Mycorrhiza Helper Bacteria: Future Prospects

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ABSTRACT

Mycorrhiza helper bacteria (MHB) are a group of organism that selectively promotes the establishment of mycorrhizal symbiosis. The establishment of mycorrhizal symbioses can be positively influenced by certain bacterial isolates, an effect exhibited by mycorrhiza helper bacteria. Nowadays MHB are considered as plant growth promoting rhizobacteria (PGPR). There are potential practical application of mycorrhiza helper bacteria in agriculture and forestry. On the basis of scientific and practical interests, supported by the development of genomics, may represent a unique opportunity to place MHB at the forefront of future mycorrhiza research and to boost the more general field of fungal-bacterial interactions in ecosystems and for the benefit of mankind.

Keywords: Mycorrhiza, symbioses, genomics, ecosystems, fungal.

INTRODUCTION

'Mycorrhiza helper bacteria' (MHB) was first coined to refer only to bacteria that promote the establishment of the root-fungus symbiosis, or bacteria associated with roots and mycorrhizal fungi that selectively promote the establishment of mycorrhizal symbiosis. The establishment of mycorrhizal symbioses can be positively influenced by certain bacterial isolates, an effect exhibited by mycorrhiza helper bacteria. The MHB include a variety of Gram-negative and Gram-positive species usually associated with mycorrhizal symbiosis. Many MHB are considered nowadays as plant growth promoting rhizobacteria (PGPR), such as Pseudomonas sp. Another factor that complicates the distinction of the two terms (PGPR and MHB) is that studies with PGPR generally exclude the evaluation of mycorrhization. However, it is interesting to note that some fungal signaling pathways are mutually regulated by different rhizobacteria, while others are specific to some MHB. According to Garbaye, the MHB are not plant specific, but are clearly selective about the fungal species, and the term fungus-specific can be used. Among ectomycorrhizal fungi, only basidiomycetes have been described to be interacting with MHB.

MHB are the best studied inhabitants of the mycorrhizosphere. Although most studies of MHB have been conducted in ectomycorrhizal systems, MHB have also been shown to occur in arbuscular symbioses. In ectomycorrhizal systems, MHB have been isolated from the mycorrhizospheres of many tree-fungus symbioses in nursery, plantation and semi natural situations. Garbaye (1994) proposed five main mechanisms by which MHB could promote mycorrhiza formation, although to date there has been little robust evidence for the relative importance of any of them. Most evidence for MHB mechanisms has been gathered from in vitro studies of the interactions between mycorrhizal fungi and bacteria or their metabolites in the absence of the host plant, although there is evidence that some MHB require close proximity or contact with the plant to exert MHB effects.

[¹]
Occurrence of mycorrhiza helper bacteria

There are different types of mycorrhizal symbiosis, the arbuscular mycorrhizal one being the most common and the most frequent all over the world. The arbuscular mycorrhizal symbiosis is formed by obligately symbiotic fungi from the Glomeromycota. Colonization of plant roots by arbuscular mycorrhizal fungi is achieved via spores and mycelia originating from infected roots. The hyphae enter the root tissues and develop inter- and intracellularly, forming running hyphae, coils and arbuscules. Ectomycorrhizal symbioses are formed by a large number of fungal species, mainly Basidiomycetes, but also Ascomycetes. The fungus forms a mantle which encloses the root. Some hyphae extend to the surrounding soil; others pass between the epidermal and cortical cells and form the so-called Hartig net, the site for nutritional exchange between the fungal and plant cells. Many bacterial strains have been reported to be able to promote either arbuscular or ectomycorrhizal. [2,3] Many plant models have been used to study the MHB effect (Table 1).

<table>
<thead>
<tr>
<th>Mycorrhizal fungi</th>
<th>Identity of the MHB isolates</th>
<th>Host plant</th>
<th>Ecological origin of MHB isolates</th>
<th>Effect of MHB</th>
<th>References</th>
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<tbody>
<tr>
<td>Amanita muscaria, Suillus bovinus</td>
<td>Streptomyces</td>
<td>Picea abies, Pinus sylvestris</td>
<td>A. muscaria-containing spruce stand</td>
<td>1.2-1.7-fold increase in the second-order root mycorrhizal rate</td>
<td>[4]</td>
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<tr>
<td>Laccaria bicolor/laccata</td>
<td>Pseudomonas fluorescens, Pseudomonas sp., Bacillus sp.</td>
<td>Pseudostugia menziesii</td>
<td>L. laccata sporocarps and mycorrhizas</td>
<td>1.2-1.4-fold increase in the ectomycorrhizal infection</td>
<td>[5]</td>
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<tr>
<td>Laccaria frater, Laccaria laccata</td>
<td>Bacillus sp., Pseudomonas sp.</td>
<td>Eucalyptus diversicolor</td>
<td>Sporocarps and ectomycorrhizae of L. fraterl</td>
<td>1.8-3.9-fold increase in the ectomycorrhizal infection</td>
<td>[6]</td>
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<tr>
<td>Lactarius rufus</td>
<td>Paenibacillus sp., Rhizosphere</td>
<td>Pinus sylvestris</td>
<td>L. rufus ectomycorrhizas</td>
<td>1.9-2.4-fold increase in the ectomycorrhizal infection</td>
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<td>Pisolithus alba</td>
<td>Pseudomonas monteillii, Pseudomonas resinosorans</td>
<td>Acacia holosericea</td>
<td>Rhizosphere</td>
<td>2.2-fold increase in the ectomycorrhizal infection</td>
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<td>Pisolithus sp.</td>
<td>Flourescent pseudomonads</td>
<td>Acacia holosericea</td>
<td>Rhizosphere, mycorrhizosphere, galls</td>
<td>1.7-2.3-fold increase in the ectomycorrhizal infection</td>
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<td>Rhizopogon luteolus</td>
<td>Unidentified bacterial isolates</td>
<td>Pinus radiate</td>
<td>Rhizopogon luteolus ectomycorrhizas</td>
<td>1.2-2.3-fold increase in the ectomycorrhizal infection</td>
<td>[9]</td>
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<td>Different species of Scleroterma and Pisolithus</td>
<td>Pseudomonas monteillii</td>
<td>Different Acacia species</td>
<td>Rhizosphere,</td>
<td>1.4-2.8-fold increase in the ectomycorrhizal infection</td>
<td>[10]</td>
</tr>
<tr>
<td>Suillus luteus</td>
<td>Bacillus</td>
<td>Pinus sylvestris</td>
<td>S. luteus ectomycorrhizas</td>
<td>2.1-fold increase in the percentage of arbuscular mycorrhizal infection of the root</td>
<td>[11]</td>
</tr>
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</table>

Multiple mechanisms exhibited by mycorrhiza helper bacteria

The type of mycorrhization both ecto and endo are dependent upon helper bacteria. Bacteria associated with AMF spores colonize mainly the outer wall layer and rarely penetrate into the inner layers. Nevertheless, some bacteria have been found in the cytoplasm of AMF spores. The role of AMF spore associated bacteria is not clear. They could stimulate spore germination by eroding spore walls by producing stimulatory compounds such as CO₂ and other volatiles, or by influencing AMF phosphorus acquisition. Root exudation could enhance spore germination by stimulating the growth of bacteria beneficial for AMF. Lehr et al. [12] found that Streptomyces sp. AcH505 acting as MHB suppressed the plant defence response. Streptomyces sp. AcH505 acts as an MHB by expressing a combination of
mechanisms which add to existing signalling mechanisms operating between the mycorrhizal fungus and plant symbionts. The metabolite, auxofuran, appears to stimulate presymbiotic growth of the fungus, adding to the action of root-derived metabolites which direct growth of mycorrhizal hyphae towards the root. MHB promote the establishment of symbiosis by stimulating mycelial extension; increasing root-fungus contacts and colonization; and reducing the impact of advers environmental conditions on the mycelium of the mycorrhizal fungi. Using established microbiological techniques, the screening process showed the presence of bacteria species from Firmicutes, Actinobacteria, Alphaproteobacteria and gamma proteobacteria represented by six genera namely - Bacillus, Paenibacillus, Curtobacterium, Rhizobium, Enterobacter and Stenotrophomonas. Among the isolates, Bacillus and Rhizobium have been previously reported as potential MHBs. [13,14] The major hypotheses by Garbaye (1994) provide evidence for a multitude of MHB mechanisms (Fig. 1). A direct effect of the helper bacteria on the root receptivity to mycorrhizal fungi has been frequently evoked in the different papers that deal with the mechanisms of the mycorrhiza helper effect. However, the main mechanism favoured so far in all these studies is the direct effect of helper bacteria on the presymbiotic survival and growth of the mycorrhizal fungi in the soil (Founoune et al. 2002). At the molecular level, this mechanism likely relies on the modification of the fungal nutrient use efficiency and on the regulation of the fungal cell cycle by the helper bacteria. Little is known about the signal molecules produced by the helper bacteria, the fungal factors that recognize the bacterial signal molecules as well as the fungal gene networks underlying the fungal–bacterial interactions. It was demonstrated the involvement of bacterial Nod factors in the helper effect of Bradyrhizobium japonicum on the Glomus mosseae–soybean endomycorrhizal symbiosis. It twas reported a relationship between the mycorrhizal formation on Pseudotsuga menziesii (Douglas fir) and Pseudomonas fluorescens BBC6, where there was a decrease in mycorrhizal colonization with increasing bacterial doses. [15]

Paenibacillus spp. has been shown to be capable of degrading complex carbohydrates. For instance, the production of cellulolytic enzymes, such as xylanases and cellulases that degrade insoluble polysaccharides (cellulose and xylan) by Paenibacillus curdlanolyticus.

![Fig.1: Simplified representation of the rhizosphere pointing out five possible ways by which a bacterium can promote mycorrhizal establishment: (1) effect on the root receptivity to mycorrhizal fungi; (2) effect on the root-fungus recognition and attachment; (3) effect on the fungus survival and growth; (4) effect on the physico-chemical properties of the soil; and (5) effect on the germination of fungal propagules.](image)

Close associations of Rhizobium with mycorrhizal plants have previously been described (Garbaye, 1994) to indirectly influence mycorrhizal formation. The isolation of a close relative of Rhizobium from the ascocarp harvested could have originated from leguminous trees such as Acacia erioloba at the site of ascocarp collection. The isolation of Rhizobium from T. borchii ascocarps was previously reported. Similarly, nitrogen fixation was confirmed by in Tuber magnatum. [17] The significant stimulatory effect of Rhizobium tropici on the growth of K. pfeillii mycelium could be attributed to their capability to supply the nitrogen requirement of the fungus. Some fungal regulated genes were discovered in Pseudomonas aeruginosa during their investigation of the
pathogenicity of the bacterium on the *Candida albicans* fungus. [18] reported that *Bacillus megaterium* EG24 and other isolates of *Bacillus*, identified on the basis of 16S rDNA sequencing showed cellulase, pectinase and protease activities. These bacteria associated with spores of *Gigaspora margarita* showed their potentiality for stimulating root mycorrhizal stimulation and seedling growth of neem plant

**Applied prospects of mycorrhiza helper bacteria in future**

As it proven that the potential practical application of mycorrhiza helper bacteria in agriculture and forestry, there is a need for the identification of new screening criteria allowing a quick and efficient selection of performing bacterial isolates. The screening strategies used so far are too time-consuming. Therefore, any molecular approach leading to identify fungal marker genes, such as master regulators specific for the mycorrhiza helper effect, will have crucial practical outputs, especially for the improvement of tree yields in poor forest soils [19] can undisputably be considered as pioneers in the analysis of the molecular mechanisms underlying the interactions between helper bacteria and mycorrhizal fungi. However, many complementary studies on different mycorrhizal fungi are still required in order to identify and validate the preliminary marker genes that have been identified so far. In parallel to these studies, one should pay attention to the research on fungal–bacterial interactions presently developing in other fields, such as plant protection and medicine. Comparing the mycorrhizal fungus genes differentially expressed in the presence of helper bacteria to the ones identified in these other model systems would allow us to overcome, more efficiently, the bottlenecks in studying the mechanisms of the mycorrhiza helper effect. Conversely, any breakthrough in the understanding of the mechanisms underlying the interactions between helper bacteria and mycorrhizal fungi will undoubtedly benefit the other research areas where fungal–bacterial interactions play a major practical and economical role. More work on mycorrhiza helper bacteria should be dedicated to model mycorrhizal fungi that are of obvious commercial interest as well as being of use as research laboratory models. In addition, growing concern about the pollution of soils, and the resulting trend towards reducing the input of chemicals in plant production, should foster more environmentally friendly practices such as controlled mycorrhization or microbial bioremediation, for instance by using mycorrhizal fungi as carriers of depolluting bacteria. [20] MHB helps mycorrhiza establish symbiotic associations in stressful environments such as those high in toxic metals. In harsh environments, the bacteria assist in acquiring more nutrients such as nitrogen and phosphorus. MHB helps to prevent the uptake of toxic metals including lead, zinc, and cadmium. The bacteria decrease the amount of metals taken up by the plant and also promote detoxifying soil. This converging of scientific and practical interests, supported by the development of genomics, may represent a unique opportunity to place MHB at the forefront of future mycorrhiza research and to boost the more general field of fungal-bacterial interactions in ecosystems.

**REFERENCES**


4. Schrey SD, Schellhammer M, Ecke M, Hampp R, Tarkka MT. *Mycorrhiza* helper bacterium *Streptomyces* AcH 505 induces


