pathway. Consistent with



The *M. truncatula nfh1-3* mutant produces fewer nodules and young nodules have a palmate-coralloid structure. (Adapted from Cai et al. [2018] Figure 8.)

its localization in infection chambers of curled root hairs, mutants in this chitinase-like hydrolase gene had fewer infection events and the root hairs showed aberrant branching. The strongest mutant allele, *nfh1-3*, had fewer nitrogen-fixing pink nodules, and these nodules clustered together and possessed multiple meristems. The authors hypothesized that, since accumulation of uncleaved NFs resulting from a lack of NF hydrolase activity caused aberrant organogenesis, overproduction of NFs must similarly affect nodule morphology. In line with this reasoning, an *Sm* strain carrying

extra copies of NF synthesis genes induced coraloid nodules with apical meristem branching on wild type roots.

Curiously, transgenic overexpression of MtNFBH1 yielded a similar phenotype as the mutant. These lines had fewer pink nodules and the ones that formed were branched. How can too much and too little of symbiotic NF be detrimental to establishment of RNS? The authors propose that MtNFBH1 spatiotemporally limits NF levels in order to reach an optimal NF activity for initiating the right number of infections and correct nodule

morphology. It remains to be seen how *MtNFH1* relates to the phenomenon of autoregulation of nodulation in legumes, by which the plant controls the number of nodules formed.

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