Cultivos Tropicales, 2022, vol. 43, no. 2, e13
ISSN impreso: 0258-5936
ISSN digital: 1819-4087

http://ediciones.inca.edu.cu

abril-junio
Ministerio de Educación Superior. Cuba
Instituto Nacional de Ciencias Agrícolas

Review

Mycorrhizae and rhizobia: a molecular dialogue with the plant host

Andrés Zúñiga-Orozco[1*]

Ayerin Carrodeguas- Gonzalez[2]

Laura Yesenia Solís-Ramos[3]

[1]Escuela de Ciencias Exactas y Naturales, Universidad Estatal a Distancia. San Pedro, San José, Costa Rica

[2]Instituto de Investigaciones Hortícolas Lilliana Dimitrova, Mayabeque, Cuba.

[3]Escuela de Biología, Universidad de Costa Rica. San Pedro, San José, Costa Rica

[*]Author for correspondence: azunigao@uned.ac.cr

ABSTRACT

Rhizobia (Rhs) and arbuscular mycorrhizal fungi (AMF) are soil microsymbionts associated with crop roots. For Rhs most of them are legumes and for AMF there is a wider host range; however, there are crops that develop colonization by both symbionts. In any of the symbiotic relationships, crops receive benefits when colonized by these microorganisms are varied and contribute to the use of alternatives for sustainable agriculture. The mechanism by which both symbionts penetrate their plant host has been studied at the molecular level and common genes have been identified, as well as the pathways in which they are involved. Some of these genes are related to the reception of the signal mediated by Nod factors in the case of Rhs and by Myc factors in the case of AMF, others are related to the penetration mechanism and finally to the route by which the symbiont and the plant communicate. In the present review, a list of studies concerning microsymbionts, at the level of pre-colonization, colonization and shared mechanism is made. A proposal of possible common candidate genes for Rhs and AMF to apply genetic engineering is presented, in such a way that a research field called: gene optimization is explored. Because of the similarity by which these symbionts penetrate their host and the potential for genetic modification that this implies, a close molecular, metabolic and physiological relationship is described.

Key words: rhizobium arbuscular mycorrhizae, genetic engineering, symbiosis, genes.

INTRODUCTION

The creation of sustainable bioeconomies framed in the concept of circular economy demands the optimization of biological resources to improve agricultural productivity. The findings found during the last 30 years,

regarding symbiosis, the use of arbuscular mycorrhizal fungi (AMF) and rhizobia (Rhs), have been of great interest due to the impact they have on agriculture [1-3].

Symbiosis is the close mutualistic relationship between two organisms, which has a beneficial effect on adaptation, ecology, and evolution for both parties [4-6]. Among the most interesting mutualistic symbiotic relationships are those between fungi, bacteria, and plant cells [7]. AMF and Rhs originated approximately 400 and 100 million years ago, respectively [8-12].

AMF belong to the phyla *Glomeromycota* and are organisms that colonize between 70-90 % of plant species; some authors mention that they colonize all gymnosperms, 83 % of dicotyledons and 79 % of monocotyledons [12,13]; while Rhs are more restricted to the FaFaCuRo clade (Fabales, Fagales, *Cucurbitaceae* and Roses) [14-16].

AMF have received special attention from the agricultural point of view due to the benefits they provide to plants, such as greater resistance to biotic and abiotic stresses, increased surface area for water and nutrient absorption [11,15], as well as their use in bioremediation [17], among others. Rhizobia are also very important for their ability to fix atmospheric nitrogen, and there are even studies that mention that they could fix the annual amount of synthetic ammonium produced [1,18].

Currently, the natural use of nitrogen-fixing bacteria symbiosis in leguminous plants has been promoted to reduce the amount of nitrogen applied through chemical fertilizers, which can cause eutrophication and decrease the diversity of soil microorganisms [3,19].

AMF penetrate the host through the cortex of the parenchyma of finest roots and in the interior of cells they form branched structures called arbuscules [20]; while Rhs penetrate their host through the root hairs, folding them until an infection tube is formed in which the symbiosome develops in the internal end [21,22]. The colonization mechanisms of AMF and Rhs are very similar, even to the extent of activating and deactivating common genes. For their study, species of the legume family have been used, which can host both microsymbionts and as a result accumulate greater dry mass and have a larger root surface for nutrient absorption [23].

There is clear evidence for the existence of a shared mechanism in shared colonization that induces a type of self-regulation between the microsymbionts and occurs in constant communication with the plant [24].

Through studies of the composition of microorganism communities, it has been shown that diverse habitats are capable of harboring a great biological diversity of AMF and Rhs [25], which have applications in agriculture. In addition, a breakthrough has recently been made in sequencing and gene expression profiling, which has elucidated commonalities in the shared colonization of AMF and Rhs [26-30].

Taking into account the criteria written above, this article is a compilation of a series of scientific evidences that reflect the similarity of the behavior in the colonization process carried out by AMF and Rhs, in response to signals of the plant host. In this way, this compilation of information is considered a bridge to define new lines of research in genetic engineering; therefore, contributing to the state of knowledge regarding candidate genes that could be modified in the future, making the relationship between AMF and Rhs and their plant hosts more efficient.

The microsymbiont: the rhizobiums

The bacteria that are part of the soil microbiota are mainly included in the following phyla: *Acidobacteria*, *Actinobacteria*, *Bacteroidetes*, *Chloroflexi* (*Chlorobacteria*), *Firmicutes* and *Proteobacteria* [31,32]. As roots grow, they incorporate organic deposits (dead cells + organic compounds) at the rhizosphere level, resulting in rhizodeposition that modifies the structure and population composition of bacteria. Consequently, the diversity of the phyla *Acidobacteria*, *Proteobacteria* and *Actinobacteria* is reduced [32].

Bacteria are attracted to their host by cells of the rhizodermis, rhizodeposits and the mucilage that is exuded at the end of the roots. This mucilage is composed of organic and inorganic acids, siderophores, vitamins, amino acids, purines and nucleosides, but there are some compounds in particular: flavonoids and isoflavonoids, which are mainly responsible for attracting bacteria to the plant [33,34]. Upon sensing the signal, Rhs begin to secrete a lipo-chito-oligosaccharide (LCO) mediated by Nod factors (NF) [35-37]. In the case of Rhs, LCO interacts with host-emitted hydrolases NFH1 and CHIT5 to prepare for contact with the plant cell membrane [38].

For their part, Rhs colonize the plant mainly by a tubular infection that forms after the root hairs fold into a loop, although to a lesser extent, they can also gain access through wounds, or through intercellular spaces independently of *Nod* factors [31].

Therefore, the colonization process of Rhs in their host goes through several progressive stages, ranging from initial signaling, host range restriction, bacterial colonization, autoregulation of nodule number (AON), bacterial maturation, symbiosome formation, development of nodule metabolism, and transport until the final phase of senescence begins [18,38].

Proper bacteria-host chemical communication depends on Nod factors and proper coupling with plant membrane receptors. This process is key to trigger colonization, which subsequently results in a change in the calcium gradient in the nuclear membrane of the plant cell, which will be explained later.

The microsymbiont: the arbuscular mycorrhizal fungi

AMF are organisms that initiate their life cycle from a propagule that can be a spore, a fragment of hyphae or a colonized root. The propagule germinates stimulated by signals derived from the potential host, although it can also germinate in the absence of these. A germinating hyphae is produced and begins to grow in search of a host, and when it finds it, it adheres to the cortex walls of the finest roots (secondary or tertiary) producing a support structure called an appressorium. Subsequently, it penetrates the interior of the cortex without crossing the central cylinder and colonizes the plant intracellularly and extracellularly. Upon accessing the cells, it crosses the cell wall, but not the plasma membrane, a retraction of the cytoplasm occurs and the hyphae begin to branch to form arbuscules, which are the exchange structures between the fungus and its host.

In most AMF species vesicles are produced which are reserve structures [39]; however, vesicles are not present in all AMF species, some such as those belonging to the *Gigasporacea* family may instead form structures called auxiliary cells, but in the external mycelium and are recognized to have similar function to vesicles.

AMF are obligate symbionts and therefore require a host to complete their life cycle. When they find one, they reproduce rapidly and it is even known that they share several hosts at the same time through a connective network of extraradical mycelium, so their functional diversity is high and allows them to have great adaptability to diverse environmental conditions [40,41].

The fungus invaginates the internal cortical cells, where it produces an extensive ramification becoming a structure that entirely fills the cortical cells [42]. Consequently, the architecture of the host cell changes: the nucleus moves from a peripheral to a central position, the vacuole begins to fragment, and an extensive periarbuscular membrane is synthesized continuously to the plasma membrane [43]. Despite the intense activity of both symbionts allowing the formation of arbuscules in the cells, they collapse after several days, leaving the cortical cell intact and ready to host a new arbuscule [42].

There are several situations that occur in the symbiont-plant relationship, one of them is when the host has sufficient availability of nutrients, in this case, the vesicles store carbon structures as a means of survival being very similar in function to the polyhydroxybutyrate granules that are present in the Rhs [44,45]. Such is the case of phosphorus, since the plant inactivates phosphorus transporter genes when there is a high availability of this nutrient [46].

On the other hand, it has been found that arbuscules provide a greater amount of phosphorus to tissues that provide a greater amount of carbon. The above described indicates that there may be a self-regulation mechanism on the part of the plant. It has been found that molecules such as lysophosphatidylcholine (LPc) could help the host to perceive the concentration of phosphorus in the soil; however, further research is needed in this regard, since it is not well described in comparison to what happens with Rhs [47].

Another of the less explored characteristics of AMF is that they have the ability to protect host roots from pathogenic hyphae, since they grow about 100 times faster than root hairs, which allows them to colonize the root area more rapidly. Such is the case of the *Glomeraceae* family, whose species have shown high tolerance to *Fusarium* sp. and *Pythium* sp. [46], it is also possible that there is a molecular mechanism of symbiotic interaction still unexplored. This is in addition to many other studies focused on quantifying the tolerance to diverse pathogens such as: *Alternaria*, *Fusarium*, *Phytophthora*, *Pythium*, *Rhizoctonia* and *Verticillium*, bacteria such as *Ralstonia solanacearum* and *Pseudomonas syringae*, nematodes of the genera *Pratylenchus* and *Meloidogyne*) and even insects such as *Otiorhynchus sulcatus* [48-52].

Regarding the AMF colonization in their host, it is important to point out that their propagules are stimulated by flavonoids and isoflavonoids from plants, as is the case with Rhs; however, AMF are also stimulated by sesquiterpenes such as strigolactones, which stimulate the branching of germinating hyphae [53]. Flavonoids and isoflavonoids secreted by plants activate the germination process and hyphal growth; and for their part, AMF begin to secrete a lipo-chito-oligosaccharide (LCO) mediated by Myc factors [35-37] and a short-chain chitin oligomer or chito-oligosaccharide (COS) [54].

In response to Myc factors, ENOD11 gene has been reported in plants as responsible for encoding lysine-rich proteins in the membrane, thus, in a way, plants are also primed to receive AMF [55]. In addition, as will be discussed later, ENOD gene has some relationship with Rhs.

Other genes have been identified as necessary to induce the formation of the perihaustorial membrane appressorium such as the DMI2 and DMI3 genes [18]. Similarly, an orthologous gene related to penetration, called STE12, has been found in pathogenic fungi (*Magnaphorte oryzae* and *Colletotrichum inemuthianum*) [56].

Finally, plant genes coding for phosphorus transporters (PT3 and PT4) are known to be responsible for associating with hyphae, in addition to a gene called Gmar-CuZnSOD coding for superoxide dismutase, which provides the plant with tolerance to oxidative stress [57].

The macrosymbiont: the plant host and the mechanism of colonization

In this section, a general account of what has been described for the mechanism of colonization by Rhs will be made, since, although it has been described for AMF, studies related to genes involved and their regulation are still lacking. However, in the next section, similarities between both colonization pathways will be addressed.

The role played by the plant host is crucial in the attraction of Rhs and AMF, as well as in the acceptance and maintenance of the microorganisms, since the symbionts benefit from carbon sources produced by the plant, mainly sucrose, hexoses, and starch, in a kind of "mutual trade" between Rhs-AMF-plant [38,39,42,58].

First, it is important to describe how the plant carries out the process of attracting the bacteria, giving way to the pre-colonization or presymbiotic phase; in this step the CHS, CHR, FNS and IFS genes are related, which are responsible for producing flavonoids and isoflavonoids by the plant [59-61]. Some researchers show evidence that the isoflavones genistein and diadzein produced by *Glycine max* and *Phaseolus vulgaris* induce the activation of Nod genes in very species-specific symbiont bacteria, such as *Bradyrhizobium japonicum* and *Rhizobium leguminosarum* bv phaseoli [62].

Once the plant perceives the Nod factors in the membrane, genes are activated that induce the degradation of the cell wall and the folding of the root hairs so that the Rhs can lodge and the colonization tube is formed; the following genes have been identified in this process: NPL, FLOT2, FLOT4 and SYMREM1 [63-65].

After the stimulus of the Nod genes and the folding of root hairs, the plant host perceives the signal in the membrane receptors. These receptors are encoded by the genes of orthologous pairs LjNFR1/MtLYK3 and LjNFR5/MtNFP, which are receptor kinases with three extracellular domains of lysine (LysM) in which they form a homo and heteromeric complex between the cell membrane and the colonization membrane [66-68]. According to the above, communication with the factors coming from the bacteria takes place through the LCO complex, in addition to the union with the NFH1/CHIT5 hydrolases; this interaction is the entry point to trigger an internal signaling complex in the host, which initiates the infection of Rhs and in a second step to organogenesis.

In addition, related to the perception of the signal, it has been recorded that plant hormones have an important role, since they interact with Nod factors and subsequent processes. Such is the case of brassinosteroids with the BRI1 gene and strigolactones with the CCD7 gene, which collaborate in the progression of colonization by rhizobia, as well as cytokinins and auxins that initiate the process of organogenesis. Cytokinins are directly related to the TF NSP2 and auxins to the ARF16a gene, responsible for the positive regulation of the colonization process [69-72]. Other very important genes that have been shown to reduce colonization and the amount of nodules when silenced are those related to gibberellins, these genes encode for DELLA proteins and these proteins, in turn, interact with the TFs IPD3 and NSP2, necessary for the transcription of NIN (nodule initiation) [69].

After the reception of the signal in the plant membrane, a calcium gradient is induced in the nuclear membrane, which reduces the cation entry potential. It has been reported that calcium channels (LjCASTOR, LjPOLLUX/MtDMI1), MtCNGC a/b/c and its orthologue LjBRUSH, as well as nucleoporins (LjNUP85 and LjNUP133) are affected. This leads to the MtDMI3/CCaMK kinase and the transcription factors LjCYCLOPS/MtIPD3 regulating the positive expression of the NIN gene, in conjunction with two other transcription factors called NSP1 and NSP2, found in *M. truncatula* [22,73,74]. NIN, together with the NF-YA and NF-YB genes, are of great importance because they are the starting signal for organogenesis and nodule proliferation to begin [38].

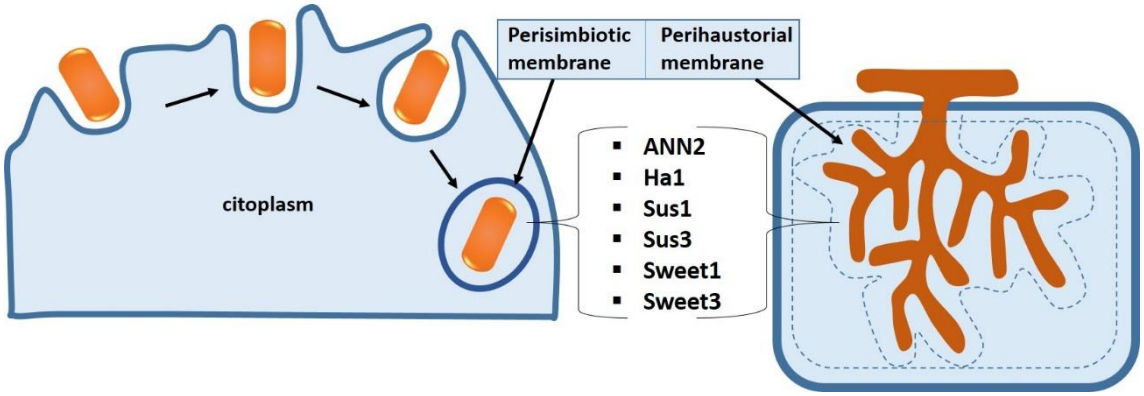
The plant has mechanisms to accept symbionts, but also emits a slight defense reaction trying to reject them [75], this reaction has been found to be very similar to the one that occurs before a pathogenic affectation and, consequently, many common genes are activated. Such is the case of *Sinorhizobium meliloti* that has the capacity to induce genes in the host, similar to those activated by the plant when it is attacked by *Pseudomonas syringae* [26]. What happens at the molecular level is that complexes of defense kinase receptor, such as LRR-RLKs and LysM-RLKs, recognize Rhs molecules while producing NBS-LRR-like proteins to neutralize the bacteria [76]. Some genes in the NBS-LRR group, such as Rj2, Rfg1, and Rj4 are associated with host restriction to a range of bacteria, because they code for family five pathogenesis-associated proteins [77,78].

In the case of Rhs, nodules cannot grow indefinitely, which is why the host tries to regulate the amount and timing of colonization. At least five main endogenous and exogenous factors that control nodulation have been identified, which, in one way or another, are related. In the first instance, there is a mechanism specific to the host called "autoregulation system (AON)", mediated by a signaling complex induced by bacteria. Also the amount of nitrogen available in the soil can influence, the presence of ethylene in the rhizosphere, the pH of the soil (mainly acid) and several biotic/abiotic factors, which can cause stress for the host plant and, consequently, produce fewer nodules by a reduction effect of carbon sources in the sink [58]. In the case of the AON mechanism, the CLE1 and CLE2 peptides in *M. truncatula*, their counterparts RIC 1 and RIC 2 in *G. max* and *P. vulgaris* or CLE-RS1 and CLE-RS2 in *Lotus japonicus*, are responsible for sending a signal to the plant stem to regulate the number of nodules. In this plant organ, a receptor complex is formed with the peptide to trigger a signal that is sent back to the roots to curb the number of symbiotic nodules [79-82].

Cultivos Tropicales, 2022, vol. 43, no. 2, e13
ISSN impreso: 0258-5936
ISSN digital: 1819-4087
http://ediciones.inca.edu.cu
abril-junio
Ministerio de Educación Superior. Cuba
Instituto Nacional de Ciencias Agrícolas

The shared colonization mechanism

To understand the process of colonization, development and reproduction of Rhs, we initially worked with mutants that nodulated little or not at all. In these experiments, the idea was to evaluate the behavior of the plant host in the absence or expression of one or several genes [83]. In this type of experiment, the activity and colonization capacity of AMF were affected. This led the researchers to think that, in some way, the mechanism by which microsymbionts made their way was more than anatomical-physiological and their origin could be found at the molecular level [83-85]. It was to be expected to find common genes since these symbiont microorganisms share a very similar mechanism of entry into the plant in the state prior to entering their host, which is separated by a highly specialized perisymbiotic membrane [86]. In Rhs a structure called the symbiosome is formed [87] and in AMF it is known as the perihaustorial membrane, which surrounds the arbuscules [18]. It is through these membranes that nutrient exchange with the host occurs [88,89]. The HA1 and ANN2 genes have been found in the model plant *Medicago truncatula* to be responsible for acidifying the perisymbiotic and perihaustorial membrane, probably to facilitate cross transport between host-symbiont in the case of HA1 and ANN2 as an inducer of the nodulation primordium, as well as cells containing arbuscules [90] (Figure 1).



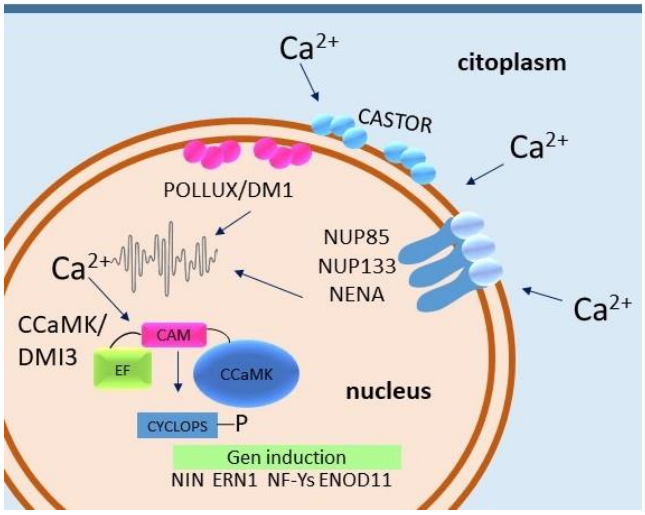
Authors' own creation

Figure 1. Responsible genes for acidifying the perisymbiotic and perihaustorial membrane (ANN2 and Ha1) and responsible genes for transporting glucose (Sweet1 and Sweet3) and hydrolyzing sucrose (Sus1 and Sus3)

Regarding the transport of carbon sources from the host to the symbionts, it has been found in *Medicago truncatula* that Sus1 and Sus3 are genes present in both AMF and Rhs, which encode for sucrose synthase, whose responsibility is to hydrolyze sources such as sucrose and starch [91]. In the case of AMF, the SWEET1b gene is expressed, which encodes for a glucose transporter from the host to the periarbuscular membrane, which becomes an important factor for intraradical mycelial growth as well as bacterial proliferation (Figure 1). SWEET genes have even been identified in pathogenic fungi and bacteria [92].

Although the mechanism by which AMF and Rhs structures develop are distinct, in the initial process of perception, colonization, and the subsequent signal translation complex that initiates nodulation and

mycorrhization in legumes, they are very similar and may even overlap. Genes that are shared at the level of the colonization process are referred to as common SYM genes [18], alluding to the process of symbiosis. Some researchers have summarized at least seven SYM genes (SYMRK, CASTOR, POLLUX, SYM3, SYM6, SYM15, and SYM24), including receptor kinases, putative protein channels, and nucleoporins, which are necessary for entry into the plant epidermis in both symbionts [93-95]. NUP85 and CYCLOPS have also been reported [16]. In crops other than legumes, such as rice (Oryza sativa L.), common genes for Rhs and HMA have also been reported, such as CASTOR, POLLUX, DMI3/CCaMK, and CYCLOPS [96] (Figure 2).



Own source

Route shared by Rhs and AMF colonization.

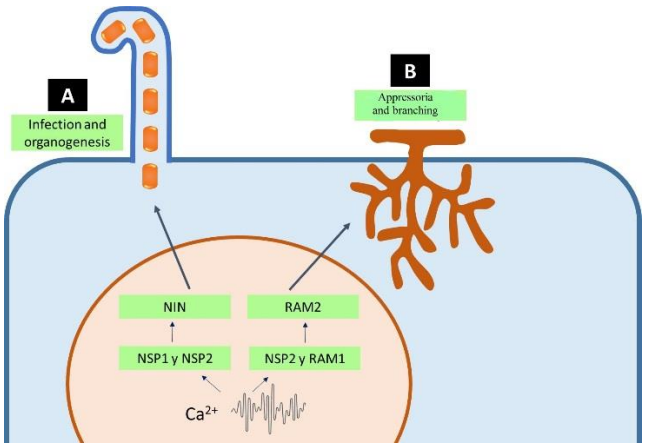
Figure 2. Genes encoding for calcium transporters (CASTOR, POLLUX/DM1, NUP85, NUP133, NENA) within the plant cell nucleus and induction of a cation gradient

Although there are SYM genes shared by Rhs and AMF, it is important to note that both symbionts developed different ways to colonize their host. In the case of Rhs, they form a tube that penetrates the cells and in the case of AMF a pre-penetration apparatus or appressorium. Some researchers suggest that the appressorium is related to transcriptional factors of the ENOD11 and ENOD12 genes that, in turn, are induced by Rhs [97]. Subsequently, other researchers found that ENODs are key for colonization and organogenesis in Rhs and are regulated by ERN1 (ethylene response factor required for nodulation 1) [55].

On the other hand, both symbionts, as mentioned above, require communication with host roots in order to develop the symbiotic process. In this communication, it was mentioned that the Nod (in Rhs) and Myc (in AMF) genes are activated, which produce LCOs and LCOs+COS, respectively. Some mycorrhiza-forming fungi such as *Rhizophagus irregularis* produce sulfated LCOs very similar to the LCOs emitted by the Nod factors of the bacterium *Sinorhizobium* meliloti in the plant *M. truncatula*. This causes, in parallel, the plant to "believe" that it will be colonized by Rhs, when in fact it will be by an AMF, thus causing curvature of root hairs and proliferation of lateral roots [36]. At the molecular level, this event triggers the legume to activate the NSP1 (Nodulation Signaling Pathway) transcription factor required for nodulation and the RAM1 (Required for Arbuscular Mycorrhization) gene required for AMF colonization [98,99] (Figure 3).

Although there are SYM genes shared by Rhs and AMF, it is important to note that both symbionts developed different ways to colonize their host. In the case of Rhs, they form a tube that penetrates the cells and in the case of AMF a pre-penetration apparatus or appressorium. Some researchers suggest that the appressorium is related to transcriptional factors of the ENOD11 and ENOD12 genes which, in turn, are induced by Rhs [97]. Subsequently, other researchers found that ENODs are key for colonization and organogenesis in Rhs and are regulated by ERN1 (ethylene response factor required for nodulation 1) [55].

On the other hand, both symbionts, as mentioned above, require communication with host roots in order to develop the symbiotic process. In this communication, it was mentioned that the Nod (in Rhs) and Myc (in AMF) genes are activated, which produce LCOs and LCOs+COS, respectively. Some mycorrhiza-forming fungi such as *Rhizophagus irregularis* produce sulfated LCOs very similar to the LCOs emitted by the Nod factors of the bacterium *Sinorhizobium meliloti* in the plant *M. truncatula*. This causes, in parallel, the plant to "believe" that it will be colonized by Rhs, when in fact it will be by an AMF, thus causing curvature of root hairs and proliferation of lateral roots [36]. At the molecular level, this event triggers the legume to activate the NSP1 (Nodulation Signaling Pathway) transcription factor required for nodulation and the RAM1 (Required for Arbuscular Mycorrhization) gene required for AMF colonization [98,99] (Figure 3).



Author's own source

A: in the first case, NIN activation is arranged to continue colonization and organogenesis of the nodules

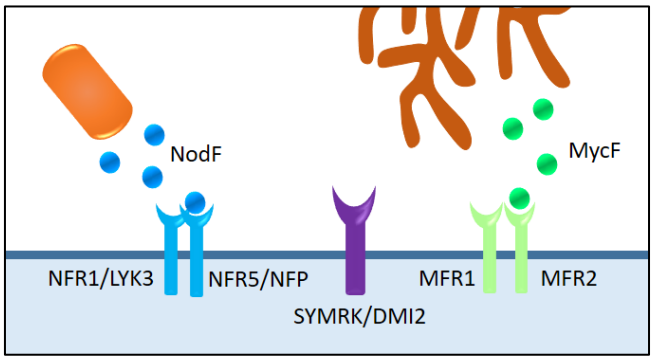
B: in the second case, hyphal branching is promoted

Figure 3. Induction of plant host acceptance mechanism upon exposure to Rhs and AMF

Another mechanism of perception of the external signal and that, in addition, interacts with the Nod/Myc factors is carried out by hormones, such is the case of brassinosteroids, with the BRI1 gene and strigolactones with the CCD7 gene, which are implicated in the signaling of the symbiosis between Rhs/AMF [69,70]. DELLA proteins, related to gibberellins, are also involved in the regulation of negative gene expression induced by Nod factors for Rhs/AM and this is manifested by their close relationship with TFs IPD3 and NSP2 [100].

At the host receptor level, the plant membrane receptors LjNFR1/MtLYK3 and LjNFR5/MtNFP sense the stimulus of Nod factors. However, there is a third gene coding for a similar type of receptor called LjSYMRK

and its ortholog MtDMI2. The latter fulfill the same function, since they are of the SYM type, which means that they are able to perceive the stimulus of both AMFs and Rhs (Figure 4) [101,102]. The LjSYMRK/MtDMI2 gene was the first gene found to be common for AMF and Rhs symbiosis. Also the MFR1 and MFR2 receptors are specific to AMF.

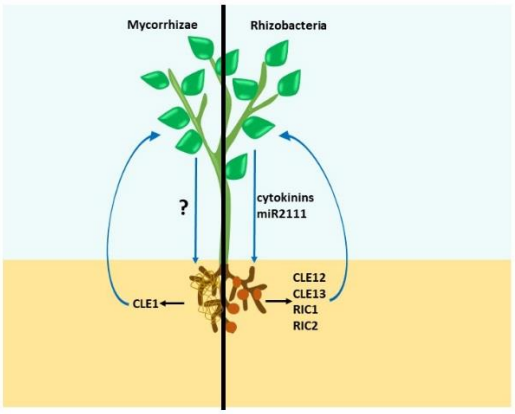


Author's own source

Figure 4. Genes coding for plant receptors involved in the recognition of nodulation and colonization factors in Rhs and AMF

Once the stimulus is perceived and inside the host cell nucleus, a depolarization of the cell membrane and a change in the flow of ions, especially calcium in the nuclear membrane, is induced. This process was described previously, as mentioned above, the similarity between Rhs and AMF was demonstrated, since the DMI1, and NENA genes, which are shared between Rhs and AMF, modulate calcium. DMI1 is related to a Ca^{2+} transporter channel and NENA to a nucleoporin transporter protein, both of which affect symbiont infection to the same extent [74]. Subsequent to Ca^{2+} depolarization in Rhs, complex formation occurs in parallel between GRAS proteins as transcription factors (NSP1, NSP2), together with RAM1 and RAM2, which induce the biosynthesis of cutin monomers and are related to the formation of appressoria in AMF (Figure 3).

Another relevant fact that is shared by Rhs and AMF is the presence of specific hormones such as CLE proteins, which mediate cell-cell communication in plants and are clearly identified in *M. truncatula*, as well as their counterparts in other legume species. Specifically, they are molecules that communicate to shoots to signal roots to stop colonization by bacteria [80,81] (Figure 5). However, these peptides are also incorporated into the roots, through AMF, to modulate root architecture, favoring lateral growth and inhibiting apical growth [103]. This, in a way, suggests that there may be a direct or indirect involvement in AMF-mediated regulation of Rhs infection, since under conditions of dual colonization, CLE peptides will activate the regulatory mechanism in plants to not allow further colonization.



Author's own creation

CLE1 is also incorporated by AMF, resulting in branching of secondary roots

Figure 5. CLE peptides delivered from nodules to shoots, which in turn activate the AON mechanism to regulate the amount of CLE peptides

Some investigators report a decrease in the presence of Rhs by AMF inoculation [104]. In other studies, on the contrary, the dual infection by inoculation with Rhs/AMF versus individual inoculation has been performed and quantified and it has been found that, although AMF can activate the AON mechanism by means of CLE peptides. They are not enough to reduce the infection by Rhs; in fact, they have even been reduced for hyphae in the tissue of their host and are more efficient when inoculated together with Rhs [23,105-107]. The above described shows how there is not only a regulation of the symbionts by the plant, but also at the level of the microorganisms themselves; however, it is not of sufficient magnitude and they rather coexist with their host.

On the other hand, Rhs have a characteristic and abundant compound: leghemoglobins, whose function is to protect bacteria from oxygen ingress so that nitrogenase can perform biological fixation of atmospheric nitrogen [21]. The VfLb29 gene is responsible for producing the proteins necessary for leghemoglobin. This gene is expressed in the same way when there is AMF infection, due to the fact that the expression of the promoter of a gene that codes for a phosphorus transporter (StPt3), activates the expression of VfLb29 [18,108].

With the advent of new technologies, such as gene expression studies through the transcriptome, it has been possible to elucidate the genes that are activated or silenced when a plant is colonized by AMF, Rhs, or both. Early studies in this area found 75 genes up-regulated (toward the 5' end) during the dual colonization event [90].

Subsequently, other investigators compared differential gene expression profiles (DEGs), and found "upstream/downstream" gene expression (toward the 5'/toward the 3' end), with 288/233 genes being common for AMF and Rhs [27]. In this study, quantitative gene expression profiles were found at three stages: biological processes (PB), molecular function (MF) and cellular components (CC). Accordingly, in PB, a high frequency of genes related to metabolic processes, energy pathways, signal translation, transport and stress response was obtained; for the FM stage, genes related to enzymes with catalytic activity such as hydrolase, oxidoreductase,

protein kinases and transferase activity were found; and finally, for the CC stage, genes for the cell wall and plasma membrane were found. In summary, the genes were grouped into three expression clusters according to the processes involved and it was determined that both AMF and Rhs share genes involved in defense processes, cell wall structure and N and P metabolism [27].

Finally, the studies presented here do not necessarily describe the totality of gene expression diversity, as they have been performed on model species and each plant host interacts uniquely with its symbiont(s). In some cases, several plant species have been inoculated with the same AMF and similar metabolic responses have been obtained; this mechanism is known as species-independent. For example, when inoculating several plant species with the fungus *Rhizophagus irregularis*, a change in the metabolome of between 18-45 % was observed in all inoculated species [109,110]. A species-dependent association has also been recorded, i.e., inoculating several plant species with a fungus resulted in a change in the metabolome of only one species [111].

Genetic engineering at the forefront of the biotechnological development of AMF and Rhs

With the advent of new technologies and tools in the field of genetic engineering, there is a tendency to develop projects in this line, due to the fact that each time a study is published, more questions and possibilities for modifications in numerous fields are opened. With regard to Rhs, it is important to highlight the potential that exists in terms of modifying the acceptance of a bacterium by the plant host.

As mentioned and as described in this paper, genes of the NBS-LRR group, such as Rj2, Rfg1 and Rj4, have been registered and are associated with host restriction to a range of bacteria, because they encode for family 5 proteins associated with pathogenesis [77,78] and this is a reason to pay attention to a possible modification of these genes. A group of researchers made the first approaches, using CRISPR/Cas9 to increase colonization of strains incompatible with soybean (*Glycine max*) [78].

Another research [112], in which modification of NCR (cysteine-rich nodule) peptides was performed, was reported. These molecules play an important role in Rhs restriction and this allowed *M. truncatula* to be colonized by a hitherto poorly infective Rhs strain [103]. Finally, there is also the possibility of modifying the LjNFR1/MtLYK3 and LjNFR5/MtNFP receptors, as they alter the degree and specificity to which an Rhs species succeeds in colonizing the plant host [113].

The above criteria indicate that there is a possibility that different plant species can be colonized by more than one strain of Rhs, although there may be some specificity between the plant and bacterial genomes, helping to optimize the expression of each gene. Despite the above, a cautious position must also be taken so that the balance towards the plant or ecosystem is always positive, however, it is an interesting line of research.

Another aspect to consider is the use of genetic transformation for transferring genes from one species of Rhs to another, in the search to improve the efficiency of host colonization. The transfer of Nod genes from some bacteria to others has been recorded; for example, from *Rhizobium leguminosarum* to *Rhizobium phaseoli* so that the latter colonize peas (*Pisum sativum*), as well as beans (*Phaseolus vulgaris*) [114]. This technology also opens the possibility of transferring Nif (nitrogen fixation) genes.

Genetic transformation can be carried out by direct or indirect methods. In this case, the use of indirect methods by means of Agrobacterium would be indicated, due to the fact that it shares many similarities with the Rhs of legumes.

In addition to genetic transformation at the bacterial level, transformation at the host level could be considered. Candidate genes for this purpose are: Rj2, Rfg1 and Rj4 from the NBS-LRR group and those related to the NFR1/LYK3 and NFR5/NFP receptors (they alter the degree and specificity in which an Rh species succeeds in colonizing the plant host).

Following the Rhs line, there is the possibility of modifying the nitrogenase enzyme (NifH, NifD, NifK, NifE and NifN), which consists of a long subunit composed of Molybdenum-Iron and a small one composed of a ferric protein or dinitrogenase reductase. The latter is coupled to an Mg-ATP complex that delivers energy and donates electrons for nitrogen reduction [114]. This enzyme has been identified in most Rhs-infected legume species, which is why it may be of special attention for genetic modification. The main objective would be to make it more efficient by improving coupling with Mg-ATP and increasing nitrogen reduction. This would result in increased ammonium delivery (mainly) to the plant.

On the other hand, molybdenum, as well as iron and sulfur are important for nitrogenase. One group of researchers identified that the MOT1.2/1.3 genes are related to transporters found in the plasma membrane of endodermal cells, which enclose the vascular branches of the nodule, and these modulate the entry as well as the distribution of Mo in the cells [115]. How Mo is transported to the symbiosome remains to be determined [38], but these genes are candidates for study to decipher the possibility of making the Mo-Fe subunit of nitrogenase more efficient.

Calcium transporters (NENA, CASTOR, POLLUX, NUP85, NUP133, DMI3 and DMI1) are also susceptible to modification, so that the calcium gradient produced in the nucleus is the reason for an increased transcription of the colonization response. It is important to emphasize that, regardless of the transporter that is chosen to be modified, what should be sought is the optimization of the process; that is, not to exceed the limits that the plant can tolerate so as not to cause an imbalance in terms of energy balance.

Regarding AMF, the most common use is the extraction of their propagules from the soil to inoculate *ex-situ* and thus increase the amount of this type of fungus in agricultural soils, this is carried out because AMF are not very specific and can colonize several hosts, thus registering many studies in this regard [116]. The identification of new species through metagenomics can support the typical *ex-situ* inoculation; recently, new species were found in environments as inhospitable as deserts and could be used in diverse crops to increase their yields [117]. In addition, sequencing of AMF could provide new discoveries in genomics and transcriptomics concerning the infection mechanism they share with Rhs.

The use of genetic engineering has great potential for agriculture, but sequencing of species is needed to identify and modify the expression of genes of interest. However, this aspect has been little investigated and only *R. irregularis* has been sequenced in one study, where a compilation of genes of interest that can be modified in this species is made [109]. On the other hand, some researchers suggest using, initially, the

information obtained in other fungi such as *Aspergillus niger* and *Penicillium chrysogenum* in terms of promoters and transcription factors [118].

Because most AMF do not reproduce sexually, it makes these organisms highly promising when using genetic engineering and synthetic biology, as genes introduced are unlikely to be cross-incorporated into other species [119]. In addition, this type of fungi is of great importance in the phytoremediation of soils contaminated with heavy metals, since there are scientific records of the possibility of improving the expression of genes that encode for chelating proteins such as phytochelatins and metallothioneins, as well as metabolites such as oxalate, which deactivate heavy metal toxicity [109,120].

Despite the benefits provided by AMF mentioned in this review, not enough research has been done in molecular genetics to increase the beneficial effects of these fungi. First, future research should focus on basic issues such as identification and expression of genes that may affect plant growth and metabolism. On the other hand, with metagenomic studies, to characterize the diversity that exists in the edaphic environment, in order to have a wide variety of options for future applications that could be beneficial to humans and different ecosystems.

CONCLUSIONS

- The microsymbionts (Rhs and AMF) are oriented to their host by the signal emitted by the host roots. Upon sensing the signal, both microsymbionts begin to activate genes that secrete compounds that bind with the membrane of their host to, in turn, activate the mechanism of acceptance and attachment by the plant.
- The plant configures its anatomy, expressing genes that allow microsymbionts to colonize and exchange chemical compounds in a continuous communication process, through the perisymbiotic and perihaustorial membrane for Rhs and AMF, respectively.
- Common genes have great potential to be candidates for genetic modification and thus make colonization more efficient, either by one or several microsymbiont species. It is important to assess when maximum efficiency is achieved individually or with a set of species/strains.
- Although most AMF are complex to sequence because they are difficult to culture at the laboratory level, it is necessary to conduct such studies, as well as to take advantage of current technologies, such as metagenomics, to identify species and genes related to colonization, regulation and expression pathways. Once these have been elucidated, new genes common to both symbionts can be identified.

BIBLIOGRAPHY

1. Gruber N, Galloway JN. An Earth-system perspective of the global nitrogen cycle. Nature [Internet]. 2008;451(7176):293–6. Available from: https://www.nature.com/articles/nature06592

2. Sakamoto K, Ogiwara N, Kaji T. Involvement of autoregulation in the interaction between rhizobial nodulation and AM fungal colonization in soybean roots. Biology and Fertility of soils [Internet]. 2013;49(8):1141–52. Available from: https://link.springer.com/article/10.1007/s00374-013-0804-8

3. Foyer CH, Lam H-M, Nguyen HT, Siddique KH, Varshney RK, Colmer TD, et al. Neglecting legumes has compromised human health and sustainable food production. Nature plants [Internet]. 2016;2(8):1–10. Available from: https://www.nature.com/articles/nplants2016112

4. Wade MJ. The co-evolutionary genetics of ecological communities. Nature Reviews Genetics [Internet]. 2007;8(3):185–95. Available from: https://www.nature.com/articles/nrg2031

5. Gilbert SF, Bosch TC, Ledón-Rettig C. Eco-Evo-Devo: developmental symbiosis and developmental plasticity as evolutionary agents. Nature Reviews Genetics [Internet]. 2015;16(10):611–22. Available from: https://www.nature.com/articles/nrg3982

6. Kiers ET, West SA. Evolving new organisms via symbiosis. Science [Internet]. 2015;348(6233):392–4. Available from: https://www.science.org/doi/abs/10.1126/science.aaa9605

7. McFall-Ngai M, Hadfield MG, Bosch TC, Carey HV, Domazet-Lošo T, Douglas AE, et al. Animals in a bacterial world, a new imperative for the life sciences. Proceedings of the National Academy of Sciences [Internet]. 2013;110(9):3229–36. Available from: https://www.pnas.org/content/110/9/3229.short

8. Simon L, Bousquet J, Lévesque RC, Lalonde M. Origin and diversification of endomycorrhizal fungi and coincidence with vascular land plants. Nature [Internet]. 1993;363(6424):67–9. Available from: https://www.nature.com/articles/363067a0

9. Remy W, Taylor TN, Hass H, Kerp H. Four hundred-million-year-old vesicular arbuscular mycorrhizae. Proceedings of the National Academy of Sciences. 1994;91(25):11841–3.

10. Doyle JJ. Phylogenetic perspectives on the origins of nodulation. Molecular Plant-Microbe Interactions [Internet]. 2011;24(11):1289–95. Available from: https://apsjournals.apsnet.org/doi/abs/10.1094/MPMI-05-11-0114

11. Lum MR, Hirsch AM. Roots and their symbiotic microbes: strategies to obtain nitrogen and phosphorus in a nutrient-limiting environment. Journal of Plant Growth Regulation [Internet]. 2002;21(4):368–82. Available from: https://d1wqtxts1xzle7.cloudfront.net/50695661/Roots_and_Their_Symbiotic_Microbes_Strat20161203-18979-zmsc1o.pdf?1480786248=&response-content-disposition=inline%3B+filename%3DRoots_and_their_symbiotic_microbes_strat.pdf&Expires=1637980035&Signature=CLS84WGYgwkPInm3BQHfvQcxuywR35uX-15tbKWTGDWhvTiOOJrRWuR-GP1M6AhDYl2pcH5beB7wk3ZL3h3Bqlq5W5phQyTb5qUqYGs7c8w~a3ZgGzYNS8ZQcFTrFA0MjAZItmdG-AsX6a3dcCrEJXkDIyC6AbkWREc8h7Ekhwb4zJ12R4w2gshuoVmXm7NbeVCkcB-K7juNRbTFff-gApnUfrPvYxydDq9c8rng9DKr8S3tynVW9-d5EW~X1x~RB7hVM83kwAfrc9Tx0zWoJzt8ardCPY6E7YVMMU1QOPvb-e1gCCNwImedD5azzJt0YNMjdNRnsZd7jGcaZ~iBsQ__&Key-Pair-Id=APKAJLOHF5GGSLRBV4ZA

12. Kennedy AC, de Luna LZ. RHIZOSPHERE. In: Hillel D, editor. Encyclopedia of Soils in the Environment [Internet]. Oxford: Elsevier; 2005 [cited 26/11/2021]. p. 399–406. Available from: https://www.sciencedirect.com/science/article/pii/B0123485304001636

13. SCHÜßLER A, Schwarzott D, Walker C. A new fungal phylum, the Glomeromycota: phylogeny and evolution. Mycological research [Internet]. 2001;105(12):1413–21. Available from: https://www.cambridge.org/core/journals/mycological-research/article/abs/new-fungal-phylum-the-glomeromycota-phylogeny-and-evolution/6A4E3EB5D8D502B5571F591F5B705C47

14. Kistner C, Parniske M. Evolution of signal transduction in intracellular symbiosis. Trends in plant science [Internet]. 2002;7(11):511–8. Available from: https://www.sciencedirect.com/science/article/abs/pii/S1360138502023567

15. Vessey JK, Pawlowski K, Bergman B. Root-based N2-fixing symbioses: legumes, actinorhizal plants, *Parasponia* sp. and cycads. Plant and soil [Internet]. 2005;274(1):51–78. Available from: https://link.springer.com/article/10.1007/s11104-005-5881-5

16. Parniske M. Arbuscular mycorrhiza: the mother of plant root endosymbioses. Nature Reviews Microbiology [Internet]. 2008;6(10):763–75. Available from: https://www.nature.com/articles/nrmicro1987

17. Solís-Ramos LY, Coto-López C, Andrade-Torres A. Role of arbuscular mycorrhizal symbiosis in remediation of anthropogenic soil pollution. Symbiosis [Internet]. 2021;1–16. Available from: https://www.researchgate.net/profile/Laura-Solis-Ramos/publication/351356453_Role_of_arbuscular_mycorrhizal_symbiosis_in_remediation_of_anthropogenic_soil_pollution/links/6102c0bd0c2bfa282a0d4330/Role-of-arbuscular-mycorrhizal-symbiosis-in-remediation-of-anthropogenic-soil-pollution.pdf

18. Manchanda G, Garg N. Endomycorrhizal and rhizobial symbiosis: How much do they share? Journal of Plant Interactions [Internet]. 2007;2(2):79–88. Available from: https://www.tandfonline.com/doi/full/10.1080/17429140701558000

19. Matson PA, Parton WJ, Power AG, Swift MJ. Agricultural intensification and ecosystem properties. Science [Internet]. 1997;277(5325):504–9. Available from: https://www.science.org/doi/abs/10.1126/science.277.5325.504

20. Bonfante P, Genre A. Mechanisms underlying beneficial plant–fungus interactions in mycorrhizal symbiosis. Nature communications [Internet]. 2010;1(1):1–11. Available from: https://www.nature.com/articles/ncomms1046?fbclid=IwAR1g7_DQ5BfH6DKqDAuMLO0CIqwsSMMgkEUVRMWED7EVdhpQcloMbKDEMfs

21. Li X, Feng H, Wen J, Dong J, Wang T. MtCAS31 aids symbiotic nitrogen fixation by protecting the leghemoglobin MtLb120-1 under drought stress in *Medicago truncatula*. Frontiers in plant science [Internet]. 2018;9:633. Available from: https://www.frontiersin.org/articles/10.3389/fpls.2018.00633/full

22. Kim G-B, Son S-U, Yu H-J, Mun J-H. MtGA2ox10 encoding C20-GA2-oxidase regulates rhizobial infection and nodule development in *Medicago truncatula*. Scientific reports [Internet]. 2019;9(1):1–13. Available from: https://www.nature.com/articles/s41598-019-42407-3

23. Sakamoto K, Ogiwara N, Kaji T, Sugimoto Y, Ueno M, Sonoda M, et al. Transcriptome analysis of soybean (*Glycine max*) root genes differentially expressed in rhizobial, arbuscular mycorrhizal, and dual symbiosis. Journal of plant research [Internet]. 2019;132(4):541–68. Available from: https://pubmed.ncbi.nlm.nih.gov/31165947/

24. Sakamoto K, Nohara Y. Soybean (*Glycine max* [L.] Merr.) shoots systemically control arbuscule formation in mycorrhizal symbiosis. Soil science and plant nutrition [Internet]. 2009;55(2):252–7. Available from: https://www.tandfonline.com/doi/full/10.1111/j.1747-0765.2009.00358.x

25. Gill AS, Purnell K, Palmer MI, Stein J, McGuire KL. Microbial Composition and Functional Diversity Differ Across Urban Green Infrastructure Types. Frontiers in Microbiology [Internet]. 2020;11:912. Available from: https://www.frontiersin.org/articles/10.3389/fmicb.2020.00912/full

26. Bozsó Z, Maunoury N, Szatmari A, Mergaert P, Ott PG, Zsíros LR, et al. Transcriptome analysis of a bacterially induced basal and hypersensitive response of *Medicago truncatula*. Plant molecular biology [Internet]. 2009;70(6):627–46. Available from: https://d1wqtxts1xzle7.cloudfront.net/46335421/s11103-009-9496-820160608-1847-i5rbjk.pdf?1465381657=&response-content-disposition=inline%3B+filename%3DTranscriptome_analysis_of_a_bacterially.pdf&Expires=1637981832&Signature=aeoaEBCzv8zj58xJM0o4sYtIYNXTA-ToUh7sY6BR1UBVterlBMLVypqejVHJyV-F-dZ4SpcQHOOgo4bchiNK4k5ZkJiwbrLMH-cTqIRYjngcIvYPydWGNnWpG8Fq2J-SgrU6laJ5ySma0kmp4SUYquIxqDR-yDgfYuqBW~wLGBhCZcr55SqmAXRsBgbqCohU1Ub1~8f4QeSW6V1IKHLF5-8qsAAV-RQp2Zr4io7yZcCYVH2ooRiPGu5v89pn-o0tmZ4VlPnW12JNtONCpn1-nD1qYs9yiUDn~~6-1sk12X7xGkjmFeoQlDGfO4yqOuRsREByVtPtQvoxw4foIkubuA__&Key-Pair-Id=APKAJLOHF5GGSLRBV4ZA

27. Nanjareddy K, Arthikala M-K, Gómez B-M, Blanco L, Lara M. Differentially expressed genes in mycorrhized and nodulated roots of common bean are associated with defense, cell wall architecture, N metabolism, and P metabolism. PloS one [Internet]. 2017;12(8):e0182328. Available from: https://journals.plos.org/plosone/article?id=10.1371/journal.pone.0182328

28. Mohammadi-Dehcheshmeh M, Niazi A, Ebrahimi M, Tahsili M, Nurollah Z, Ebrahimi Khaksefid R, et al. Unified transcriptomic signature of arbuscular mycorrhiza colonization in roots of *Medicago truncatula* by integration of machine learning, promoter analysis, and direct merging meta-analysis. Frontiers in plant science [Internet]. 2018;9:1550. Available from: https://www.frontiersin.org/articles/10.3389/fpls.2018.01550/full

29. Tromas A, Parizot B, Diagne N, Champion A, Hocher V, Cissoko M, et al. Heart of endosymbioses: transcriptomics reveals a conserved genetic program among arbuscular mycorrhizal, actinorhizal and legume-rhizobial symbioses. 2012; Available from: https://journals.plos.org/plosone/article?id=10.1371/journal.pone.0044742

30. Barea J-M. Interaction between mycorrhizal fungi and rhizosphere micro-organisms with in the context of sustainable soil-plant systems. Multitrophic interactions in terrestrial systems [Internet]. 1997;65–7. Available from: https://ci.nii.ac.jp/naid/10029653607/

31. Sprent JI, James EK. Legume evolution: where do nodules and mycorrhizas fit in? Plant physiology [Internet]. 2007;144(2):575–81. Available from: https://academic.oup.com/plphys/article/144/2/575/6106716?login=true

32. Zgadzaj R, Garrido-Oter R, Jensen DB, Koprivova A, Schulze-Lefert P, Radutoiu S. Root nodule symbiosis in Lotus japonicus drives the establishment of distinctive rhizosphere, root, and nodule bacterial communities. Proceedings of the National Academy of Sciences [Internet]. 2016;113(49):E7996–8005. Available from: https://www.pnas.org/content/113/49/E7996.short

33. Ferguson BJ, Indrasumunar A, Hayashi S, Lin M-H, Lin Y-H, Reid DE, et al. Molecular analysis of legume nodule development and autoregulation. Journal of integrative plant biology [Internet]. 2010;52(1):61–76. Available from: https://onlinelibrary.wiley.com/doi/full/10.1111/j.1744-7909.2010.00899.x

34. Sprent JI, Ardley J, James EK. Biogeography of nodulated legumes and their nitrogen-fixing symbionts. New Phytologist [Internet]. 2017;215(1):40–56. Available from: https://nph.onlinelibrary.wiley.com/doi/full/10.1111/nph.14474

35. Dénarié J, Debellé F, Promé J-C. Rhizobium lipo-chitooligosaccharide nodulation factors: signaling molecules mediating recognition and morphogenesis. Annual review of biochemistry [Internet]. 1996;65(1):503–35. Available from: https://www.annualreviews.org/doi/abs/10.1146/annurev.bi.65.070196.002443

36. Maillet F, Poinsot V, André O, Puech-Pagès V, Haouy A, Gueunier M, et al. Fungal lipochitooligosaccharide symbiotic signals in arbuscular mycorrhiza. Nature [Internet]. 2011;469(7328):58–63. Available from: https://www.nature.com/articles/nature09622

37. Persson T, Battenberg K, Demina IV, Vigil-Stenman T, Vanden Heuvel B, Pujic P, et al. Candidatus Frankia datiscae Dg1, the actinobacterial microsymbiont of *Datisca glomerata*, expresses the canonical nod genes nodABC in symbiosis with its host plant. PloS one [Internet]. 2015;10(5):e0127630. Available from: https://journals.plos.org/plosone/article?id=10.1371/journal.pone.0127630

38. Roy S, Liu W, Nandety RS, Crook A, Mysore KS, Pislariu CI, et al. Celebrating 20 years of genetic discoveries in legume nodulation and symbiotic nitrogen fixation. The Plant Cell [Internet]. 2020;32(1):15–41. Available from: https://academic.oup.com/plcell/article/32/1/15/6099066?login=true

39. Solís-Ramos LY, Andrade-Torres A. Arbuscular Mycorrhizal Fungi in tropical ecosystems towards its management? Agricultural Research & Technology: Open Access Journal [Internet]. 2020;24(4):152–5. Available from: https://www.researchgate.net/profile/Laura-Solis-Ramos/publication/343322429_Arbuscular_Mycorrhizal_Fungi_in_Tropical_Ecosystems_Towards_its_Management_Mini_Review/links/5f22ff91a6fdcccc4399dfc9/Arbuscular-Mycorrhizal-Fungi-in-Tropical-Ecosystems-Towards-its-Management-Mini-Review.pdf

40. Croll D, Giovannetti M, Koch AM, Sbrana C, Ehinger M, Lammers PJ, et al. Nonself vegetative fusion and genetic exchange in the arbuscular mycorrhizal fungus *Glomus intraradices*. New Phytologist [Internet]. 2009;181(4):924–37. Available from: https://nph.onlinelibrary.wiley.com/doi/full/10.1111/j.1469-8137.2008.02726.x

41. den Bakker HC, VanKuren NW, Morton JB, Pawlowska TE. Clonality and recombination in the life history of an asexual arbuscular mycorrhizal fungus. Molecular Biology and Evolution [Internet]. 2010;27(11):2474–86. Available from: https://academic.oup.com/mbe/article/27/11/2474/1127206?login=true

42. Paszkowski U. A journey through signaling in arbuscular mycorrhizal symbioses 2006. New Phytologist [Internet]. 2006;172(1):35–46. Available from: https://nph.onlinelibrary.wiley.com/doi/full/10.1111/j.1469-8137.2006.01840.x

43. Harrison MJ. Molecular and cellular aspects of the arbuscular mycorrhizal symbiosis. Annual review of plant biology [Internet]. 1999;50(1):361–89. Available from: https://www.annualreviews.org/doi/abs/10.1146/annurev.arplant.50.1.361

44. Johnson NC, Rowland DL, Corkidi L, Egerton-Warburton LM, Allen EB. Nitrogen enrichment alters mycorrhizal allocation at five mesic to semiarid grasslands. Ecology [Internet]. 2003;84(7):1895–908. Available from: https://esajournals.onlinelibrary.wiley.com/doi/abs/10.1890/0012-9658(2003)084[1895:NEAMAA]2.0.CO;2

45. Johnson NC. Resource stoichiometry elucidates the structure and function of arbuscular mycorrhizas across scales. New Phytologist [Internet]. 2010;185(3):631–47. Available from: https://nph.onlinelibrary.wiley.com/doi/full/10.1111/j.1469-8137.2009.03110.x

46. Maherali H, Klironomos JN. Influence of phylogeny on fungal community assembly and ecosystem functioning. science [Internet]. 2007;316(5832):1746–8. Available from: https://www.science.org/doi/abs/10.1126/science.1143082

47. Denison RF, Kiers ET. Life histories of symbiotic rhizobia and mycorrhizal fungi. Current Biology [Internet]. 2011;21(18):R775–85. Available from: https://www.sciencedirect.com/science/article/pii/S0960982211006634

48. Garcia-Garrido JM, Ocampo JA. Effect of VA mycorrhizal infection of tomato on damage caused by *Pseudomonas syringae*. Soil Biology and Biochemistry [Internet]. 1989;21(1):165–7. Available from: https://www.sciencedirect.com/science/article/abs/pii/0038071789900278

49. De La Peña E, Echeverría SR, Van Der Putten WH, Freitas H, Moens M. Mechanism of control of root-feeding nematodes by mycorrhizal fungi in the dune grass *Ammophila arenaria*. New Phytologist [Internet]. 2006;169(4):829–40. Available from: https://nph.onlinelibrary.wiley.com/doi/full/10.1111/j.1469-8137.2005.01602.x

50. Fritz M, Jakobsen I, Lyngkjær MF, Thordal-Christensen H, Pons-Kühnemann J. Arbuscular mycorrhiza reduces susceptibility of tomato to *Alternaria solani*. Mycorrhiza [Internet]. 2006;16(6):413–9. Available from: https://pubmed.ncbi.nlm.nih.gov/16614816/

51. Pozo MJ, Azcón-Aguilar C. Unraveling mycorrhiza-induced resistance. Current opinion in plant biology [Internet]. 2007;10(4):393–8. Available from: https://www.sciencedirect.com/science/article/abs/pii/S1369526607000702

52. Jung SC, Martinez-Medina A, Lopez-Raez JA, Pozo MJ. Mycorrhiza-induced resistance and priming of plant defenses. Journal of chemical ecology [Internet]. 2012;38(6):651–64. Available from: https://pubmed.ncbi.nlm.nih.gov/22623151/

53. Akiyama K, Matsuzaki K, Hayashi H. Plant sesquiterpenes induce hyphal branching in arbuscular mycorrhizal fungi. Nature [Internet]. 2005;435(7043):824–7. Available from: https://www.nature.com/articles/nature03608

54. Genre A, Chabaud M, Balzergue C, Puech-Pagès V, Novero M, Rey T, et al. Short-chain chitin oligomers from arbuscular mycorrhizal fungi trigger nuclear Ca^{2+} spiking in *Medicago truncatula* roots and their production is enhanced by strigolactone. New Phytologist [Internet]. 2013;198(1):190–202. Available from: https://nph.onlinelibrary.wiley.com/doi/full/10.1111/nph.12146

55. Andriankaja A, Boisson-Dernier A, Frances L, Sauviac L, Jauneau A, Barker DG, et al. AP2-ERF transcription factors mediate Nod factor–dependent Mt ENOD11 activation in root hairs via a novel cis-regulatory motif. The Plant Cell [Internet]. 2007;19(9):2866–85. Available from: https://academic.oup.com/plcell/article/19/9/2866/6092340?login=true

56. Heupel S, Roser B, Kuhn H, Lebrun M-H, Villalba F, Requena N. Erl1, a novel era-like GTPase from *Magnaporthe oryzae*, is required for full root virulence and is conserved in the mutualistic symbiont *Glomus intraradices*. Molecular plant-microbe interactions [Internet]. 2010;23(1):67–81. Available from: https://apsjournals.apsnet.org/doi/abs/10.1094/MPMI-23-1-0067

57. Lanfranco L, Novero M, Bonfante P. The mycorrhizal fungus *Gigaspora margarita* possesses a CuZn superoxide dismutase that is up-regulated during symbiosis with legume hosts. Plant Physiology [Internet]. 2005;137(4):1319–30. Available from: https://academic.oup.com/plphys/article/137/4/1319/6112689?login=true

58. Ferguson BJ, Mens C, Hastwell AH, Zhang M, Su H, Jones CH, et al. Legume nodulation: The host controls the party. Plant, cell & environment [Internet]. 2019;42(1):41–51. Available from: https://onlinelibrary.wiley.com/doi/abs/10.1111/pce.13348

59. Wasson AP, Pellerone FI, Mathesius U. Silencing the flavonoid pathway in *Medicago truncatula* inhibits root nodule formation and prevents auxin transport regulation by rhizobia. The Plant Cell [Internet]. 2006;18(7):1617–29. Available from: https://academic.oup.com/plcell/article/18/7/1617/6115313?login=true

60. Zhang J, Subramanian S, Stacey G, Yu O. Flavones and flavonols play distinct critical roles during nodulation of *Medicago truncatula* by *Sinorhizobium meliloti*. The Plant Journal [Internet]. 2009;57(1):171–83. Available from: https://onlinelibrary.wiley.com/doi/full/10.1111/j.1365-313X.2008.03676.x

61. Subramanian S, Stacey G, Yu O. Endogenous isoflavones are essential for the establishment of symbiosis between soybean and *Bradyrhizobium japonicum*. The Plant Journal [Internet]. 2006;48(2):261–73. Available from: https://onlinelibrary.wiley.com/doi/full/10.1111/j.1365-313X.2006.02874.x

62. Bolaños-Vásquez MC, Werner D. Effects of *Rhizobium tropici*, *R. etli* and *R. leguminosarum* bv. phaseoli on nod gene-inducing flavonoids in root exudates of *Phaseolus vulgaris*. Molecular plant-microbe interactions [Internet]. 1997;10(3):339–46. Available from: https://apsjournals.apsnet.org/doi/abs/10.1094/MPMI.1997.10.3.339

63. Xie F, Murray JD, Kim J, Heckmann AB, Edwards A, Oldroyd GE, et al. Legume pectate lyase required for root infection by rhizobia. Proceedings of the National Academy of Sciences [Internet]. 2012;109(2):633–8. Available from: https://www.pnas.org/content/109/2/633.short

64. Haney CH, Long SR. Plant flotillins are required for infection by nitrogen-fixing bacteria. Proceedings of the National Academy of Sciences [Internet]. 2010;107(1):478–83. Available from: https://www.pnas.org/content/107/1/478.short

65. Lefebvre B, Timmers T, Mbengue M, Moreau S, Hervé C, Tóth K, et al. A remorin protein interacts with symbiotic receptors and regulates bacterial infection. Proceedings of the National Academy of Sciences [Internet]. 2010;107(5):2343–8. Available from: https://www.pnas.org/content/107/5/2343.short

66. Haney CH, Riely BK, Tricoli DM, Cook DR, Ehrhardt DW, Long SR. Symbiotic rhizobia bacteria trigger a change in localization and dynamics of the *Medicago truncatula* receptor kinase LYK3. The Plant Cell [Internet]. 2011;23(7):2774–87. Available from: https://academic.oup.com/plcell/article/23/7/2774/6097218?login=true

67. Broghammer A, Krusell L, Blaise M, Sauer J, Sullivan JT, Maolanon N, et al. Legume receptors perceive the rhizobial lipochitin oligosaccharide signal molecules by direct binding. Proceedings of the National Academy of Sciences [Internet]. 2012;109(34):13859–64. Available from: https://www.pnas.org/content/109/34/13859.short

68. Moling S, Pietraszewska-Bogiel A, Postma M, Fedorova E, Hink MA, Limpens E, et al. Nod factor receptors form heteromeric complexes and are essential for intracellular infection in *Medicago nodules*. The Plant Cell [Internet]. 2014;26(10):4188–99. Available from: https://academic.oup.com/plcell/article/26/10/4188/6101576?login=true

69. Liu H, Zhang C, Yang J, Yu N, Wang E. Hormone modulation of legume-rhizobial symbiosis. Journal of Integrative Plant Biology [Internet]. 2018;60(8):632–48. Available from: https://onlinelibrary.wiley.com/doi/abs/10.1111/jipb.12653

70. Cheng X, Gou X, Yin H, Mysore KS, Li J, Wen J. Functional characterisation of brassinosteroid receptor MtBRI1 in *Medicago truncatula*. Scientific reports [Internet]. 2017;7(1):1–12. Available from: https://www.nature.com/articles/s41598-017-09297-9

71. Grunewald W, Van Noorden G, Van Isterdael G, Beeckman T, Gheysen G, Mathesius U. Manipulation of auxin transport in plant roots during Rhizobium symbiosis and nematode parasitism. The Plant Cell [Internet]. 2009;21(9):2553–62. Available from: https://academic.oup.com/plcell/article/21/9/2553/6096165?login=true

72. Ariel F, Brault-Hernandez M, Laffont C, Huault E, Brault M, Plet J, et al. Two direct targets of cytokinin signaling regulate symbiotic nodulation in *Medicago truncatula*. The Plant Cell [Internet]. 2012;24(9):3838–52. Available from: https://academic.oup.com/plcell/article/24/9/3838/6100604?login=true

73. Charpentier M, Bredemeier R, Wanner G, Takeda N, Schleiff E, Parniske M. Lotus japonicus CASTOR and POLLUX are ion channels essential for perinuclear calcium spiking in legume root endosymbiosis. The Plant Cell [Internet]. 2008;20(12):3467–79. Available from: https://academic.oup.com/plcell/article/20/12/3467/6092747?login=true

74. Groth M, Takeda N, Perry J, Uchida H, Dräxl S, Brachmann A, et al. NENA, a Lotus japonicus homolog of Sec13, is required for rhizodermal infection by arbuscular mycorrhiza fungi and rhizobia but dispensable for cortical endosymbiotic development. The Plant Cell [Internet]. 2010;22(7):2509–26. Available from: https://academic.oup.com/plcell/article/22/7/2509/6096023?login=true

75. Bapaume L, Reinhardt D. How membranes shape plant symbioses: signaling and transport in nodulation and arbuscular mycorrhiza. Frontiers in Plant Science [Internet]. 2012;3:223. Available from: https://www.frontiersin.org/articles/10.3389/fpls.2012.00223/full

76. Cao Y, Halane MK, Gassmann W, Stacey G. The role of plant innate immunity in the legume-rhizobium symbiosis. Annual review of plant biology [Internet]. 2017;68:535–61. Available from: https://www.annualreviews.org/doi/abs/10.1146/annurev-arplant-042916-041030

77. Yang S, Tang F, Gao M, Krishnan HB, Zhu H. R gene-controlled host specificity in the legume–rhizobia symbiosis. Proceedings of the National Academy of Sciences [Internet]. 2010;107(43):18735–40. Available from: https://www.pnas.org/content/107/43/18735.short

78. Tang F, Yang S, Liu J, Zhu H. Rj4, a gene controlling nodulation specificity in soybeans, encodes a thaumatin-like protein but not the one previously reported. Plant Physiology [Internet]. 2016;170(1):26–32. Available from: https://academic.oup.com/plphys/article/170/1/26/6114002?login=true

79. Ferguson BJ, Li D, Hastwell AH, Reid DE, Li Y, Jackson SA, et al. The soybean (*Glycine max*) nodulation-suppressive CLE peptide, Gm RIC 1, functions interspecifically in common white bean (*Phaseolus vulgaris*), but not in a supernodulating line mutated in the receptor Pv NARK. Plant Biotechnology Journal [Internet]. 2014;12(8):1085–97. Available from: https://onlinelibrary.wiley.com/doi/full/10.1111/pbi.12216

80. Mortier V, Den Herder G, Whitford R, Van de Velde W, Rombauts S, D'haeseleer K, et al. CLE peptides control *Medicago truncatula* nodulation locally and systemically. Plant Physiology [Internet]. 2010;153(1):222–37. Available from: https://academic.oup.com/plphys/article/153/1/222/6108407?login=true

81. Okamoto S, Ohnishi E, Sato S, Takahashi H, Nakazono M, Tabata S, et al. Nod factor/nitrate-induced CLE genes that drive HAR1-mediated systemic regulation of nodulation. Plant and Cell Physiology [Internet]. 2009;50(1):67–77. Available from: https://academic.oup.com/pcp/article/50/1/67/1851930?login=true

82. Reid DE, Ferguson BJ, Gresshoff PM. Inoculation-and nitrate-induced CLE peptides of soybean control NARK-dependent nodule formation. Molecular Plant-Microbe Interactions [Internet]. 2011;24(5):606–18. Available from: https://apsjournals.apsnet.org/doi/abs/10.1094/MPMI-09-10-0207

83. Harrison MJ. Signaling in the arbuscular mycorrhizal symbiosis. Annu. Rev. Microbiol. [Internet]. 2005;59:19–42. Available from: https://www.annualreviews.org/doi/abs/10.1146/annurev.micro.58.030603.123749

84. Catoira R, Galera C, de Billy F, Penmetsa RV, Journet E-P, Maillet F, et al. Four genes of Medicago truncatula controlling components of a Nod factor transduction pathway. The Plant Cell [Internet]. 2000;12(9):1647–65. Available from: https://academic.oup.com/plcell/article/12/9/1647/6009341?login=true

85. Senoo K, Solaiman MZ, Kawaguchi M, Imaizumi-Anraku H, Akao S, Tanaka A, et al. Isolation of two different phenotypes of mycorrhizal mutants in the model legume plant *Lotus japonicus* after EMS-treatment. Plant and Cell Physiology [Internet]. 2000;41(6):726–32. Available from: https://academic.oup.com/pcp/article/41/6/726/1923261?login=true

86. Provorov NA, Borisov AY, Tikhonovich IA. Developmental genetics and evolution of symbiotic structures in nitrogen-fixing nodules and arbuscular mycorrhiza. Journal of Theoretical Biology [Internet]. 2002;214(2):215–32. Available from: https://www.sciencedirect.com/science/article/abs/pii/S0022519301924531

87. Roth LE, Stacey G. Bacterium release into host cells of nitrogen-fixing soybean nodules: the symbiosome membrane comes from three sources. European journal of cell biology [Internet]. 1989;49(1):13–23. Available from: https://europepmc.org/article/med/2759097

88. Day DA, Kaiser BN, Thomson R, Udvardi MK, Moreau S, Puppo A. Nutrient transport across symbiotic membranes from legume nodules. Functional Plant Biology [Internet]. 2001;28(7):669–76. Available from: https://www.publish.csiro.au/fp/pp01028

89. Parniske M. Intracellular accommodation of microbes by plants: a common developmental program for symbiosis and disease? Current opinion in plant biology [Internet]. 2000;3(4):320–8. Available from: https://www.sciencedirect.com/science/article/abs/pii/S1369526600000881

90. Manthey K, Krajinski F, Hohnjec N, Firnhaber C, Pühler A, Perlick AM, et al. Transcriptome profiling in root nodules and arbuscular mycorrhiza identifies a collection of novel genes induced during *Medicago truncatula* root endosymbioses. Molecular Plant-Microbe Interactions [Internet]. 2004;17(10):1063–77. Available from: https://apsjournals.apsnet.org/doi/abs/10.1094/MPMI.2004.17.10.1063

91. Hohnjec N, Perlick AM, Pühler A, Küster H. The *Medicago truncatula* sucrose synthase gene MtSucS1 is activated both in the infected region of root nodules and in the cortex of roots colonized by

arbuscular mycorrhizal fungi. Molecular Plant-Microbe Interactions [Internet]. 2003;16(10):903–15. Available from: https://apsjournals.apsnet.org/doi/abs/10.1094/MPMI.2003.16.10.903

92. Chen L-Q, Hou B-H, Lalonde S, Takanaga H, Hartung ML, Qu X-Q, et al. Sugar transporters for intercellular exchange and nutrition of pathogens. Nature [Internet]. 2010;468(7323):527–32. Available from: https://www.nature.com/articles/nature09606

93. Imaizumi-Anraku H, Takeda N, Charpentier M, Perry J, Miwa H, Umehara Y, et al. Plastid proteins crucial for symbiotic fungal and bacterial entry into plant roots. Nature [Internet]. 2005;433(7025):527–31. Available from: https://www.nature.com/articles/nature03237

94. Kistner C, Winzer T, Pitzschke A, Mulder L, Sato S, Kaneko T, et al. Seven Lotus japonicus genes required for transcriptional reprogramming of the root during fungal and bacterial symbiosis. The Plant Cell [Internet]. 2005;17(8):2217–29. Available from: https://academic.oup.com/plcell/article/17/8/2217/6114612?login=true

95. Kanamori N, Madsen LH, Radutoiu S, Frantescu M, Quistgaard EM, Miwa H, et al. A nucleoporin is required for induction of Ca^{2+} spiking in legume nodule development and essential for rhizobial and fungal symbiosis. Proceedings of the National Academy of Sciences [Internet]. 2006;103(2):359–64. Available from: https://www.pnas.org/content/103/2/359.short

96. Gutjahr C, Banba M, Croset V, An K, Miyao A, An G, et al. Arbuscular mycorrhiza–specific signaling in rice transcends the common symbiosis signaling pathway. The Plant Cell [Internet]. 2008;20(11):2989–3005. Available from: https://academic.oup.com/plcell/article/20/11/2989/6092513?login=true

97. Genre A, Bonfante P. Building a mycorrhizal cell: how to reach compatibility between plants and arbuscular mycorrhizal fungi. Journal of Plant Interactions [Internet]. 2005;1(1):3–13. Available from: https://www.tandfonline.com/doi/full/10.1080/17429140500318986

98. Gobbato E, Marsh JF, Vernié T, Wang E, Maillet F, Kim J, et al. A GRAS-type transcription factor with a specific function in mycorrhizal signaling. Current Biology [Internet]. 2012;22(23):2236–41. Available from: https://www.sciencedirect.com/science/article/pii/S0960982212011463

99. Charpentier M, Sun J, Martins TV, Radhakrishnan GV, Findlay K, Soumpourou E, et al. Nuclear-localized cyclic nucleotide–gated channels mediate symbiotic calcium oscillations. Science [Internet]. 2016;352(6289):1102–5. Available from: https://www.science.org/doi/abs/10.1126/science.aae0109

100. Jin Y, Liu H, Luo D, Yu N, Dong W, Wang C, et al. DELLA proteins are common components of symbiotic rhizobial and mycorrhizal signalling pathways. Nature Communications [Internet]. 2016;7(1):1–14. Available from: https://www.nature.com/articles/ncomms12433?origin=ppub

101. Endre G, Kereszt A, Kevei Z, Mihacea S, Kaló P, Kiss GB. A receptor kinase gene regulating symbiotic nodule development. Nature [Internet]. 2002;417(6892):962–6. Available from: https://www.nature.com/articles/nature00842

102. Stracke S, Kistner C, Yoshida S, Mulder L, Sato S, Kaneko T, et al. A plant receptor-like kinase required for both bacterial and fungal symbiosis. Nature [Internet]. 2002;417(6892):959–62. Available from: https://www.nature.com/articles/nature00841

103. Wang G, Zhang G, Wu M. CLE peptide signaling and crosstalk with phytohormones and environmental stimuli. Frontiers in plant science [Internet]. 2016;6:1211. Available from: https://www.frontiersin.org/articles/10.3389/fpls.2015.01211/full

104. Ballesteros-Almanza L, Altamirano-Hernandez J, Pena-Cabriales JJ, Santoyo G, Sanchez-Yanez JM, Valencia-Cantero E, et al. Effect of co-inoculation with mycorrhiza and rhizobia on the nodule trehalose content of different bean genotypes. The open microbiology journal [Internet]. 2010;4:83. Available from: https://www.ncbi.nlm.nih.gov/pmc/articles/PMC3023947/

105. González RL, Sosa BN, Díaz RB. Efecto de la aplicación de Rhizobium y Mycorriza en el crecimiento del frijol (*Phaseolus vulgaris* L.) variedad CC-25-9 negro. Centro Agrícola [Internet]. 2012;39(4):17–20. Available from: http://cagricola.uclv.edu.cu/descargas/pdf/V39-Numero_4/cag044121877.pdf

106. Quintana LJO, Peraza RH, Gómez EF, Rodríguez CH. Efecto de inoculaciones conjuntas de Rhizobium-micorrizas arbusculares en *Leucaena leucocephala* cv: Perú. Centro Agrícola [Internet]. 2014;41(3):17–21. Available from: http://cagricola.uclv.edu.cu/descargas/pdf/V41-Numero_3/cag033141982.pdf

107. Tajini F, Trabelsi M, Drevon J-J. Combined inoculation with *Glomus intraradices* and *Rhizobium tropici* CIAT899 increases phosphorus use efficiency for symbiotic nitrogen fixation in common bean (*Phaseolus vulgaris* L.). Saudi Journal of Biological Sciences [Internet]. 2012;19(2):157–63. Available from: https://www.sciencedirect.com/science/article/pii/S1319562X11000726

108. Fehlberg V, Vieweg MF, Dohmann EM, Hohnjec N, Pühler A, Perlick AM, et al. The promoter of the leghaemoglobin gene VfLb29: functional analysis and identification of modules necessary for its activation in the infected cells of root nodules and in the arbuscule-containing cells of mycorrhizal roots. Journal of Experimental Botany [Internet]. 2005;56(413):799–806. Available from: https://academic.oup.com/jxb/article/56/413/799/550015?login=true

109. French KE. Engineering mycorrhizal symbioses to alter plant metabolism and improve crop health. Frontiers in Microbiology [Internet]. 2017;8:1403. Available from: https://internal-journal.frontiersin.org/articles/10.3389/fmicb.2017.01403/full

110. Schweiger R, Baier MC, Persicke M, Müller C. High specificity in plant leaf metabolic responses to arbuscular mycorrhiza. Nature Communications [Internet]. 2014;5(1):1–11. Available from: https://www.nature.com/articles/ncomms4886

111. Rivero J, Gamir J, Aroca R, Pozo MJ, Flors V. Metabolic transition in mycorrhizal tomato roots. Frontiers in Microbiology [Internet]. 2015;6:598. Available from: https://internal-journal.frontiersin.org/articles/10.3389/fmicb.2015.00598/full

112. Wang Q, Yang S, Liu J, Terecskei K, Ábrahám E, Gombár A, et al. Host-secreted antimicrobial peptide enforces symbiotic selectivity in *Medicago truncatula*. Proceedings of the National Academy of Sciences [Internet]. 2017;114(26):6854–9. Available from: https://www.pnas.org/content/114/26/6854.short

113. Radutoiu S, Madsen LH, Madsen EB, Felle HH, Umehara Y, Grønlund M, et al. Plant recognition of symbiotic bacteria requires two LysM receptor-like kinases. Nature [Internet]. 2003;425(6958):585–92. Available from: https://www.nature.com/articles/nature02039

114. Kumaar SA, Babu RP, Vivek P, Saravanan D. Role of Nitrogen Fixers as Biofertilizers in Future Perspective: A Review. Research Journal of Pharmacy and Technology [Internet]. 2020;13(5):2459–67. Available from: https://www.indianjournals.com/ijor.aspx?target=ijor:rjpt&volume=13&issue=5&article=070

115. Gil-Díez P, Tejada-Jiménez M, León-Mediavilla J, Wen J, Mysore KS, Imperial J, et al. MtMOT1. 2 is responsible for molybdate supply to *Medicago truncatula* nodules. Plant, cell & environment [Internet]. 2019;42(1):310–20. Available from: https://onlinelibrary.wiley.com/doi/abs/10.1111/pce.13388

116. Torrez V, Ceulemans T, Mergeay J, De Meester L, Honnay O. Effects of adding an arbuscular mycorrhizal fungi inoculum and of distance to donor sites on plant species recolonization following topsoil removal. Applied Vegetation Science [Internet]. 2016;19(1):7–19. Available from: https://onlinelibrary.wiley.com/doi/abs/10.1111/avsc.12193

117. Symanczik S, Błaszkowski J, Chwat G, Boller T, Wiemken A, Al-Yahya'ei MN. Three new species of arbuscular mycorrhizal fungi discovered at one location in a desert of Oman: *Diversispora omaniana*, *Septoglomus nakheelum* and *Rhizophagus arabicus*. Mycologia [Internet]. 2014;106(2):243–59. Available from: https://www.tandfonline.com/doi/abs/10.3852/106.2.243

118. Polli F, Meijrink B, Bovenberg RA, Driessen AJ. New promoters for strain engineering of *Penicillium chrysogenum*. Fungal Genetics and Biology [Internet]. 2016;89:62–71. Available from: https://www.sciencedirect.com/science/article/abs/pii/S1087184515300517

119. Pawlowska TE. Genetic processes in arbuscular mycorrhizal fungi. FEMS Microbiology Letters [Internet]. 2005;251(2):185–92. Available from: https://academic.oup.com/femsle/article/251/2/185/601349?login=true

120. Sayer JA, Gadd GM. Solubilization and transformation of insoluble inorganic metal compounds to insoluble metal oxalates by *Aspergillus niger*. Mycological Research [Internet]. 1997;101(6):653–61. Available from: https://www.cambridge.org/core/journals/mycological-research/article/abs/solubilization-and-transformation-of-insoluble-inorganic-metal-compounds-to-insoluble-metal-oxalates-by-aspergillus-niger/0A498FDCFD1784B980BAD284216A8EAB