

We are IntechOpen, the world's leading publisher of Open Access books Built by scientists, for scientists

6,900

Open access books available

186,000

International authors and editors

200M

Downloads

Our authors are among the

154

Countries delivered to

TOP 1%

most cited scientists

12.2%

Contributors from top 500 universities



WEB OF SCIENCE™

Selection of our books indexed in the Book Citation Index
in Web of Science™ Core Collection (BKCI)

Interested in publishing with us?
Contact book.department@intechopen.com

Numbers displayed above are based on latest data collected.
For more information visit www.intechopen.com



Effects of Phytohormones on Nodulation and Nitrogen Fixation in Leguminous Plants

Maki Nagata and Akihiro Suzuki

Additional information is available at the end of the chapter

<http://dx.doi.org/10.5772/57267>

1. Introduction

Leguminous plants form root nodules, in which symbiotic rhizobia fix atmospheric nitrogen. The nodulation process in the legume-rhizobium symbiosis consists of a series of events initiated by an exchange of specific signaling compounds between the two partners. The roots of leguminous plants secrete flavonoids, which trigger the synthesis of lipochitin-oligosaccharide signaling molecules, Nod factors (NFs), by rhizobia. NFs activate nodule organogenesis in the roots by stimulating the division of cortical cells. Many nodule development stages resemble other plant organ development such as cell divisions and differentiations. Because phytohormones are signal molecules involved in most plant physiological activities, they are likely to positively or negatively regulate nodulation and nitrogen fixation in the legume-rhizobium symbiosis.

In this chapter, we review the roles of several key phytohormones, namely auxins, cytokinins, gibberellins, ethylene, brassinosteroids, abscisic acid, salicylic acid, jasmonic acid and strigolactones in nodulation and nitrogen fixation in leguminous plants.

2. Auxins

Auxins were the first class of plant hormones discovered, and play a central role on the regulation of germination, plant growth, flower bud formation and flowering, and other developmental processes. In addition, auxins are involved in responses to environmental stimuli such as temperature, light and gravity. The most important member of the auxin family is indole-3-acetic acid (IAA), a native auxin in plants. The highest auxin levels are found in the cells undergoing cell division, elongation, differentiation and vascular bundle formation.

Therefore, auxin may play a significant role in nodulation. The possible involvement of auxins in nodule formation was first reported by Thimann in 1936. Thimann reported that the nodules of *Pisum sativum* contained auxin and that the auxin content increased during root nodule development [1]. Since then, this indirect evidence for an involvement of auxin in nodulation has been supported by various experiments. Hirsch et al. (1989) showed that auxin transport inhibitors, such as N-(1-naphthyl) phthalamic acid (NPA) and 2,3,5-triiodobenzoic acid (TIBA), caused the formation of nodule-like structures in *Medicago sativa* [2]. In *Trifolium repens*, transgenic plants carrying an auxin-responsive promoter (GH3) fused to the GUS reporter gene have been used to visualize the presence of auxins inside roots during nodule organogenesis [3]. The authors found that rhizobia cause a localized, temporary and early inhibition of auxin transport, which subsequently leads to an accumulation of auxins at the site of nodule initiation. A similar expression pattern was also observed in *Medicago truncatula* by using a DR5::GUS auxin-responsive promoter. In addition, *MtPIN*-silencing plants had significantly fewer nodules than control plants [4]. van Noorden et al. (2006) showed that the supernodulating *sun1* mutant had increased auxin transport and auxin content and that long-distance auxin transport regulation by rhizobia was defective [5]. de Billy et al. (2001) showed that genes in *M. truncatula* related to the auxin import carrier gene *AtAUX1*, which they named *MtLAX*, are predominantly expressed in regions of the root tips and nodule primordia where the vasculature arises (i.e., in the center of the lateral roots and at the peripheral region of the nodules) [6]. These results suggest that auxins are required during the development and differentiation of nodule primordia and of the vasculature within the nodules [6].

These reports also suggest that the auxin transport system is an important control on the number of indeterminate-type nodules. The effects of treatment with auxin transport inhibitors (NPA and TIBA) and with an auxin antagonist, α -(phenylethyl-2-one)-indole-3-acetic acid (PEO-IAA), on determinate-type nodulation were investigated by using *Lotus japonicus*. Both the nodule number and nodule development decreased and the formation of lenticels, which normally develop on the root surface and originate from the root outer cortex, was also inhibited by the treatment [7, 8]. The GH3:GUS transformant of *L. japonicus* showed auxin responses during nodule development. In rhizobia-inoculated roots, GH3-driven expression started to increase in the outer cortical cells where cell divisions occurred in the nodule primordia. GH3-driven expression was connected to the main root vascular tissues. These results suggest that auxins play an important role in the development of nodule vasculature, regardless of the nodule type [7-9]. In indeterminate-type nodule-forming plants, auxins accumulate at the site of rhizobia inoculation. This is caused by the inhibition of polar auxin transport by an accumulation of flavonoids, which are known as to be auxin transport regulators, around the infection site. In contrast, flavonoid-regulated auxin transport inhibition is not crucial during root nodule formation in *Glycine max*, which produces determinate-type nodules [10]. In the determinate-type nodules of *L. japonicus*, no inhibition of auxin transport was observed [9]. These differences in auxin distribution and transport inhibition between plants with determinate and indeterminate nodules have been attributed to a difference in the developmental pattern of the two nodule types [3, 11].

The role of auxins in nodulation is linked to the development of other root structures, such as lateral roots. Both lateral roots and root nodules development are known to be regulated by the auxin-to-cytokinin ratio. An increase in the auxin concentration stimulated lateral root formation, whereas an increase of in the cytokinin concentration or an inhibition of auxin transport induced the development of pseudo-nodules. Previously, it has been assumed that nodule initiation in plants that form indeterminate-type nodules is stimulated by a low auxin-to-cytokinin ratio [2]. Recently, Suzaki et al. (2012) investigated auxin distribution during root nodule development by using a DR5::GFP transformant of *L. japonicus* [12]. The accumulation of auxin in the dividing cortical cells was positively regulated by the nodule inception (NIN), transcription factor in nodule development and was inhibited by a negative systemic regulatory mechanism called autoregulation of nodulation. Moreover, auxin accumulation was observed in uninoculated roots of the *L. japonicus* mutant *spontaneous nodule formation 2 (snf2)*, which has a gain-of-function mutation in *LHK1* encoding the putative cytokinin receptor LOTUS HISTIDINE KINASE 1. Therefore, it appears that auxins are involved in the division of cortical cells and acts downstream of cytokinin signaling [12, 13] (Figure 1).

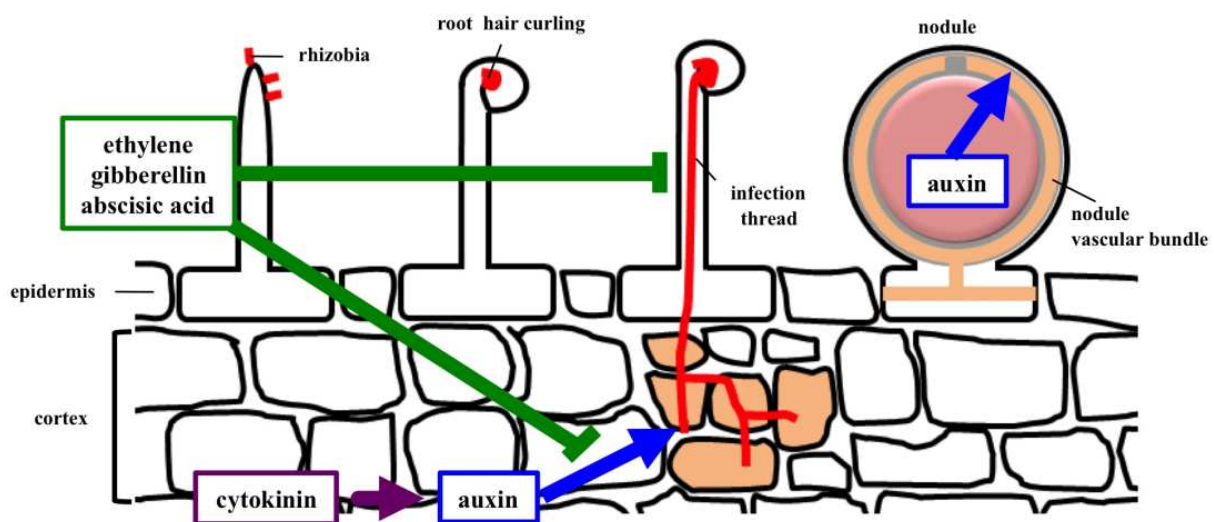


Figure 1. Effects of phytohormones on nodulation process in leguminous plants. Effects of phytohormones on nodulation process in leguminous plants. Auxin and cytokinin are required for cortical cell division and auxin acts downstream of cytokinin signaling. Ethylene, gibberellin, and abscisic acid inhibit the cortical cell divisions and rhizobial infection.

3. Cytokinins

Cytokinins have been found in all living cells of intact higher plants. Cytokinins play roles ranging from minor to major throughout development, from germination to leaf and plant senescence and modulate physiological processes important throughout the life of the plant,

including photosynthesis and respiration [14, 15]. Cytokinins are also involved in as the control of root architecture development, including root nodulation.

In *Medicago polymorpha*, the synthetic cytokinin 6-benzyl-amino-purine (BAP) significantly increased the number of nodules [16]. In addition, BAP had a dose-dependent effect on nodulation in *P. sativum*. High levels of BAP results in few, flat, pale nodules and abnormal infection threads in wild-type *P. sativum* [17]. Expression of the alfalfa nodule-specific marker gene *MsENOD2* was induced by the exogenous application of a cytokinin in *M. sativa* [18]. The exogenous application of BAP on *M. sativa* induced cortical cell divisions and the expression of the early nodulin *ENOD40* gene [19]. In *G. max*, the cytokinin *trans*-zeatin stimulated nodulation at concentrations lower than 2.5×10^{-8} M, whereas at higher concentrations (5.0×10^{-8} M) it inhibited the formation of nodules [20]. Lohar et al. (2004) developed cytokinin-resistant transgenic *L. japonicus* hairy roots by using cytokinin oxidase (CKX) genes from *Arabidopsis thaliana* (*AtCKX3*) and maize (*ZmCKX1*), and showed that these roots had significantly reduced numbers of root nodules compared to control roots [21]. These reports suggest that cytokinins play a significant role in nodule organogenesis.

Recent research has confirmed that cytokinin receptors also play a significant role in nodule organogenesis [22-24]. The gain-of-function *snf2* mutant of the *L. japonicus* cytokinin receptor gene (*LHK1*) can form spontaneous nodules without rhizobial infection. This result indicates that cytokinin signaling is indispensable for cell division and for initiating nodule development [24]. In addition, *L. japonicus* plants homozygous for a mutation in the *hyperinfected 1* (*hit1*) locus exhibit abundant infection thread formation but fail to initiate cortical cell division in response to rhizobial signaling [23]. In addition, RNA interference of the cytokinin receptor homolog *cytokinin response 1* (*MtCRE1*) led to the development of cytokinin-insensitive roots, which showed an increased number of lateral roots and a strong reduction in nodulation [22]. Based on these reports, it has become clear that cytokinins act downstream of early NF signaling to mediate nodule formation. Cytokinins may therefore be the most important differentiation signal for cortical cell division and differentiation and for nodule organogenesis (Figure 1).

Exogenous application of cytokinins enhanced nitrogenase activity in nodules at all stages [25]. Rao et al. (1984) reported that the stimulatory effect of cytokinins increased the efficiency of nitrogen fixation. The application of cytokinins stimulated nitrate-induced nitrate reductase activity in the dark. Stimulation of nitrate reductase by cytokinins was inhibited significantly 6-methylpurine and cycloheximide, suggesting a requirement of RNA and protein synthesis [26].

4. Gibberellins

Gibberellins (GAs) are another important family of growth regulators in higher plants. GAs are also involved in root nodule symbiosis. Mutants deficient in GA biosynthesis or signaling develop dwarf phenotypes [27]. The GA-deficient mutants of *P. sativum* also developed significantly fewer nodules than wild-type plants. The application of an exogenous GA restored the nodule number in these mutants, although the addition of higher concentrations

of GAs no longer restored nodule formation in these mutants. These results suggest that reduced levels of root GAs significantly decrease the number of nodules, and that nodule formation is considered to be strictly controlled by the GA concentration [28]. On the other hand, GAs are downstream signals of NFs for root hair curling process and for formation of infection pockets and infection threads at lateral root bases, and are essential for nodule primordium formation and differentiation in *Sesbania rostrata* [29]. The application of GA₃ at concentrations from 10⁻⁷ to 10⁻⁴ M results in the formation of nodule-like structures in the roots, and this response is sensitive to nitrogen levels [30]. Maekawa et al. (2009) also reported details of the effects of GAs on root nodulation in *L. japonicus*. Exogenous application of GA₃ (at more than 10⁻⁸ M) inhibited the number of infection threads and nodules. In contrast, the formation of both infection threads and nodules were stimulated by the application of Uniconazole-P, an inhibitor of GA₃ biosynthesis. Moreover, the degree of NF-induced root hair deformation was attenuated by the application of GA₃. The GA₃-treated *snf1* (a gain-of-function mutation of calcium/calmodulin-dependent kinase, CCaMK) and *snf2* (a gain-of-function mutation of a cytokinin receptor) showed a significant reduction in the number of spontaneous nodules. The cytokinin-dependent induction of *NIN* was suppressed by GA₃ treatment. These results suggest that GAs are involved in the cytokinin signaling pathway for nodulation in *L. japonicus* [31] (Figure 1).

The F-box containing protein, SLEEPY 1 (SLY1), functions as a positive regulator in GA signaling. In the presence of GAs, SLY1 interacts with negative regulators of GA signaling, leading to the degradation of these negative regulators [32, 33]. The over-expression of *L. japonicus* SLY1 carrying a gain-of-function mutation resulted in a reduced number of nodules, though the number of lateral roots and the degree of root growth were not significantly affected [31]. The constitutive GA signaling mutants of *P. sativum*, *la cry-s* mutants, also form significantly fewer nodules than wild-type plants. However, GA deficiency that results from the *na* mutation in *P. sativum* causes a reduction in nodulation. These results suggest that there is an optimal degree of GA signaling required for nodule formation and that the GA signal, and not the concentration of bioactive GA is important for nodulation [34].

The application of a GA decreased nodulation and nitrogen fixation under optimal growth conditions [35]. However, in *G. max* under low soil temperature conditions, nodulation and nitrogen fixation were decreased by GA₃ treatment during early plant development, but were increased during later development. The application of GA₃ increased nodulation and nitrogen accumulation after the early pod filling stage. These results suggest that GAs applied to soybean seeds at the time of planting did not influence final grain and protein yield [36].

5. Ethylene

Ethylene is a gaseous phytohormone involved in fruit ripening, leaf and fruit abscission, germination, seedling morphogenesis, root emergence, root hair elongation, promotion of flowering, senescence and stress response. Several studies have shown that ethylene production can have a negative effect on nodule formation. For example, ethylene production significant-

ly increased in roots infected by rhizobia, and added exogenous ethylene can decrease the number of nodules [37, 38]. In *P. sativum*, exogenous ethylene did not decrease the number of infections per lateral root, but nearly all of the infections were blocked when the infection thread was in the basal epidermal cell or in the outer cortical cells [38]. In addition, ethylene inhibited all of the early plant responses that were tested, including the initiation of calcium spiking in *M. truncatula*. This finding suggests that ethylene acts upstream or at the point of calcium spiking in the NF signal transduction pathway [39]. Nodule formation can be stimulated by treatment of *M. truncatula* roots with aminoethoxyvinyl glycine (AVG) or Ag⁺, which are inhibitors of ethylene synthesis and perception, respectively [40, 41]. The hypernodulation phenotype of the *sickle* mutant of *M. truncatula* has been attributed to a mutation causing ethylene insensitivity [42]. This mutant is defective in the ortholog of Arabidopsis *ETHYLENE-INSENSITIVE 2* (*EIN2*), involved in ethylene signaling [43]. In *L. japonicus*, root nodule formation was suppressed by 1-aminocyclopropane-1-carboxylic acid (ACC), a precursor of ethylene, but was enhanced by AVG and silver thiosulfate [44]. These results suggest that an ethylene-mediated signaling pathway is involved in the nodulation process, regardless of the nodule type. Nukui et al. (2004) produced transgenic *L. japonicus* carried the mutated melon ethylene receptor gene *Cm-ERS1/H70A*, which confers ethylene insensitivity [45]. When inoculated with *Mesorhizobium loti*, transgenic plants showed markedly higher numbers of infection threads and nodule primordia in their roots than did control plants. This result is consistent with the result reported for the *sickle* mutant of *M. truncatula* [42]. In addition, transcripts of *NIN* increased in the inoculated transgenic plants as compared with levels in the wild-type plants. In leguminous plants, the early stage of nodule development, including infection thread formation and the emergence of nodule primordia, are likely to be negatively regulated by ethylene signaling (Figure 1). In *snf* mutants, ethylene inhibits spontaneous nodulation. Therefore, ethylene plays a role in nodule formation downstream of the cytokinin signaling pathway [46]. However, ethylene may not play a significant role in nodule formation in all species. In soybean plants that form determinate-type nodules, the application of exogenous ethylene did not inhibit nodulation, and treatment with AVG or Ag⁺ did not increase nodule number [38, 47-49]. In addition, ethylene leads to the formation of infection pockets and the initiation of nodule primordia in *S. rostrata* [50].

ACC deaminase catalyzes the degradation of ACC into ammonium and α -ketobutyrate. The ACC deaminase gene (*acdS*) has been found in many rhizosphere bacteria [51, 52]. Through the action of this enzyme, ACC deaminase-containing bacteria can reduce ethylene biosynthesis in plants. In *M. loti*, *acdS* was found in the symbiosis island, and the enhancing effect of this gene on enhancing the nodulation of *L. japonicus* was demonstrated by using an *M. loti* *acdS* disruption mutant [53]. ACC diaminase in *Rhizobium leguminosarium* bv. *viciae* has been confirmed to enhance nodulation of *P. sativum* [54]. Reports concerning rhizobial strategies to reduce the amount of ethylene synthesized in the host leguminous plant suggest the importance of ethylene-mediated interactions in the establishment of symbiosis between the partners, by decreasing the negative effect of ethylene on nodulation [55].

6. Brassinosteroids

Brassinosteroids (BRs) are a group of plant steroid hormones that regulate a wide range of physiological responses, including cell elongation, photomorphogenesis, xylem differentiation, and seed germination. BRs are present in the plants in extremely low concentrations, but they are highly mobile within the plant. BRs supplied via the root system remarkably promoted the elongation of cotyledon petioles and the hypocotyls of young radish and tomato plants [56]. This study clearly demonstrated the mobility of BRs in the plant system.

In addition, application of BRs affected nodulation and nitrogen fixation in groundnut (*Arachis hypogaea*), pea and soybean [57-59]. In groundnuts, BRs enhanced the growth and yield of the plants, and the growth promotion was associated with enhanced levels of nucleic acids, soluble proteins and carbohydrates [60]. The effect of BRs on nodulation and nitrogen fixation was also investigated. Exogenous application of BR increased in nodulation. Foliar application of BRs also increased the nitrogenase activity [57]. The application of 24-epibrassinosteroid also increased the nodule number, nodule fresh and dry mass, and nitrogenase activity in relation to those of the control in pea [59]. In the hypernodulating En6500 mutant of the 'Enrei' soybean cultivar, the application of BR to the leaves not only induced stem elongation but also repressed root nodule formation, depending on the dose. However, this effect was not observed in the wild-type. On the other hand, foliar treatment with brassinazole, an inhibitor of BR biosynthesis, increased the nodule number and significantly reduced stem elongation in wild-type 'Enrei'. These results suggest that BRs in the shoots may contribute mainly to the regulation of nodule formation and that brassinazole transferred to the shoot from the culture medium subsequently reduced the level of endogenous BRs in the leaves [58]. From these results, BRs are clearly involved in nodulation and nitrogen fixation. The BR synthesis mutants *lk* and *lkb* exhibit a severe reduction in level of bioactive BRs in the shoot [61, 62]. A reduction in BR levels in the roots has also been confirmed for *lkb* [63]. These mutants *lk* and *lkb* and the BR response mutant *lka* also have fewer nodules than wild-type plants. These mutants also have fewer and shorter lateral roots. However, the average nodule dry weight increased significantly. Thus, although the root system dry weight decreased, the average nodules dry weight increased. This finding illustrated that nodule size is not simply a reflection of root system dry weight. Ferguson et al. (2005) also suggested that BRs affect the nodulation mechanism of the shoot that is involved in regulating the nodule numbers of the root [28]. They found that uniconazole-P, which is a GA₃ biosynthesis inhibitor, partially inhibits BR biosynthesis. In *L. japonicus*, the application of BR combined with Uniconazole-P significantly decreased the number of infection threads, compared with a treatment with uniconazole-P alone, but the decrease was less significant than the decrease in GA levels. However, no significant effects on the nodule number, and on shoot and root lengths, were observed. These results suggest that uniconazole-P inhibits BR biosynthesis in the root hairs, and thus lowers the number of infection threads that develop [31] (Figure 1).

7. Absciscic acid

Absciscic acid (ABA) plays crucial roles in plant growth, development and responses to environment stresses such as cold, drought and high salinity. ABA has been reported to play negative roles at different stages of nodule development. The application of ABA inhibited nodulation in *P. sativum* [64], *G. max* [65, 66], *L. japonicus* [67], *T. repens* [67] and *M. truncatula* [68]. ABA application to wild-type *G. max* and to a hypernodulation mutant, NOD1-3, reduced both nodule numbers and isoflavonoid accumulation. It has been shown that isoflavonoids in *G. max* are responsible for the activation of nodulation, thus ABA could have an indirect role in nodule organogenesis through its effects on isoflavonoid synthesis. The effect of ABA on *T. repens* and *L. japonicus*, which form indeterminate and determinate type nodules respectively, was examined. Both leguminous plants showed a decrease in nodule numbers in response to ABA application. Similarly, after the application of abamine, an ABA biosynthesis inhibitor, nodule number increased in *L. japonicus*. The application of ABA on *T. repens* blocked root hair deformation at the stage between root hair swelling and curling [67]. In addition, ABA treatment inhibited infection thread formation in *L. japonicus* and *M. truncatula* [68, 69]. Moreover, calcium spiking after NF perception was inhibited by ABA treatment in *M. truncatula* [68]. Phillips (1971) found that exogenous ABA inhibited root nodule formation by inhibiting the cytokinin-induced cortical cell divisions required for nodule initiation [64]. These results suggest that ABA controls root nodulation by regulating root hair deformation, infection thread formation, and cytokinin-induced cortical cell division in leguminous plants (Figure 1).

The nitrogenase activity of nodules treated with ABA was lower than in untreated wild-type in *Phaseolus vulgaris* [70] and *P. sativum* [71]. In *P. sativum*, ABA application stimulated an abrupt stress situation of severe drought which led to leghemoglobin reduction. Thus, an effect of ABA on nodule oxygen diffusion might also be involved in the decline of nitrogen fixation. The *enhanced nitrogen fixation1* (*enf1*) mutant of *L. japonicus* was isolated by screening *L. japonicus* seedlings for survival on an agar medium containing 70 μ M ABA. The *enf1* mutants showed both increased root nodule numbers and enhanced nitrogen fixation activity. The low ABA sensitivity of the *enf1* mutants was caused by lower endogenous ABA concentration. Moreover, nitrogen fixation activity in the *enf1* mutants increased as a result of decreased nitric oxide production in the nodules [72].

The role of ABA in autoregulation of nodulation was investigated in the *G. max* hypernodulation mutant *nts382*. The basal levels of ABA in the roots of wild-type *G. max* cv. 'Bragg' were higher than those in *nts382*, regardless of *Bradyrhizobium* inoculation. The ABA concentration in the shoot increased at the onset of autoregulation in 'Bragg' but not in *nts382*. The ABA-to-cytokinin ratio in the roots was also consistently higher in 'Bragg' than in *nts382*. This phytohormone ratio had been suggested to be involved in root-to-shoot signaling and photosynthetic gas exchange in *M. sativa* [73]. A model was proposed to explain the possible influence of the ABA-to-cytokinin ratio in autoregulation of nodulation. However, Biswas et al. (2009) proposed that ABA was not directly involved in the systemic autoregulation of nodulation, because an ABA insensitive mutant of *L. japonicus* cv. 'Beyma' did not exhibit

altered autoregulation of nodulation, and the application of ABA on one side of the roots inhibited nodulation locally but not systemically in a split-root experiment [74].

8. Salicylic acid

Salicylic acid (SA) is involved in plant responses to pathogen, and its mode of action has been well characterized. SA is an inducer of systemic acquired resistance in the defense responses to pathogen attacks. In terms of the interaction between plants and rhizobia, several reports have shown that SA strongly inhibits nodulation and nodule development, leading to decreased nitrogen fixation activity. When *M. sativa* was inoculated with compatible *Rhizobium meliloti*, SA levels in the roots either decreased or remained close to their basal levels. However, when *M. sativa* was inoculated with incompatible *Rhizobium leguminosarum* or the *nod* mutant of *R. meliloti*, that was defective in NF biosynthesis, SA accumulated in roots. These results suggest the involvement of NFs produced by compatible rhizobia in the inhibition of the SA-mediated defense in leguminous plants [75]. In another study, inoculation of *P. sativum sym30* mutant (*nod*⁻) with compatible *R. leguminosarum* increased SA levels in the roots. Similarly, SA accumulation in the roots was found in *P. sativum* inoculated with a *NodC* mutant. However, SA levels in roots either remained at the basal level or decreased when *P. sativum* plants were inoculated with compatible *R. leguminosarum*. These results suggest that the *sym30* gene could be involved in a common pathway that leads to the suppression of an SA-dependent defense mechanism in leguminous plants against compatible rhizobia, thus allowing establishment of the symbiosis [76]. van Spronsen et al. (2003) found that SA application completely inhibited the formation of indeterminate-type nodules in *Vicia sativa* subsp. *nigra* and *P. sativum* [77]. However, SA application did not inhibit the formation of determinate-type nodules in *L. japonicus*, *Glycine soja*, *G. max* and *P. vulgaris*. On the other hand, SA application at higher concentrations decreased the number of determinate-type nodules and the dry mass of *G. max* seedlings, leading to a low photosynthetic rate and decreased nitrogen fixation [78]. The inhibitory effect of SA on the nodulation of hypernodulating soybean mutants NOD1-3 and NOD2-4 was significantly less pronounced than that in wild-type soybean. These results indicate that SA is directly involved in signal transmission in the autoregulation [79]. In addition, when endogenous SA levels were modulated through the transgenic expression of salicylate hydroxylase (*NahG*) in both *L. japonicus* and *M. truncatula*, a marked reduction in SA levels was correlated with an increase in the number of infections and the number of nodules [80]. These results suggest that endogenous SA levels affect nodulation in both determinate and indeterminate-type nodule-forming species.

9. Jasmonic acid

Jasmonic acid (JA) is also involved in plant defenses against pathogens and in wound responses. JA has been reported to be a negative regulator of nodulation. In *L. japonicus*, shoot-applied methyl jasmonate (MeJA) strongly suppressed nodulation, including infection thread

formation and *NIN* gene expression in wild-type plants and even in the *har1* hypernodulation mutant [81]. In *M. truncatula*, nodulation was strongly inhibited in a growth medium containing JA. A high JA concentration significantly decreased the plant's responsiveness to NFs, resulting in a lower number of root hairs that exhibited calcium spiking. In addition, JA inhibited the expression of the early rhizobium-responsive genes, *RIP1* and *ENOD11* [82]. In contrast, the JA concentration in leaves of a *G. max* hypernodulating *nts* mutant was higher than in those of wild-type *G. max* under natural growth conditions. In addition, transcription levels of JA responsive genes increased in the hypernodulating *nts* mutant, which suggests that the *nts* mutation induces changes in certain pathways, including JA synthetic metabolism, resulting in the activation of JA-responsive genes [83]. Kinkema and Gresshoff (2008) also showed that the expression of JA biosynthetic and responsive genes in the leaves of wild-type *G. max* is normally suppressed by inoculation with rhizobia, but not suppression was seen in a hypernodulating *nts* mutant. Furthermore, foliar application of n-propyl gallate, a JA biosynthesis inhibitor, significantly decreased nodulation specifically in the hypernodulating *nts* mutant [84]. Recently, Suzuki et al. (2011) reported that nodulation was enhanced by treatment with a low concentration of JA. Both infection thread formation and nodulation were increased by JA treatment of wild-type *L. japonicus* grown under low red/far-red (R/FR) light condition. These results indicate that nodulation is photomorphogenetically controlled by sensing the R/FR ratio through JA signaling [85]. Therefore, JA functions as a positive regulator of nodulation over a certain range of concentrations in these plant species. Previous studies have shown that JA can act as a signal molecule in the early stages of the development of leguminous plants-rhizobia symbioses; for example, it induces the expression of the *nod* genes in *B. japonicum* [86] and *Rhizobium* [87]. However, it is unclear whether the plant responses, rhizobial responses or combination of both responses are responsible for the positive effects of JA on nodulation. Suzuki et al. (2011) also found that nodulation was suppressed in the roots of a *phytochrome B* (*phyB*) mutant of *L. japonicus* that had not only decreased levels of photo-assimilates but also a reduced concentration of JA-Ile (the active JA derivative). In fact, the number of root nodules in the *phyB* mutant was restored by JA treatment, providing further evidence that JA can act as a positive regulator of nodulation in leguminous plants [85].

10. Strigolactones

Strigolactones (SLs) have been identified as phytohormones that are involved in the regulation of shoot branching in plants and thus have been suggested to be ubiquitous in the plant kingdom [88]. SLs are released by roots into the rhizosphere, and appear to stimulate germination of the seeds of parasitic plants such as *Striga* spp. and *Orobancha* spp. [89]. Recently, they have been shown to play key roles as signaling compounds in the interaction between plants and arbuscular mycorrhizal fungi [90]. Interestingly, SLs also increase nodulation in *M. sativa*, *P. sativum* and *L. japonicus* [91-93]. In *M. sativa*, treatment with the synthetic SL analogue GR24 clearly increased nodulation. When *M. sativa* plants were treated with GR24, the biosynthesis and the metabolism of the SLs increased, thus, resulting in an enhanced formation of indeterminate-type nodules [91]. The SL deficient *rms1* mutants of *P. sativum* also produce

fewer nodules than wild-type plants. Treatment with GR24 elevated nodule number in wild-type *P. sativum* and also elevated nodule number in *rms1* mutant to a level similar to that seen in untreated wild-type plants. These results indicated that endogenous SLs increase nodulation in *P. sativum* [92]. The role of SLs in *L. japonicus* was studied by using transgenic lines in which *CAROTENOID CLEAVAGE DIOXYGENASE 7* (*LjCCD7*), an orthologue of *Arabidopsis More Axillary Growth 3*, was silenced. Silencing of *LjCCD7* is expected to reduce SL levels. The plants with silenced *LjCCD7* produced fewer nodules than control plants; this suggests that SLs have a slight positive effect on the formation of determinate nodules [93].

Author details

Maki Nagata¹ and Akihiro Suzuki^{1,2*}

*Address all correspondence to: azuki@cc.saga-u.ac.jp

1 Department of Agricultural Sciences, Faculty of Agriculture, Saga University, Honjyo-machi, Saga, Saga, Japan

2 United Graduate School of Agricultural Sciences, Kagoshima University, Korimoto, Kagoshima, Kagoshima, Japan

References

- [1] Thimann KV (1936) On the physiology of the formation of nodules on legume roots. *Proc Natl Acad Sci USA* 22: 511-514
- [2] Hirsch AM, Bhuvaneswari TV, Torrey JG and Bisseling T (1989) Early nodulin genes are induced in alfalfa root outgrowths elicited by auxin transport inhibitors. *Proc Natl Acad Sci USA* 86: 1244-1248
- [3] Mathesius U, Schlaman HRM, Spaink HP, Sautter C, Rolfe BG and Djordjevic MA (1998) Auxin transport inhibition precedes root nodule formation in white clover roots and is regulated by flavonoids and derivatives of chitin oligosaccharides. *Plant J* 14: 23-34
- [4] Huo X, Schnabel E, Hughes K and Frugoli J (2006) RNAi phenotypes and the localization of a Protein::GUS Fusion imply a role for *Medicago truncatula* PIN genes in nodulation. *J Plant Growth Regul* 25: 156-165
- [5] van Noorden GE, Ross JJ, Reid JB, Rolfe BG and Mathesius U (2006) Defective long-distance auxin transport regulation in the *Medicago truncatula* super numeric nodules mutant. *Plant physiol* 140: 1494-1506

- [6] de Billy F, Grosjean C, May S, Bennett M and Cullimore JV (2001) Expression studies on AUX1-like genes in *Medicago truncatula* suggest that auxin is required at two steps in early nodule development. *Mol Plant Microbe Interact* 14:267-277
- [7] Takanashi K, Sugiyama A, Yazaki K (2011) Involvement of auxin distribution in root nodule development of *Lotus japonicus*. *Planta* 234: 73-81
- [8] Takanashi K, Sugiyama A, Yazaki K (2011) Auxin distribution and lenticel formation in determinate nodule of *Lotus japonicus*. *Plant Signal Behav* 6: 1405-1407
- [9] Pacios-Bras C, Schlaman HRM, Boot K, Admiraal P, Langerak JM, Stougaard J, Spaink HP (2003) Auxin distribution in *Lotus japonicus* during root nodule development. *Plant Mol Biol* 52: 1169-1180
- [10] Subramanian S, Stacey G and Yu O (2006) Endogenous isoflavones are essential for the establishment of symbiosis between soybean and *Bradyrhizobium japonicum*. *Plant J* 48: 261-273
- [11] Mathesius U (2008) Auxin: at the root of nodule development? *Funct Plant Biol* 35: 651-668
- [12] Suzaki T, Yano K, Ito M, Umehara Y, Suganuma N and Kawaguchi M (2012) Positive and negative regulation of cortical cell division during root nodule development in *Lotus japonicus* is accompanied by auxin response. *Development* 139: 3997-4006
- [13] Suzaki T, Ito M, Kawaguchi M (2013) Genetic basis of cytokinin and auxin functions during root nodule development. *Front Plant Sci* 4: 1-6
- [14] Arshad M, and Frankenberger WT (1993) Microbial production of plant growth regulators. In *Soil microbial ecology. Applications in agricultural and environmental management*. F.B. Metting, Jr. (edit.), Marcel Dekker, Inc., New York; 307-343
- [15] Mok MC (1994) Cytokinins and plant development: An overview. In *Cytokinins: Chemistry, Activity and Function*, D.W.S Mok and M.C. Mok, eds (Boca Raton, FL: CRC Press); 155-156.
- [16] Yahalom E, Okon Y, Dovrat A (1990) Possible mode of action of *Azospirillum brasilense* strain Cd on the root morphology and nodule formation in burr medic (*Medicago polymorpha*). *Can J Microbiol* 36: 10-14
- [17] Lorteau MA, Ferguson BJ and Guinel FC (2001) Effects of cytokinin on ethylene production and nodulation in pea (*Pisum sativum*) cv. Sparkle. *Physiol Plant* 112: 421-428
- [18] Cooper JB and Long SR (1994) Morphogenetic rescue of *Rhizobium meliloti* nodulation mutants by trans-Zeatin secretion. *Plant Cell* 6: 215-225
- [19] Fang Y and Hirsch AM (1998) Studying early nodulin gene ENOD40 expression and induction by nodulation factor and cytokinin in transgenic alfalfa. *Plant Physiol* 116: 53-68

- [20] Taller BJ, Sturtevant DB (1991) Cytokinin production by rhizobia. In Hennecke H, Verma DPS (eds) *Advances in Molecular Genetics of Plant-Microbe Interactions* 1: 215-221
- [21] Lohar DP, Schaff JE, Laskey JG, Kieber JJ, Bilyeu KD and Bird DM (2004) Cytokinins play opposite roles in lateral root formation, and nematode and rhizobial symbioses. *Plant J* 38: 203-214
- [22] Gonzalez-Rizzo S, Crespi M and Frugier F (2006) The *Medicago truncatula* CRE1 cytokinin receptor regulates lateral root development and early symbiotic interaction with *Sinorhizobium meliloti*. *Plant Cell* 18: 2680-2693
- [23] Murray JD, Karas BJ, Sato S, Tabata S, Amyot L, Szczyglowski K (2007) A cytokinin perception mutant colonized by *Rhizobium* in the absence of nodule organogenesis. *Science* 315: 101-104
- [24] Tirichine L, Sandal N, Madsen LH, Radutoiu S, Albrechtsen AS, Sato S, Asamizu E, Tabata S, Stougaard J (2007) A gain-of-function mutation in a cytokinin receptor triggers spontaneous root nodule organogenesis. *Science* 315: 104-107
- [25] Fatima Z, Bano A, Sial R and Aslam M (2008) Response of chickpea to plant growth regulators on nitrogen fixation and yield. *Pak. J. Bot* 40: 2005-2008
- [26] Rao LVM, Datta N, Mahadevan M, Guha-Mukherjee S and Sopory SK (1984) Influence of cytokinins and phytochrome on nitrate reductase activity in etiolated leaves of maize. *Phytochemistry* 23: 1875-1879
- [27] Hooley R (1994) Gibberellins: perception, transduction and responses. *Plant Mol Biol* 26: 1529-1555
- [28] Ferguson BJ, Ross JJ and Reid JB (2005) Nodulation phenotypes of gibberellin and brassinosteroid mutants of pea. *Plant physiol* 138: 2396-2405
- [29] Lievens S, Goormachtig S, Herder JD, Capoen W, Mathis R, Hedden P, Holsters M (2005) Gibberellins are involved in nodulation of *Sesbania rostrata*. *Plant Physiol* 139: 1366-1379
- [30] Kawaguchi M, Imaizumi-Anraku H, Fukai S, Syōno K (1996) Unusual branching in the seedling of *Lotus japonicus*-gibberellins reveal the nitrogen-sensitive cell divisions within the pericycle on roots. *Plant Cell Physiol* 37: 461-470
- [31] Maekawa T, Maekawa-Yoshikawa M, Takeda N, Imaizumi-Anraku H, Murooka Y and Hayashi M (2009) Gibberellin controls the nodulation signaling pathway in *Lotus japonicus*. *Plant J* 58: 183-194
- [32] McGinnis KM, Thomas SG, Soule JD, Strader LC, Zale JM, Sun T and Steber CM (2003) The *Arabidopsis* SLEEPY1 gene encodes a putative F-box subunit of an SCF E3 ubiquitin ligase. *Plant Cell* 15: 1120-1130

- [33] Dill A, Thomas SG, Hu J, Steber CM, Sun T (2004) The Arabidopsis F-box protein SLEEPY1 targets gibberellin signaling repressors for gibberellin-induced degradation. *Plant Cell* 16: 1392-1405
- [34] Ferguson BJ, Foo E, Ross JJ and Reid JB (2011) Relationship between gibberellin, ethylene and nodulation in *Pisum sativum*. *New Phytologist* 189: 829-842
- [35] Williams PM and Sicardi de Mallorca M (1984) Effect of gibberellins and the growth retardant CCC on the nodulation of soya. *Plant Soil* 77: 53-60
- [36] Zhang F, Pan B and Smith DL (1997) Application of gibberellic acid to the surface of soybean seed (*Glycine max* (L.) Merr.) and symbiotic nodulation, plant development, final grain and protein yield under short season conditions. *Plant Soil* 188: 329-335
- [37] Ligeró F, Lluch C, Olivares J (1987) Evolution of ethylene from roots and nodulation rate of alfalfa (*Medicago sativa* L.) plants inoculated with *Rhizobium meliloti* as affected by the presence of nitrate. *J Plant Physiol* 129: 461-467
- [38] Lee KH and LaRue TA (1992) Exogenous ethylene inhibits nodulation of *Pisum sativum* L. cv Sparkle. *Plant Physiol* 100: 1759-1763
- [39] Oldroyd GED, Engstrom EM and Long SR (2001) Ethylene inhibits the Nod factor signal transduction pathway of *Medicago truncatula*. *Plant Cell* 13: 1835-1849
- [40] Peters NK and Crist-Estes DK (1989) Nodule formation is stimulated by the ethylene inhibitor aminoethoxyvinylglycine. *Plant physiol* 91: 690-693
- [41] Caba JM, Recalde L and Ligeró F (1998) Nitrate-induced ethylene biosynthesis and the control of nodulation in alfalfa. *Plant Cell Environ* 21: 87-93
- [42] Penmetsa RV and Cook DR (1997) A legume ethylene-insensitive mutant hyperinfected by its rhizobial symbiont. *Science* 275: 527-530
- [43] Penmetsa RV, Uribe P, Anderson J, Lichtenzweig J, Gish JC, Nam YW, Engstrom E, Xu K, Sckisel G, Pereira M, Baek JM, Lopez-Meyer M, Long SR, Harrison MJ, Singh KB, Kiss GB, Cook DR (2008) The *Medicago truncatula* ortholog of Arabidopsis EIN2, sickle, is a negative regulator of symbiotic and pathogenic microbial associations. *Plant J* 55: 580-595
- [44] Nukui N, Ezura H, Yuhashi K, Yasuta T and Minamisawa K (2000) Effects of ethylene precursor and inhibitors for ethylene biosynthesis and perception on nodulation in *Lotus japonicus* and *Macroptilium atropurpureum*. *Plant Cell Physiol* 41: 893-897
- [45] Nukui N, Ezura H, Minamisawa K (2004) Transgenic *Lotus japonicus* with an ethylene receptor gene Cm-ERS1/H70A enhances formation of infection threads and nodule primordia. *Plant Cell Physiol* 45:427-435
- [46] Tirichine L, James EK, Sandal N and Stougaard J (2006) Spontaneous root-nodule formation in the model legume *Lotus japonicus*: A novel class of mutants nodulates in the absence of rhizobia. *Mol Plant Microbe Interact* 19: 373-382

- [47] Hunter WJ (1993) Ethylene production by root nodules and effect of ethylene on nodulation in *Glycine max*. *Appl Environ Microbiol* 59: 1947-1950
- [48] Suganuma N, Yamauchi H, Yamamoto K (1995) Enhanced production of ethylene by soybean roots after inoculation with *Bradyrhizobium japonicum*. *Plant Sci* 111: 163-168
- [49] Schmidt JS, Harper JE, Hoffman TK and Bent AF (1999) Regulation of soybean nodulation independent of ethylene signaling. *Plant physiol* 119: 951-959
- [50] D'Haeze W, Rycke RD, Mathis R, Goormachtig S, Pagnotta S, Verplancke C, Capoen W and Holsters M (2003) Reactive oxygen species and ethylene play a positive role in lateral root base nodulation of a semiaquatic legume. *Proc Natl Acad Sci USA* 100: 11789-11794
- [51] Honma M and Shimomura T (1978) Metabolism of 1-aminocyclopropane-1-carboxylic acid. *Agric Biol Chem* 42: 1825-1831
- [52] Blaha D, Prigent-Combaret C, Mirza MS and Moënne-Loccoz (2006) Phylogeny of the 1-aminocyclopropane-1-carboxylic acid deaminase-encoding gene *acdS* in phytobeneficial and pathogenic Proteobacteria and relation with strain biogeography. *FEMS Microbiol Ecol* 56: 455-470
- [53] Uchiumi T, Ohwada T, Itakura M, Mitsui H, Nukui N, Dawadi P, Kaneko T, Tabata S, Yokoyama T, Tejima K, Saeki K, Omori H, Hayashi M, Maekawa T, Sriprang R, Murooka Y, Tajima S, Simomura K, Nomura M, Suzuki A, Shimoda Y, Sioya K, Abe M, Minamisawa K (2004) Expression islands clustered on the symbiosis island of the *Mesorhizobium loti* genome. *J Bacteriol* 186: 2439-2448
- [54] Ma W, Guinel FC and Glick BR (2003) *Rhizobium leguminosarum* biovar *viciae* 1-aminocyclopropane-1-carboxylate deaminase promotes nodulation of pea plants. *Appl Environ Microbiol* 69: 4396-4402
- [55] Ma W, Penrose DM, Glick BR (2002) Strategies used by rhizobia to lower plant ethylene levels and increase nodulation. *Can J Microbiol* 48: 947-954
- [56] Takatsuto S, Yazawa N, Ikekawa N, Takematsu T, Takeuchi Y and Koguchi M (1983) Structure activity relationship of brassinosteroids. *Phytochemistry* 22: 2437-2441
- [57] Vardhini BV and Rao SSR (1999) Effect of brassinosteroids on nodulation and nitrogenase activity in groundnut (*Arachis hypogaea* L.). *Plant Growth Regul* 28: 165-167
- [58] Terakado J, Fujihara S, Goto S, Kuratani R, Suzuki Y, Yoshida S and Yoneyama T (2005) Systemic effect of a brassinosteroid on root nodule formation in soybean as revealed by the application of brassinolide and brassinazole. *Soil Sci Plant Nutr* 51: 389-395
- [59] Shahid MA, Pervez MA, Balal RM, Mattson NS, Rashid A, Ahmad R, Ayyub CM and Abbas T (2011) Brassinosteroid (24-epibrassinolide) enhances growth and alleviates

- the deleterious effects induced by salt stress in pea (*Pisum sativum* L.). *Aust J Crop Sci* 5: 500-510
- [60] Vardhini BV and Rao SSR (1998) Effect of brassinosteroids on growth, metabolite content and yield of *Arachis hypogaea*. *Phytochem* 48: 927-930
- [61] Nomura T, Nakayama M, Reid JB, Takeuchi Y, Yokota T (1997) Blockage of brassinosteroid biosynthesis and sensitivity causes dwarfism in garden pea. *Plant Physiol* 113: 31-37
- [62] Nomura T, Jager CE, Kitasaka Y, Takeuchi K, Fukami M, Yoneyama K, Matsushita Y, Nyunoya H, Takatsuto S, Fujioka S, Smith JJ, Kerckhoffs LHJ, Reid JB, Yokota T (2004) Brassinosteroid deficiency due to truncated steroid 5 α -reductase causes dwarfism in the lk mutant of pea. *Plant Physiol* 135: 2220-2229
- [63] Symons GM and Reid JB (2004) Brassinosteroids do not undergo long distance transport in pea. Implications for the regulation of endogenous brassinosteroid levels. *Plant Physiol* 135: 2196-2206
- [64] Phillips, DA (1971) Absciscic acid inhibition of root nodule initiation in *Pisum sativum*. *Planta* 100:181-190
- [65] Cho M-J, Harper JE (1993) Effect of abscisic acid application on root isoflavonoid concentration and nodulation of wild-type and nodulation- mutant soybean plants. *Plant and Soil* 152:145-149
- [66] Bano A, Harper JE (2002) Plant growth regulators and phloem exudates modulate root nodulation of soybean. *Funct Plant Biol* 29:1299-1307
- [67] Suzuki A, Akune M, Kogiso M, Imagama Y, Osuki K, Uchiumi T, Higashi S, Han SY, Yoshida S, Asami T, Abe M (2004) Control of nodule number by the phytohormone abscisic acid in the roots of two leguminous species. *Plant Cell Physiol* 45:914-922
- [68] Ding Y, Kalo P, Yendrek C, Sun J, Liang Y, Marsh JF, Harris JM, Oldroyd GED (2008) Absciscic acid coordinates Nod factor and cytokinin signaling during the regulation of nodulation in *Medicago truncatula*. *Plant Cell* 20:2681-2695
- [69] Nakatsukasa-Akune M, Yamashita K, Shimoda Y, Uchiumi T, Abe M, Aoki T, Kamizawa A, Ayabe S, Higashi S, Suzuki A (2005) Suppression of root nodule formation by artificial expression of the TrEnodDR1 (coat protein of white clover cryptic virus 1) gene in *Lotus japonicus*. *Mol Plant Microbe Interact* 18:1069-1080
- [70] Khadri M, Tejera NA and Lluch C (2006) Alleviation of salt stress in common bean (*Phaseolus vulgaris*) by exogenous abscisic acid supply. *J Plant Growth Regul* 25: 110-119
- [71] González EM, Galvez L, Arrese-Igor C (2001) Absciscic acid induces a decline in nitrogen fixation that involves leghemoglobin, but is independent of sucrose synthase activity. *J Exp Bot* 52: 285-293

- [72] Tominaga A, Nagata M, Futsuki K, Abe H, Uchiumi T, Abe M, Kucho K, Hashiguchi M, Akashi R, Hirsch AM, Arima S, Suzuki A (2009) Enhanced nodulation and nitrogen fixation in the abscisic acid low-sensitive mutant enhanced nitrogen fixation1 of *Lotus japonicus*. *Plant physiol* 151: 1965-1976
- [73] Goicoechea N, Antolín MC, Sánchez-Díaz M (1997) Gas exchange is related to hormone balance in mycorrhizal or nitrogen-fixing alfalfa subjected to drought. *Physiol Plant* 100: 989-997
- [74] Biswas B, Chan PK, Gresshoff PM (2009) A novel ABA insensitive mutant of *Lotus japonicus* with a wilted phenotype displays unaltered nodulation regulation. *Mol Plant* 2: 487-499
- [75] Martínez-Abarca F, Herrera-Cervera JA, Bueno P, Sanjuan J, Bisseling T, Olivares J (1998) Involvement of salicylic acid in the establishment of the *Rhizobium meliloti*-alfalfa symbiosis. *Mol Plant Microbe Interact* 11:153-155
- [76] Blilou I, Ocampo JA, García-Garrido JM (1999) Resistance of pea roots to endomycorrhizal fungus or *Rhizobium* correlates with enhanced levels of endogenous salicylic acid. *J Exp Bot* 50: 1663-1668
- [77] van Spronsen PC, Tak T, Rood AM, van Brussel AA, Kijne JW, Boot KJ (2003) Salicylic acid inhibits indeterminate-type nodulation but not determinate-type nodulation. *Mol Plant Microbe Interact* 16: 83-91
- [78] Lian B, Zhou X, Miransari M, Smith DL (2000) Effects of salicylic acid on the development and root nodulation of soybean seedlings. *J Agron Crop Sci* 185: 187-192
- [79] Sato T, Fujikake H, Ohtake N, Sueyoshi K, Takahashi T, Sato A and Ohshima T (2002) Effect of exogenous salicylic acid supply on nodule formation of hypernodulating mutant and wild type of soybean. *Soil Sci Plant Nutr* 48: 413-420
- [80] Stacey G, McAlvin CB, Kim SY, Olivares J, Soto MJ (2006) Effects of endogenous salicylic acid on nodulation in the model legumes *Lotus japonicus* and *Medicago truncatula*. *Plant physiol* 141: 1473-1481
- [81] Nakagawa T and Kawaguchi M (2006) Shoot-applied MeJA suppresses root nodulation in *Lotus japonicus*. *Plant Cell Physiol* 47: 176-180
- [82] Sun J, Cardoza V, Mitchell DM, Bright L, Oldroyd G, Harris JM (2006) Crosstalk between jasmonic acid, ethylene and Nod factor signaling allow integration of diverse inputs for regulation of nodulation. *Plant J* 46: 961-970
- [83] Seo HS, Li J, Lee SY, Yu JW, Kim KH, Lee SH, Lee IJ, Paek NC (2006) The hypernodulating mutation induces jasmonate synthetic pathway in soybean leaves. *Mol Cells* 24: 185-193

- [84] Kinkema M and Gresshoff PM (2008) Investigation of downstream signals of the soybean autoregulation of nodulation receptor kinase GmNARK. *Mol Plant Microbe Interact* 21:1337-1348
- [85] Suzuki A, Suriyagoda L, Shigeyama T, Tominaga A, Sasaki M, Hiratsuka Y, Yoshinaga A, Arima S, Agarie S, Sakai T, Inada S, Jikumaru Y, kamiya Y, Uchiumi T, Abe M, Hashiguchi M, Akashi R, Sato S, Kaneko T, Tabata S, Hirsch AM (2011) *Lotus japonicus* nodulation is photomorphogenetically controlled by sensing the red/far red (R/FR) ratio through jasmonic acid (JA) signaling. *Proc Natl Acad Sci USA* 108: 16837-16842
- [86] Mabood F, Smith DL (2005) Pre-inoculation of *Bradyrhizobium japonicum* with jasmonates accelerates nodulation and nitrogen fixation in soybean (*Glycine max*) at optimal and suboptimal root zone temperatures. *Physiol Plant* 125: 311-323
- [87] Rosas S, Soria R, Correa N, Abdala G (1998) Jasmonic acid stimulates the expression of nod genes in *Rhizobium*. *Plant Mol Biol* 38: 1161-1168
- [88] Umehara M, Hanada A, Yoshida S, Akiyama K, Arita T, Takeda-Kamiya N, Magome H, Kamiya Y, Shirasu K, Yoneyama K, Kyoizuka J, Yamaguchi S (2008) Inhibition of shoot branching by new terpenoid plant hormones. *Nature* 455: 195-200
- [89] Bouwmeester HJ, Roux C, López-Ráez JA, Bécard G (2007) Rhizosphere communication of plants, parasitic plants and AM fungi. *Trends Plant Sci* 12: 224-230
- [90] Akiyama K, Matsuzaki K, Hayashi H (2005) Plant sesquiterpenes induce hyphal branching in arbuscular mycorrhizal fungi. *Nature* 435: 824-827
- [91] Soto MJ, Fernández-Aparicio M, Castellanos-Morales V, García-Garrido JM, Ocampo JA, Delgado MJ, Vierheilig H (2010) First indications for the involvement of strigolactones on nodule formation in alfalfa (*Medicago sativa*). *Soil Biol Biochem* 42: 383-385
- [92] Foo E and Davies NW (2011) Strigolactones promote nodulation in pea. *Planta* 234: 1073-1081
- [93] Liu J, Novero M, Charnikhova T, Ferrandino A, Schubert A, Ruyter-Spira C, Bonfante P, Lovisolo C, Bouwmeester H, Cardinale F (2013) CAROTENOID CLEAVAGE DIOXYGENASE 7 modulates plant growth, reproduction, senescence, and determinate nodulation in the model *Lotus japonicus*. *J Exp Bot* 64: 1967-1981