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Ectomycorrhizal Fungi as Biofertilizers in Forestry

José Alfonso Domínguez-Núñez and Ada S. Albanesi

Abstract

Ectomycorrhizal (ECM) fungi play a fundamental role in the nutrient cycle in terrestrial ecosystems, especially in forest systems. In this chapter, the value of ECM fungi is reviewed from a global framework, not only to increase the production of edible fruit bodies and biomass of plants but also for the regular practices of reforestation and restoration of ecosystems, with implicit applications in biofertilization, bioremediation, and control of soil pathogens. Ecological functions of the ECM fungi are briefly reviewed. The direct implications of the ECM fungi in forestry are described. To do so, its role as a biotechnological tool in forest nursery production is briefly analyzed, as well as the role of mycorrhizal helper bacteria (MHB). Subsequently, the direct role as biofertilizers of the ECM fungi in forest management is discussed: reforestation, plantation management, and ecosystem restoration. The importance of ECM fungi to increase the tolerance of plants against biotic or abiotic stresses is analyzed.

Keywords: forestry, ectomycorrhiza, restoration, sustainable development, nutrients

1. Introduction

It was Albert Bernhard Frank (1885), a forest pathologist, who for the first time introduced the term mycorrhiza. In the Greek language, “mykes” refers to fungus and “rhiza” refers to root. Since Frank’s description of mycorrhizal association in the 1880s [1], a lot of work has been generated by different investigators as a consequence of which it is estimated that 86% of terrestrial plant species are benefited as they acquire their mineral nutrients via mycorrhizal roots [2]. These groups of fungi establish a symbiotic relationship with the roots of plants, called mycorrhizas. Frank established two large subdivisions of mycorrhizas, ecto- and endomycorrhizas. Ectomycorrhizal fungi form mantle and Hartig network of intercellular hyphae in the roots of forest species. Endomycorrhizas are classified as arbuscular mycorrhizas, ericoid mycorrhizas, arbutoid mycorrhizas, monotropoid mycorrhizas, ectendomycorrhizas, or orchid mycorrhizas [3]. The Arbuscular Mycorrhizal fungi (AM) form arbuscules and vesicles, they are more variable than ECM fungi since they form symbiosis with trees and herbaceous plants. Each of these categories is characterized by the invasion of plant root cells by fungal hyphae but differs in the nature of intracellular hyphal development [4, 5].

Ectomycorrhizal fungi are predominantly *Basidiomycetes*, some *Ascomycetes*, and a very few *Zygomycetes*. In these symbiotic structures, the Hartig network is the interface for the metabolic exchange between the fungus and the root. The mycorrhizal mantle is connected to the filaments of fungi that extend into the soil

(extraradical mycelium), directly involved in the mobilization, absorption, and translocation of soil nutrients and water to the roots. Molecular clock analysis on the reconciled tree suggested that ECM fungi evolved far later than the appearance of the last common ancestor of brown and white rot fungi about 300 mya [6]. These results supported the long-standing hypothesis that ECM fungi evolved polyphyletically from multiple saprophytic species. More than 7000 species of fungi form ectomycorrhizas [7], many of them with important commercial trees such as poplar, birch, oak, pine, and spruce [8]. The reproductive structures (fruiting bodies) of the macromycetes are known as mushrooms when they grow in the soil and, like truffles, when they grow underground.

The community of mycorrhizal fungi can be determinant in the structure of the plant community [9]. Therefore, the identification of the mycobiont partner and its functional structure [10] are fundamental to understand the ecological importance of this symbiotic relationship. ECM fungal diversity studies were initially based on studies of fruiting bodies and, more recently, on the direct identification of ectomycorrhizal morphoanatomical characters [11]. Despite recent advances in the use of molecular techniques, there are still many advantages associated with classical methods for studying ECM fungal diversity. For the recognition of fungal relationship and type of mycorrhizal association is advantageous over molecular method [7]. Sometimes morphoanatomical-based taxonomy is not well supported by molecular taxonomy. To overcome such discrepancy, the combined approach of morphoanatomical and molecular characterization of ectomycorrhizas in combination with phylogeny was applied [12].

Most of the cultivated species of edible fungi are saprophytes, and only some of them are ECM fungi [13]. The tickets (*Boletus edulis*), the chanterelles (*Cantharellus* spp.), the matsutake mushroom (*Tricholoma matsutake*), and the truffle (many species of the *Tuber* genus) are some ECM fungi for which the crop has been studied [14–16]. The black truffle or Périgord, *Tuber melanosporum*, is widely grown, while other species of ECM mushrooms have not yet been cultivated, including fungi porcini (*Boletus edulis* S.) and the high-priced Italian fungus, white truffles (*Tuber magnatum*).

2. Ecological functions of ECM fungi

In different forest ecosystems, ECM fungi have been reported to play an important role in seedling survival, establishment, and growth [3, 17, 18]. Researches have confirmed that ECM fungi play a key role in terrestrial ecosystems as drivers of global carbon and nutrient cycles [19].

Some of these traditionally known functions of the ECM fungi on the ecosystem are:

ECM fungi increase the water and nutrient supply plant, extending the volume of land accessible to the plants.

Different fungal species (drought-sensitive hydrophilic or drought-tolerant hydrophobic) can have different effects on hydraulic redistribution patterns [20]. The mechanisms to enhance the acquisition of P by tree mycorrhizal roots are the extension of extramatrical mycorrhizal hyphae, the increase of inorganic P transfer, the increase of inorganic P transporters in the fungus/soil interface, the mobilization of organic P (labile) by emission of phosphatases, and the mobilization of mineral insoluble P by the emission of organic acids (LMWOAs) [21, 22].

The mechanisms of improvement in nitrogen (N) absorption would be the intervention in the mineral N cycle (NH_4^+ , NO_3^-) and the assimilation of organic N (by emitting proteases, chitinases, and others) [23, 24].

Currently, recent advances in the knowledge of nutrient translocation processes in the fungus-plant and fungus-soil interaction are especially interesting, in particular the priority role of transporters of P, N, and C [25]. The inorganic P and mineral or organic forms of N, such as NH_4^+ , NO_3^- , and amino acids (AA), are absorbed by transporters specialized located in the fungal membrane in the extraradical mycelium. $\text{NH}_3/\text{NH}_4^+$ and inorganic P (from polyphosphates) are imported from the symbiotic interface to the cells of the plant through selective transporters. Transporters of hexoses import carbon of plant origin into the fungus. The nutritional strategies seem to be different between symbiotic and pathogenic fungi, for example, in the translocation of C. Even different transport strategies have been found between ECM symbionts *Ascomycota* and *Basidiomycota*. The understanding of the different systems of transporters or nutrient channels involved both at the level of the extraradical mycelium and at the level of the symbiotic interface will clarify in the future the processes of nutrition in the plant-fungus and fungus-soil interaction. Also, looking at the fungal factors in the establishment of the symbiotic relationship, chitin-related molecules seem to be shared by pathogenic and arbuscular mycorrhizal fungi, opening the question of whether they could also function in signaling in ectomycorrhizal symbioses [26].

On the other hand, the ECM colonization of the root can provide protection against soil pathogens [27]. Also, the non-nutritive benefits to plants due to changes in water relations, the level of phytohormones, the assimilation of carbon, etc. have already been verified [3]. The carbon is transferred through the ECM fungal mycelium that connects different species of plants. This can reduce competition among plants and contribute to the stability and diversity of ecosystems [28]. The extraradical mycelium of the ECM fungi provides a direct pathway for the translocation of photosynthesized carbon to microsites in the soil and a large surface area for interaction with other microorganisms [29, 30]. Recently, Hupperts et al. [31] proposed two competing models to explain carbon mobilization by ectomycorrhizal fungi. “Saprotrophy model”, where decreased allocation of carbon may induce saprotrophic behaviour in ectomycorrhizal fungi, resulting in the decomposition of organic matter to mobilize carbon and second, “nutrient acquisition model”, where decomposition may instead be driven by the acquisition of nutrients locked within soil organic matter compounds. Moreover, epigeous and hypogaeal sporocarps of ECM fungi are important food sources for placental and marsupial mammals [32]. The ectomycorrhizal roots, the mycelium, and the fruiting bodies of the fungi are important as food sources and habitats for invertebrates [33]. The hyphal networks produced by ECM fungi significantly alter and improve the structure of the soil [34]. In a global way, the ECM fungi improve the plant tolerance to (biotic and abiotic) environmental stresses.

3. Applications: ECM fungi to forestry

Much of our understanding of the functions of ECM fungi has come from research directed toward practical application in forestry. Some of the most common criteria considered for the selection of a most valued species or strain of ECM fungi (some of them implicit in others) are the abiotic criteria: climatic conditions such as temperature, insolation, and humidity; improvement of soil properties, such as texture and permeability; abiotic soil stress mitigation; soil contamination mitigation; soil metal mobilization; or nutrient cycling. There may also be criteria regarding the host, such as the plant/fungus specificity, the improvement of plant health, or the increase in the biomass of the plant. Finally, there are criteria regarding the fungus, such as abundance, effectiveness, propagules competitiveness,

fungus growth rate, or edibility. Other criteria may be the conservation of native biodiversity, the functioning of the ecosystem, human health, food, nutraceutical value, etc. [30, 35].

3.1 ECM fungi in forest nurseries

Since the late 1950s, mycorrhizal fungi were utilized as biofertilizers to promote plant growth, because of their ability to increase the plant uptake of P, N, mineral nutrients, and water [36–38]. The idea of inoculating ECM fungi on seedlings in plant nurseries was developed by Fortin [39]. Vozzo and Hacskeylo [40] while working on ECM in the United States experimentally demonstrated that field survival and growth of tree seedlings with specific potential ECM enhance the performance of seedlings and contribute to the proper functioning of forest ecosystems.

Although successful inoculation of tree seedlings (already planted) in the field has been known, nursery inoculation is more common. Seedlings inoculated in the nursery can establish a healthy ECM system before planting. The challenge in the controlled synthesis of the ectomycorrhizal symbiosis is to produce a quality mycorrhizal plant, only colonized by the desired fungus. Accurate identification of the inoculum used and avoiding contamination during the growth of the inoculated plants are essential parts of the production process to avoid the introduction of unwanted species and to avoid the mixing of their genetic material with indigenous species [41]. The appropriate selection of suitable plant-host species is essential for the success of mycorrhization [42]. Relatively fast-growing fungi are generally preferred for inoculation because of their short incubation period. Unfortunately, many otherwise desirable ECM fungi grow slowly. According to Marx [43], fresh cultures are preferred to cultures repeatedly transferred and stored for several years. He further suggests passing important fungus cultures through a host inoculation and mycorrhiza formation followed by re-isolation, every few years to maintain mycorrhiza-forming capacity. Moreover, fungi, which produce large hyphal stands of rhizomorphs in the culture of the soil, may be superior in soil exploration and mineral uptake to those which lack rhizomorphic growth. On the other hand, the fruiting of the ECM fungi species is not based solely on the mycorrhizal state of the seedlings. After planting, in addition to the presence of indigenous competitors, the biotic and physicochemical characteristics of the soil also influence the persistence and spread of the cultivated fungus [44]. The type of ECM material used for inoculation can affect the success of a mycorrhizal inoculation program. In addition to remaining viable during storage and transport, the inoculant must also maintain its infectivity for several months after its introduction [45].

There are three main sources of fungal inoculum: soil, spores, and mycelium.

Initially, the soil or humus collected from the mycorrhizal plantation area was frequently used. Its main disadvantage is the lack of control of ECM species in the soil or of microorganisms and harmful germs. Another problem with this type of inoculant is that large amounts of soil are required to inoculate nursery plants. This method is widely used in developing countries, although it is currently discarded in mycorrhization programs. Also, planting mycorrhizal “nurse” seedlings or incorporating chopped roots of ECM hosts into nursery beds as a source of fungi for neighboring young seedlings has been successful [46].

Other sources of inoculum are the spores of fruit bodies collected in the field. The main advantages are that the spores do not require the extension of the aseptic culture and that the spore inoculum is not heavy [47]. Most of the recent research has been with *Pisolithus tinctorius*. Inoculation with spores of *Rhizopogon* species also appears promising. Abundant *Rhizopogon* mycorrhizas formed on seedlings produced from the coated seed of *Pinus radiata* D. Don with

basidiospores of *Rhizopogon luteolus* [5]. However, it has three main drawbacks: (a) significant quantities of fruiting bodies are required and may not be available each year, (b) the success of the inoculation is highly dependent on the viability of the spores, and (c) the lack of genetic definition. Freeze-drying and storage at a low temperature in the dark is helpful to maintain its viability. The spores can be mixed with physical supports before the soil inoculation; suspended in water and soaked in the soil; sprinkled, sprayed or pelleted, and emitted to the ground; and encapsulated or coated on the seeds, and they can be embedded in hydrocolloid chips [47].

The most appropriate inoculum is the use of hyphae in a solid or liquid medium or substrate. Hyphae are cultivated mainly from sterile parts of fruiting bodies, less frequently from mycorrhiza due to their low (approx. 5–20%) success rate [48] and rarely from sclerotia [49] or sexual spores [50]. It is considered the most appropriate method since it allows the selection of particular strains of a fungus previously tested for its ability to promote the growth of plants [43]. Many species do grow well in culture, e.g., most species of *Suillus*, *Hebeloma*, *Laccaria*, *Amanita*, *Rhizopogon*, and *Pisolithus* genus. Liquid substrates have the advantage over solids because they are easily mixed and produce more uniform conditions for crop growth, but the risk of bacterial contamination and costs are higher [45]. On the other hand, the main advantages of the solid medium [51] are the reduction of bacterial contamination due to the lower water content, the low costs of the equipment, and the simplified design of the bioreactors. The main drawbacks of the use of mycelial inocula are that several species of ECM fungi are difficult to grow under laboratory conditions, or growth is very slow (due to the absence of their symbiont), and it is not always easy to produce large amounts of inoculum viable for large-scale nursery inoculation programs. Some advances have been made using mycelium encapsulated in “beads” of calcium alginate (e.g., [52]), but they have to be refrigerated. Inoculant beads can remain viable for several months under refrigeration, although the results vary between fungal species. For several species, the mycelial inoculum has been tested with trees of economic interest. This technique has great potential for the inoculation of seedlings in reforestation programs. For example, Rossi et al. [45] designed a bioreactor with the capacity to produce inoculum for 300 000 seedlings, enough to reforest 200 hectares. Based on a global demand of 3.0 billion cubic meters of wood, an estimated 4.3 tons of mycelium would be needed to inoculate 12 billion seedlings (5 g of dry mycelium per plant [45]). An advantage of alginate gel is the possibility of preparing a multimicrobial inoculant.

3.1.1 *Ectomycorrhizal helper bacteria*

The concept of “mycorrhiza helper bacteria” (MHB) was introduced in a “Tansley Review”: Helper Bacteria—a new dimension of mycorrhizal symbiosis [53], which has led to new research in the plant-fungus model system, as for the meaning of these bacteria that promote the formation of mycorrhizas and cause many physiological effects of mutualistic interaction. In general, the ability of some microorganisms to influence the formation and functioning of the symbiosis is known, through activities of various kinds such as the activation of infective propagules of the fungus in presymbiotic stages [54], facilitating the formation of entry points in the root [55] and increase of the growth rate [56]. The MHB improve mycorrhiza formation, although the same MHB can benefit mycorrhization for certain fungi and be negative for others [57]. The above reflects the fungal specificity by isolate, which exemplifies the genetic distance between isolates of different origin.

Among the mechanisms presented by the MHB are:

- a. Promotion of the establishment of the symbiosis by stimulation of the mycelial extension. The germination of spores and mycelial growth are improved by the production of growth factors [58].
- b. Increased contact and colonization root-fungus: increase in the number of lateral roots, mediated by the production of phytohormones [59] and the improvement of radical colonization by induction of flavonoid production [60].
- c. Reduction of the impact of adverse environmental factors on the mycelium of the mycorrhizal fungus. Bacteria can detoxify soils, restoring their conductivity, similarly freeing them from contamination generated by heavy metals [61], and reducing the concentrations of phenolic antagonist compounds produced by the same mycorrhizal fungi [62]. The rhizospheric microorganisms also have an effect on the growth of the plants, reaching a synergistic effect, where the presence of the microfungus and the other microorganism produces an increase in the growth, vigor, and protection of the plant [63]. These effects are based on activities such as the acquisition of nutrients, inhibition of the growth of pathogenic fungi [64], and improvement of the root ramification [65]. In recent years, a potential capacity of bacteria associated with ectomycorrhizas to fix atmospheric nitrogen has been suggested [66]. Several studies suggest a real possibility that the bacteria present in mycorrhizal tissues contribute to the nutritional needs of both the fungus (ascocarp development) and consequently the plants, by providing them with available nitrogen derived from atmospheric nitrogen (N_2).

MHB belong to a wide range of genera (*Burkholderia*, *Paenibacillus* [67]; *Pseudomonas*, *Bacillus* [68]; *Streptomyces* [69]). However, the molecular mechanisms by which MHB induce the growth of ECM fungi are not well described. Recently, changes in expression of genes involved in the development of certain ECM fungi have been studied at the molecular level in confrontations with MHB [70–73].

Research in mycorrhizas should, therefore, strive toward an improved understanding of the functional and molecular mechanisms involved in interactions in the mycorrhizosphere, in order to develop ad hoc biotechnology that allows the application of optimized combinations of microorganisms as effective inoculators within sustainable systems of plant production [74].

3.1.2 Polymicrobial formulations

A polymicrobial formulation containing a diverse mixture of beneficial rhizosphere microorganisms with multiple functionalities is attractive because combining different classes of soil organisms can take advantage of multiple plant growth-promoting mechanisms and could be applied to multiple crops [75–79]. A key concept in constructing effective polymicrobial multifunctional formulations is the selection and use of a right combination of rhizosphere bacteria and fungi that are mutually compatible, have complementary functionalities, effectively colonize the rhizosphere of the crop(s) of interest, and bring about a synergistic promotion of growth and yield of crop(s) [75, 80–82]. It is to be expected that well-designed multifunctional formulations such as the one described would be a welcome addition to the fast-growing inoculant enterprises worldwide. Such an inoculant is also expected to be eco-friendly and suitable for organic farming and other integrated production systems, where synthetic fertilizer inputs are not allowed or restricted by law. However, construction of such complex formulations is technically demanding [83].

Ectomycorrhizal fungi exhibit synergistic interactions with other plant-beneficial organisms such as symbiotic N₂-fixers. For example, ectomycorrhizal symbiosis enhanced the efficiency of inoculation of two *Bradyrhizobium* strains on the growth of legumes [84]. It is also of interest that similar synergies were seen when AM fungus (*Glomus mosseae*), ECM fungus (*Pisolithus tinctorius*), and *Bradyrhizobium* sp. were used together to inoculate *Acacia nilotica*; enhancement of N₂ fixation, growth, and dry biomass were observed when all three organisms were present [85, 86].

Also, using plant growth-promoting microorganism (PGPM) strains that form stable and effective biofilms could be a strategy for producing commercially viable inoculant formulations [78, 87]. A majority of plant-associated bacteria found on roots and in the soil are found to form biofilms [88]. Bacterial, fungal, and bacteria/fungal biofilms were suggested as possible inoculants. This is a novel and interesting idea, but to what extent this approach would be practiced remains to be seen [83].

3.2 Application of ECM fungi in forest management: restoration of ecosystems

The inoculation of ECM fungi can be done with the objective of producing edible carpophores but also because of its considerable value in forest management; in particular, they have had great importance in reforestation programs where it was expected that the quality and economic productivity of the plantations would increase [89]. The success of the plantations with mycorrhizal seedlings from the nursery depends on their ability to quickly access the nutrients and water available within the soil matrix [90]. The relationships between the various native edible ECM fungi have been, until relatively recently, insufficiently considered in the strategies of forest management [91].

In ectomycorrhizal plantations (productive or conservation reforestations), a consequence of the recognition of the advantages of fungal diversity in ecosystems will be an increase in the refusal to introduce potentially dominant species in mixed communities. On the other hand, unfortunately, it seems that many of those fungi selected for optimal colonization in the nursery have been poor competitors in the field, especially when the planting sites contained indigenous populations of mycorrhizal fungi. There are several possible explanations for the inoculation failure (from the nursery) to produce beneficial effects in the planting sites. Probably, among the most important of these is the inability of inoculum introduced to persist in the roots of the plant after the transfer of the nursery to the field. The soil conditions experienced in the nursery and with the plant growing in a container are very different from those of most of the planting sites; in addition, the raising, storage, and transport of seedlings can reduce the vigor of fine roots and their fungal associates. Species such as *Pisolithus tinctorius* (15 sub spp.), in circumstances such as degraded environments, with absence or scarcity of autochthonous mycorrhizal populations, have achieved the greatest success in inoculation programs [92]. In the case of edible ECM fungi, such as *Tuber melanosporum* (black truffle), the establishment of mycorrhizal plantations has always aimed at the production of carpophores, leaving aside the contribution of ecological functions of the symbiosis (in the plant, in the soil, and, in general, in the ecosystem) [93]. The example of mycorrhizal plantations for truffle production has been generally successful [94], obtaining productions from 6 to 7 years of implantation.

In the restoration of ecosystems, the biofertilization, the bioremediation, and the biocontrol of soil pathogens are prominent roles of the ectomycorrhizal fungi. Degraded ecosystems are the result of a wide range of characteristics and factors related to unfavorable land management or industrial activities. Environmental degradation of the soil is increasing worldwide at an alarming rate due to erosion,

acidity, salinization, compaction, depletion of organic matter, and water scarcity. On the contrary, in a healthy ecosystem, there is a balanced microbiota of the soil, in such a way that the potential of pathogenic and mycorrhizal fungi coexists in apparent harmony. Ectomycorrhizal fungi can survive in extreme habitats with high or low temperature [95, 96], salt and metal concentration [97, 98], drought [99], and other circumstances related to the degradation of the ecosystem. The importance of ECM fungi in the balance of the ecosystem can be enormous, since they can be used to increase the tolerance of plants against biotic or abiotic stresses, especially their capacity to fix heavy metals or to degrade a wide variety of persistent organic compounds; to interact with soil bacteria; to attack fungi, bacteria, and pathogenic nematodes; and to improve the vegetative growth and the nutritional status of its symbiont plant.

It has been documented by several authors that mycorrhizal fungi improve the disease resistance of their host plant primarily by direct competition, enhanced or altered plant growth, nutrition and morphology, induced resistance, and development of antagonist microbiota. Direct competition or inhibition is reported to be due to the production and release of antibiotics and physical sheathing by the mantle of ECM [27, 100–102]. For example, ECM fungi have been shown to protect trees from *Phytophthora cinnamomi* infection along with supporting their survival and growth in comparison to non-mycorrhizal seedlings [35, 101, 102]. Thus, ECM fungi can also be used as a fungicide in nursery plantations for better growth, survival, and establishment of seedlings.

Under drought stress, ectomycorrhizal symbiosis has been documented to possess a remarkable capacity to the uptake of water and alter hydraulic properties of plant roots by altering both apoplastic and symplastic pathways and by their impact on plant aquaporins (AQPs) [103–106]. A symbiosis between plants and ECM fungi has been documented to help plants to cope with salt stress [97, 107–109]. Li et al. [110] reported that there is ECM fungus-mediated remodeling of ion flux which helps to maintain K^+/Na^+ homeostasis by increasing the release of Ca^{2+} . Also, ECM fungi have been reported to change the plant phytohormone balance during salt stress [111, 112]. Research efforts are still in progress to select new pioneer symbiotic couples for land reforestation [113].

Till date, most studies have indicated that ECM plants accumulate less metal inside their tissue and grow better than non-mycorrhizal plants when exposed to heavy metal stress [114–118]. Also, Meharg and Cairney [119] revised potential ways in which ectomycorrhizal fungi might support rhizosphere remediation of persistent organic pollutants (POPs). Krupa and Kozdrój [120] documented the importance of mycorrhizal fungi in forming an efficient biological barrier for checking the movement of heavy metals into the host tissues. Recently, the importance of LMW organic acids and metal chelating agents (such as siderophores) from ECM fungi in the fixation of metal ions and their transmission or not to the root of the host plant has been described [121]. The cellular mechanisms involved in detoxification of heavy metals by mycorrhizal fungi include biosorption of metals to fungal cell wall, chelation of metal ion in the cytosol by compounds such as glutathione and metallothioneins, metal exclusion mechanisms in metal-tolerant ECM fungi, and the compartmentation of metals in the vacuole, where metal ions are probably complexed in a chemically inactive form [98, 118, 122, 123].

4. Conclusions

The ectomycorrhizal fungi are predominantly *Basidiomycetes* and *Ascomycetes*, which establish a symbiotic relationship with the roots of forest plants, and these

are directly involved in the mobilization, absorption, and translocation of soil nutrients and water to the roots. Most of the known cultivated species of edible fungi are saprophytes, and some of them are ectomycorrhizal fungi, but there is a promising potential in the study and knowledge of new species of ECM fungi as potential wild collected edible mushrooms. ECM fungi play a key role in terrestrial ecosystems as drivers of global carbon and nutrient cycles; in the fungus-plant interface, the role of C and nutrient transporters seems a priority. Research in ectomycorrhizal fungi should focus on better understanding the functional and molecular mechanisms involved in fungus-plant and fungus-soil interactions. For decades, our understanding of the functioning of ectomycorrhizal fungi has allowed us their application in the forest area. In the nursery, the inoculation of ECM fungi is a more common method to produce ectomycorrhizal forest seedlings, and the mycelial inoculation has great potential in reforestation programs. We should aim to find the appropriate technology for the commercial techniques of multiplication and large-scale inoculation of the mycorrhizal inoculum and the application of optimized combinations of plant-microorganisms (e.g., MHB, PGPB) adopted under well-defined environmental and soil conditions. The role of ECM fungi as biofertilizers in bioremediation or biocontrol in plantations, reforestation, and environmental restoration has been fundamental up to now, and its importance in the balance of the ecosystem can be enormous, increasing the tolerance of plants against biotic and abiotic stress. The application of ectomycorrhizal fungi in current environmental problems as the oaks or pines decline, or the phytoremediation of contaminated soils, seems promising. Research is still underway to select new pioneer symbiotic relationships for land restoration and reforestation.

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References

- [1] Frank B. Ueber die auf Wurzelsymbiose beruhende Ernährung gewisser Bäume durch unterirdische Pilze. *Berichte der Deutschen Botanischen Gesellschaft*. 1885;3:128-145
- [2] Brundrett MC. Mycorrhizal associations and other means of nutrition of vascular plants: Understanding the global diversity of host plants by resolving conflicting information and developing reliable means of diagnosis. *Plant and Soil*. 2009;320:37-77. DOI: 10.1007/s11104-008-9877-9
- [3] Smith SE, Read DJ. *Mycorrhizal Symbiosis*. 3rd ed. London: Academic Press; 2008. p. 787
- [4] Peterson RL, Massicotte HB, Melville LH. *Mycorrhizas: Anatomy and Cell Biology*. Wallingford, Oxon: CABI Publishing, CAB International; 2004
- [5] Sharma R. Ectomycorrhizal mushrooms: Their diversity, ecology and practical applications. In: Varma A, Prasad R, Tuteja N, editors. *Mycorrhiza - Function, Diversity, State of the Art*. Switzerland: Springer International Publishing; 2017. pp. 99-131
- [6] Marcel G, van der Heijden A, Martin FM, Selosse MA, Sanders IR. Mycorrhizal ecology and evolution: The past, the present, and the future. *New Phytologist*. 2015;205:1406-1423. DOI: 10.1111/nph.13288
- [7] Rinaldi AC, Comandini O, Kuyper TW. Ectomycorrhizal fungal diversity: Separating the wheat from the chaff. *Fungal Diversity*. 2008;33:1-45
- [8] Wiensczyk AM, Gamiet D, Durall DM, Jones MD, Simard SW. Ectomycorrhizas and forestry in British Columbia: A summary of current research and conservation strategies. *B.C. Journal of Ecosystems and Management*. 2002;2:1-20
- [9] Fitter AH. Darkens visible: Reflections on underground ecology. *Journal of Ecology*. 2005;93:231-243
- [10] Agerer R. Exploration types of ectomycorrhizas. A proposal to classify ectomycorrhizal mycelial systems according to their patterns of differentiation and putative ecological importance. *Mycorrhiza*. 2001;11:107-114
- [11] Horton TR, Bruns TD. The molecular revolution in ectomycorrhizal ecology: Peeking into the black-box. *Molecular Ecology*. 2001;10:1855-1871
- [12] Mrak T, Kühdorf K, Grebenc T, Štraus I, Münzenberger B, Kraigher H. *Scleroderma areolatum* ectomycorrhiza on *Fagus sylvatica* L. *Mycorrhiza*. 2017;27:283-293. DOI: 10.1007/s00572-016-0748-6
- [13] Savoie JM, Largeteau ML. Production of edible mushrooms in forests: Trends in development of a mycosilviculture. *Applied Microbiology and Biotechnology*. 2011;89:971-979
- [14] Chang ST, Hayes WA. *The Biology and Cultivation of Edible Mushrooms*. New York: Academic; 1978
- [15] Chevalier G. The truffle cultivation in France: Assessment of the situation after 25 years of intensive use of mycorrhizal seedlings. In: *Proceedings of the first International Meeting on Ecology, Physiology and Cultivation of Edible Mycorrhizal Mushrooms*. 3-4 July; Uppsala, Sweden: Swedish University of Agricultural Sciences; 1998
- [16] Bencivenga M. Ecology and cultivation of *Tuber magnatum* Pico. In: *Proceedings of the first International Meeting on Ecology, Physiology and Cultivation of Edible Mycorrhizal Mushrooms*. 3-4 July; Uppsala, Sweden:

Swedish University of Agricultural Sciences; 1998

[17] Sebastiana M, Martins J, Figueiredo A, Monteiro F, Sardans J, Peñuelas J, et al. Oak protein profile alterations upon root colonization by an ectomycorrhizal fungus. *Mycorrhiza*. 2017;**27**:109-128. DOI: 10.1007/s00572-016-0734-z

[18] Tedersoo L, May TW, Smith ME. Ectomycorrhizal lifestyle in fungi: Global diversity, distribution, and evolution of phylogenetic lineages. *Mycorrhiza*. 2010;**20**:217-263. DOI: 10.1007/s00572-009-0274-x

[19] Kumar J, Atri NS. Studies on ectomycorrhiza: An appraisal. *The Botanical Review*. 2018;**84**:108-155. DOI: 10.1007/s12229-017-9196-z

[20] Prieto I, Roldán A, Huygens D, Alguacil MM, Navarro-Cano JA, Querejeta JI. Species-specific roles of ectomycorrhizal fungi in facilitating interplant transfer of hydraulically redistributed water between *Pinus halepensis* saplings and seedlings. *Plant and Soil*. 2016;**406**:15-27

[21] Harley JL, Smith SE. *Mycorrhizal Symbiosis*. Toronto: Academic Press; 1983

[22] Plassard C, Dell B. Phosphorus nutrition of mycorrhizal trees. *Tree Physiology*. 2010;**30**(9):1129-1139. DOI: 10.1093/treephys/tpq063

[23] Harley JL. The significance of mycorrhiza. *Mycological Research*. 1989;**92**:129-139

[24] Chalot M, Plassard C. Ectomycorrhiza and nitrogen provision to the host tree. In: Polacco JC, Todd CD, editors. *Ecological Aspects of Nitrogen Metabolism in Plants*. England: Wiley-Blackwell; 2011. pp. 69-94

[25] Bonfante P, Genre A. Mechanisms underlying beneficial plant-fungus

interactions in mycorrhizal symbiosis. *Nature Communications*. 2010;**27**:1-48. DOI: 10.1038/ncomms1046

[26] Bonfante P. The future has roots in the past: The ideas and scientists that shaped mycorrhizal research. *New Phytologist*. 2018;**220**:982-995

[27] Duchesne LC, Peterson RL, Ellis BE. Pine root exudate stimulates antibiotic synthesis by the ectomycorrhizal fungus *Paxillus involutus*. *New Phytologist*. 1988;**108**:471-476. DOI: 10.1111/j.1469-8137.1988.tb04188.x

[28] Simard SW, Jones MD, Durall DM, Perry DA, Myrold DD, Molina R. Reciprocal transfer of carbon isotopes between ectomycorrhizal *Betula papyrifera* and *Pseudotsuga menziesii*. *New Phytologist*. 1997;**137**(3):529-542

[29] Sun Y-P, Unestam T, Lucas SD, Johanson KJ, Kenne L, Finlay RD. Exudation-reabsorption in mycorrhizal fungi, the dynamic interface for interaction with soil and other microorganisms. *Mycorrhiza*. 1999;**9**:137-144

[30] Suz L, Azul AM, Pino-Bodas R, Martín MP. Ectomycorrhizal fungi in biotechnology: Present and future perspectives. In: Kumar A, Prasad R, editors. *Environment and Biotechnology*. Lambert Academic Publishing. AG & Co; 2012. pp. 472-542

[31] Hupperts SF, Karst J, Pritsch K, Landhäuser SM. Host phenology and potential saprotrophism of ectomycorrhizal fungi in the boreal forest. *Functional Ecology*. 2017;**31**: 116-126. DOI: 10.1111/1365-2435.12695

[32] Claridge AW, May TW. Mycophagy by Australian mammals. *Australian Journal of Ecology*. 1994;**19**:251-275

[33] Fogel R, Peck SB. Ecological studies of hypogeous fungi. I Coleoptera

associated with sporocarps. *Mycologia*. 1975;**67**:741-747

[34] Griffiths RP, Caldwell BA. Mycorrhizal mat communities in forest soils. In: Read DJ, Lewis DH, Fitter AH, Alexander IJ, editors. *Mycorrhizas in Ecosystems*. Wallingford, UK: CAB International; 1992. pp. 98-105

[35] Azul AM, Nunes J, Ferreira I, Coelho AS, Verissimo P, Trovao J, et al. Valuing native ectomycorrhizal fungi as a mediterranean forestry component for sustainable and innovative solutions. *Botany-Botanique*. 2014;**92**(2):161-171

[36] Feldmann F, Hutter I, Schneider C. Best production practice of arbuscular mycorrhizal inoculum. In: Varma A, Kharkwal AC, editors. *Symbiotic Fungi: Principles and Practice*. Vol. 18. Berlin Heidelberg: Springer; 2009. pp. 319-336

[37] Koide R, Mosse B. A history of research on arbuscular mycorrhiza. *Mycorrhiza*. 2004;**14**:145-163

[38] Miransari M. Soil microbes and plant fertilization. *Applied Microbiology and Biotechnology*. 2011;**92**:875-885

[39] Fortin JA. Synthesis of mycorrhizas on explants of the root hypocotyls of *Pinus sylvestris* L. *Canadian Journal of Botany*. 1966;**44**:1087-1092. DOI: 10.1139/b66-116

[40] Vozzo JA, Hacskeylo E. Inoculation of *Pinus caribaea* with ectomycorrhizal fungi in Puerto Rico. *Forest Science*. 1971;**17**:239-245

[41] Murat C, Martin F. Sex and truffles: First evidence of Périgord black truffle outcrosses. *The New Phytologist*. 2008;**180**:260-263

[42] Olivier JM. Progress in the cultivation of truffles. In: Van Griensven LJLD, editor. *Science and*

Cultivation of Edible Fungi. Rotterdam: A. Balkema; 2000. pp. 937-942

[43] Marx DH. Ectomycorrhizal fungus inoculations: A tool for improving forestation practices. In: Mikola P, editor. *Tropical Mycorrhiza Research*. London: Oxford University Press; 1980. pp. 13-71

[44] Hortal S, Pera J, Parladé J. Field persistence of the edible ectomycorrhizal fungus *Lactarius deliciosus*: Effect of inoculation strain, initial colonization level, and site characteristics. *Mycorrhiza*. 2009;**19**:167-177

[45] Rossi MJ, Furigo A, Oliveira VL. Inoculant production of ectomycorrhizal fungi by solid and submerged fermentations. *Food Technology and Biotechnology*. 2007;**45**:277-286

[46] Sim M-Y, Eom A-H. Effects of ectomycorrhizal fungi on growth of seedlings of *Pinus densiflora*. *Mycobiology*. 2006;**34**:191-195

[47] Marx DH, Cordell CE. The use of specific ectomycorrhizas to improve artificial forestation practices. In: Whipps JM, Lumsden RD, editors. *Biotechnology of Fungi for Improving Plant Growth: Symposium of the British*. Cambridge: Cambridge University Press; 1989. pp. 1-25

[48] Molina R, Palmer JG. Isolation, maintenance, and pure culture manipulation of ectomycorrhizal fungi. In: Schenck NC, editor. *Methods and Principles of Mycorrhizal Research*. St Paul, Minnesota, USA: The American Phytopathological Society; 1982. pp. 115-129

[49] Trappe JM. Studies on *Cenococcum graniforme*. An efficient method for isolation from sclerotia. *Canadian Journal of Botany*. 1969;**47**:1389-1390

- [50] Fries N, Birraux D. Spore germination in *Hebeloma* stimulated by living plant roots. *Experimentia*. 1980;**36**:1056-1057
- [51] Cannel E, Moo-Young M. Solid-state fermentation systems. *Process Biochemistry*. 1980;**15**:24-28
- [52] Le Tacon F, Jung G, Mugnier J, Michelot P. Efficiency in a forest nursery of an inoculant of an ectomycorrhizal fungus produced in a fermentor and entrapped in polymeric gels. *Annals of Forest Science*. 1983;**40**:165-176
- [53] Garbaye J. Helper bacteria: A new dimension to the mycorrhizal symbiosis. *New Phytologist*. 1994;**128**(2):197-210
- [54] Azcón-Aguilar C, Barea JM. Arbuscular mycorrhizas and biological control of soil-borne plant pathogens - an overview of the mechanisms involved. *Mycorrhiza*. 1996;**6**:457-464
- [55] Linderman RG. Mycorrhizal interactions with the Rhizosphere Microflora - the rhizosphere effect. *Phytopathology*. 1988;**78**:366-371
- [56] Carpenter-Boggs L, Loynachan TE, Stahl PD. Spore germination of *Gigaspora margarita* stimulated by volatiles of soil-isolated actinomycetes. *Soil Biology and Biochemistry*. 1995;**27**:1445-1451
- [57] Garbaye J, Duponnois R. Specificity and function of mycorrhization helper bacteria (MHB) associated with the *Pseudotsuga menziesii*-*Laccaria laccata* symbiosis. *Symbiosis*. 1992;**14**:335-344
- [58] Keller S, Schneider K, Rüdiger S. Structure elucidation of auxofuran, a metabolite involved in stimulating growth of fly agaric, produced by the mycorrhiza helper bacterium *Streptomyces* Ach 505. *Journal of Antibiotics*. 2006;**59**:801-803
- [59] Bending GD, Poole EJ, Whipps JM, Read DJ. Characterisation of bacteria from *Pinus sylvestris*-*Suillus luteus* mycorrhizas and their effects on root-fungus interactions and plant growth. *FEMS Microbiology Ecology*. 2002;**39**:219-227
- [60] Xie ZP, Staehelin C, Vierheilig H, Iemken W, Jabbouri S, Broughton WJ, et al. Rhizobial nodulation factors stimulate mycorrhizal colonization of nodulating and nonnodulating soybeans. *Plant Physiology*. 1995;**108**:1519-1525
- [61] Brulé C, Frey-Klett P, Pierrat JC, Courier S, Gérard F, Lemoine MC, et al. Survival in the soil of the ectomycorrhizal fungus *Laccaria bicolor* and effect of a mycorrhiza helper *Pseudomonas fluorescens*. *Soil Biology and Biochemistry*. 2001;**33**:1683-1694
- [62] Duponnois R, Garbaye J. Some mechanisms involved in growth stimulation of ectomycorrhizal fungi by bacteria. *Canadian Journal of Botany*. 1990;**68**:2148-2152
- [63] Domínguez JA, Martín A, Anriquez A, Albanesi A. The combined effects of *Pseudomonas fluorescens* and *Tuber melanosporum* on the quality of *Pinus halepensis* seedlings. *Mycorrhiza*. 2012;**22**(6):429-436
- [64] Budi SW, Van Tuinen D, Martinotti MG, Gianiazzi S. Isolation from the *Sorghum bicolor* mycorrhizosphere of a bacterium compatible with arbuscular mycorrhiza development and antagonistic towards soil-borne fungal pathogens. *Applied and Environmental Microbiology*. 1999;**65**:148-150
- [65] Gamalero E, Trotta A, Massa N, Copetta A, Martinotti MG, Berta G. Impact of two fluorescent pseudomonads and an arbuscular mycorrhizal fungus on tomato plant growth, root architecture

and P acquisition. Mycorrhiza. 2004;**14**:185-192

[66] Frey-Klett P, Garbaye J, Tarkka M. The Mycorrhiza helper bacteria revisited. New Phytologist. 2007;**176**:22-36

[67] Poole EJ, Bending GD, Whipps JM, Read DJ. Bacteria associated with *Pinus sylvestris*-*Lactarius rufus* ectomycorrhizas and their effects on mycorrhiza formation in vitro. New Phytologist. 2001;**151**:743-751

[68] Duponnois R, Garbaye J. Mycorrhizal helper bacteria associated with the Douglas fir *Laccaria laccata* symbiosis: Effects in aseptic and in glasshouse conditions. Annales des Sciences Forestières. 1991;**48**:239-251

[69] Maier A, Riedlinger J, Fiedler HP, Hampp R. Actinomycetales bacteria from a spruce stand: Characterization and effects on growth of root symbiotic and plant parasitic soil fungi in dual culture. Mycological Progress. 2004;**3**(2):129-136

[70] Schrey SD, Schellhammer M, Ecke M, Hampp R, Tarkka MT. Mycorrhiza helper bacterium *Streptomyces* AcH 505 induces differential gene expression in the ectomycorrhizal fungus *Amanita muscaria*. New Phytologist. 2005;**168**:205-216

[71] Riedlinger J, Schrey SD, Tarkka MT, Hampp R, Kapur M, Fiedler HP. Auxofuran, a novel metabolite that stimulates the growth of fly agaric, is produced by the mycorrhiza helper bacterium *Streptomyces* strain AcH 505. Applied and Environmental Microbiology. 2006;**72**:3550-3557

[72] Deveau A, Palin B, Delaruelle C, Peter M, Kohler A, Pierrat JC, et al. The mycorrhiza helper *Pseudomonas fluorescens* BBc6R8 has a specific priming

effect on the growth, morphology and gene expression of the ectomycorrhizal fungus *Laccaria bicolor* S238N. New Phytologist. 2007;**175**:743-755

[73] Zhou AD, Wu XQ, Shen L, Xu XL, Huang L, Ye JR. Profiling of differentially expressed genes in ectomycorrhizal fungus *Pisolithus tinctorius* responding to mycorrhiza helper *Brevibacillus reuszeri* MPt17. Biologia. 2014;**69**(4):435-442

[74] Artursson V, Finlay RD, Jansson JK. Interactions between arbuscular mycorrhizal fungi and bacteria and their potential for stimulating plant growth. Environmental Microbiology. 2006;**8**:1-10

[75] Avis TJ, Gravel V, Autoun H, Tweddel RJ. Multifaceted beneficial effects of rhizosphere microorganisms on plant health and productivity. Soil Biology and Biochemistry. 2008;**40**:1733-1740

[76] Gravel V, Antoun H, Tweddell RJ. Growth stimulation and growth yield improvement of greenhouse tomato plants by inoculation with *Pseudomonas putida* and *Trichoderma atroviride*: Possible role of indole acetic acid (IAA). Soil Biology and Biochemistry. 2007;**39**:1968-1977

[77] Hayat R, Ali S, Amara U, Khalid R, Ahmed I. Soil beneficial bacteria and their role in plant growth promotion: A review. Annales de Microbiologie. 2010;**60**:579-598

[78] Malusa E, Sas-Paszt L, Ciesielska J. Technologies for beneficial microorganisms inocula used as biofertilizers. The Scientific World Journal. 2012;**2012**:12. DOI: 10.1100/2012/491206. Article ID: 491206

[79] Vestberg M, Kukkonen S, Saari K, Parikka P, Huttunen J, Tainio L, et al.

Microbial inoculation for improving the growth and health of micropropagated strawberry. *Applied Soil Ecology*. 2004;27:243-258

[80] Azcón-Aguilar C, Barea JM, Gianinazzi S, Gianinazzi-Pearson V, editors. *Mycorrhizas Functional Processes and Ecological Impact*. Berlin: Springer; 2009

[81] Barea JM, Azcón R, Azcón-Aguilar C. Interactions between mycorrhizal fungi and bacteria to improve plant nutrient cycling and soil structure. In: Varma A, Buscot F, editors. *Microorganisms in Soils: Roles in Genesis and Functions*, Vol. 3. Heidelberg: Springer Berlin; 2005. pp. 195-212

[82] Hata S, Kobae Y, Banba M. Interactions between plants and arbuscular mycorrhizal fungi. In: Kwang WJ, editor. *International Review of Cell and Molecular Biology*, Vol. 281. San Diego, USA: Academic Press; 2010. pp. 1-48

[83] Reddy CA, Saravanan RS. Polymicrobial multi-functional approach for enhancement of crop productivity. In: Sariaslani S, Gadd GM, editors. *Advances in Applied Microbiology*. Vol. 82. Michigan, USA: Elsevier; 2013. pp. 53-113

[84] Andre S, Galiana A, Le Roux C, Prin Y, Neyra M, Duponnois R. Ectomycorrhizal symbiosis enhanced the efficiency of inoculation with two *Bradyrhizobium* strains and *Acacia holosericea* growth. *Mycorrhiza*. 2005;15:357-364

[85] Saravanan RS, Natarajan K. Effect of *Pisolithus tinctorius* on the nodulation and nitrogen fixing potential of *Acacia nilotica* seedlings. *Kavaka*. 1996;24:41-49

[86] Saravanan RS, Natarajan K. Effect of ecto- and endomycorrhizal fungi along with *Bradyrhizobium sp.* on the growth and nitrogen fixation in

Acacia nilotica seedlings in the nursery. *Journal of Tropical Forest Science*. 2000;12:348-356

[87] Seneviratne G, Zavahir J, Bandara W, Weerasekara M. Fungal-bacterial biofilms: Their development for novel biotechnological applications. *World Journal of Microbiology and Biotechnology*. 2008;24:739-743

[88] Ude S, Arnold DL, Moon CD, Timms-Wilson T, Spiers AJ. Biofilm formation and cellulose expression among diverse environmental *Pseudomonas* isolates. *Environmental Microbiology*. 2006;8:1997-2011

[89] Garbaye J. Use of mycorrhizas in forestry. In: Strullu DG, editor. *Les mycorhizes des Arbres et Plantes Cultivées*. Paris, France: Lavoisier; 1990. pp. 197-248

[90] Duñabeitia M, Rodríguez N, Salcedo I, Sarrionandia E. Field mycorrhization and its influence on the establishment and development of the seedlings in a broadleaf plantation in the Basque country. *Forest Ecology and Management*. 2004;195:129-139

[91] Dahlberg A, Genney DR, Heilmann-Clausen J. Developing a comprehensive strategy for fungal conservation in Europe: Current status and future needs. *Fungal Ecology*. 2010;3(2):50-64

[92] McAfee BJ, Fortin JA. Competitive interactions of ectomycorrhizal mycobionts under field conditions. *Canadian Journal of Botany*. 1986;64:848-852

[93] Domínguez JA, Selva J, Rodríguez Barreal JA, de Omeñaca S. The influence of mycorrhization with *Tuber melanosporum* in the afforestation of a Mediterranean site with *Quercus ilex* and *Quercus faginea*. *Forest Ecology and Management*. 2006;231:226-233

- [94] Olivier JM, Savignac JC, Sourzat P. Truffe et Trufficulture. Perigueux, France: Ed Fanlac; 1996
- [95] Tibbett M, Cairney JWG. The cooler side of mycorrhizas: Their occurrence and functioning at low temperatures. Canadian Journal of Botany. 2007;**85**:51-62
- [96] Geml J, Timling I, Robinson CH, Lennon N, Nusbaum HC, Brochmann C, et al. Antarctic community of symbiotic fungi assembled by long-distance dispersers: Phylogenetic diversity of ectomycorrhizal basidiomycetes in Svalbard based on soil and sporocarp DNA. Journal of Biogeography. 2011;**39**:74-88
- [97] Guerrero-Galán C, Calvo-Polanco M, Zimmermann SD. Ectomycorrhizal symbiosis helps plants to challenge salt stress conditions. Mycorrhiza. 2019;**29**:291-301. DOI: 10.1007/s00572-019-00894-2
- [98] Colpaert JV, Wevers JHL, Krzmaric E, Adriaensen K. How metal-tolerant ecotypes of ectomycorrhizal fungi protect plants from heavy metal pollution. Annals of Forest Science. 2011;**68**:17-24
- [99] Azul AM, Sousa JP, Agerer R, Martín MP, Freitas H. Land use practices and ectomycorrhizal fungal communities from oak woodlands dominated by *Quercus suber* L. considering drought scenarios. Mycorrhiza. 2010;**20**:73-88
- [100] Blom JM, Vannini A, Vettraino AM, Hale MD, Godbold DL. Ectomycorrhizal community structure in a healthy and a *Phytophthora*-infected chestnut (*Castanea sativa* Mill.) stand in central Italy. Mycorrhiza. 2009;**20**:25-38. DOI: 10.1007/s00572-009-0256-z
- [101] Branzanti MB, Rocca E, Pisi A. Effect of ectomycorrhizal fungi on chestnut ink disease. Mycorrhiza. 1999;**9**:103-109. DOI: 10.1007/s005720050007
- [102] Corcobado T, Moreno G, Azul AM, Solla A. Seasonal variations of ectomycorrhizal communities in declining *Quercus ilex* forests: Interactions with topography, tree health status and *Phytophthora cinnamomi* infections. Forestry. 2015;**88**:257-266. DOI: 10.1093/forestry/cpu056
- [103] Lehto T, Zwiazek JJ. Ectomycorrhizas and water relations of trees: A review. Mycorrhiza. 2011;**21**(2):71-90. DOI: 10.1007/s00572-010-0348-9
- [104] Maurel C, Plassard C. Aquaporins: For more than water at the plant-fungus interface? New Phytologist. 2011;**190**(4):815-817. DOI: 10.1111/j.1469-8137.2011.03731.x
- [105] Nehls U, Dietz S. Fungal aquaporins: Cellular functions and ecophysiological perspectives. Fungal Aquaporins: Cellular functions and Ecophysiological perspectives. Applied Microbiology and Biotechnology. 2014;**98**(21):8835-8851. DOI: 10.1007/s00253-014-6049-0
- [106] Xu H, Kemppainen M, El Kayal W, Lee SH, Pardo AG, Cooke JEK, et al. Overexpression of *Laccaria bicolor* aquaporin JQ585595 alters root water transport properties in ectomycorrhizal white spruce (*Picea glauca*) seedlings. New Phytologist. 2015;**205**:757-770. DOI: 10.1111/nph.13098
- [107] Ishida TA, Nara K, Ma S, Takano T, Liu S. Ectomycorrhizal fungal community in alkaline-saline soil in northeastern China. Mycorrhiza. 2009;**19**(5):329-335. DOI: 10.1007/s00572-008-0219-9
- [108] Luo ZB, Li K, Gai Y, Gobel C, Wildhagen H, Jiang XN, et al. The ectomycorrhizal fungus (*Paxillus*

involutus) modulates leaf physiology of poplar towards improved salt tolerance. *Environmental and Experimental Botany*. 2011;72:304-311. DOI: 10.1016/j.envexpbot.2011.04.008

[109] Richard F, Roy M, Shahin O, Sthultz C, Duchemin M, Joffre R, et al. Ectomycorrhizal communities in a Mediterranean forest ecosystem dominated by *Quercus ilex*: Seasonal dynamics and response to drought in the surface organic horizon. *Annals of Forest Science*. 2011;68:57-68. DOI: 10.1007/s13595-010-0007-5

[110] Li J, Bao S, Zhang Y, Ma X, Mishra-Knyrim M, Sun J, et al. *Paxillus involutus* strains MAJ and NAU mediate K(+)/Na(+) homeostasis in ectomycorrhizal *Populus x canescens* under sodium chloride stress. *Plant Physiology*. 2012;159:1771-1786. DOI: 10.1104/pp.112.195370

[111] Luo ZB, Janz D, Jiang X, Göbel C, Wildhagen H, Tan Y, et al. Upgrading root physiology for stress tolerance by ectomycorrhizas: Insights from metabolite and transcriptional profiling into reprogramming for stress anticipation. *Plant Physiology*. 2009;151:1902-1917. DOI: 10.1104/pp.109.143735

[112] Szuba A. Ectomycorrhiza of *Populus*. *Forest Ecology and Management*. 2015;347:156-169. DOI: 10.1016/j.foreco.2015.03.012

[113] Beaudoin-Nadeau M, Gagné A, Bissonnette C, Bélanger P, Fortin J, Roy S, et al. Performance of ectomycorrhizal alders exposed to specific Canadian oil sands tailing stressors under in vivo bipartite symbiotic conditions. *Canadian Journal of Microbiology*. 2016;62(7):543-549. DOI: 10.1139/cjm-2015-0703

[114] Adriaensen K, van der Lelie D, Van Laere A, Vangronsveld J, Colpaert JV. A zinc-adapted fungus

protects pines from zinc stress. *New Phytologist*. 2004;161:549-555. DOI: 10.1046/j.1469-8137.2003.00941.x

[115] Adriaensen K, Vrålstad T, Noben JP, Vangronsveld J, Colpaert JV. Copper-adapted *Suillus luteus*, a symbiotic solution for pines colonizing Cu mine spoils. *Applied and Environmental Microbiology*. 2005;71:7279-7284. DOI: 10.1128/AEM.71.11.7279-7284.2005

[116] Jourand P, Ducouso M, Reid R, Majorel C, Richert C, Riss J, et al. Nickel-tolerant ectomycorrhizal *Pisolithus albus* ultramafic ecotype isolated from nickel mines in New Caledonia strongly enhance growth of the host plant *Eucalyptus globulus* at toxic nickel concentrations. *Tree Physiology*. 2010;30:1311-1319. DOI: 10.1093/treephys/tpq070

[117] Kayama M, Yamanaka T. Growth characteristics of ectomycorrhizal seedlings of *Quercus glauca*, *Quercus salicina*, and *Castanopsis cuspidata* planted on acidic soil. *Trees*. 2014;28:569-583. DOI: 10.1007/s00468-013-0973-y

[118] Luo ZB, Wua C, Zhang C, Lic H, Lipkad U, Polle A. The role of ectomycorrhizas in heavy metal stress tolerance of host plants. *Environmental and Experimental Botany*. 2014;108:47-62. DOI: 10.1016/j.envexpbot.2013.10.018

[119] Meharg AA, Cairney JWG. Ectomycorrhizas: Extending the capacities of rhizosphere remediation? *Soil Biology and Biochemistry*. 2000;32:1475-1484

[120] Krupa P, Kozdrój J. Accumulation of heavy metals by ectomycorrhizal fungi colonizing birch trees growing in an industrial desert soil. *World Journal of Microbiology and Biotechnology*. 2004;20(4):427-430. DOI: 10.1023/B:WIBI.0000033067.64061.f3

[121] Machuca A. Metal-chelating agents from ectomycorrhizal fungi and their biotechnological potential. In: Rai M, Varma A, editors. *Diversity and Biotechnology of Ectomycorrhizas*. Soil Biology. Berlin, Heidelberg: Springer-Verlag; 2011:25

[122] Daghino S, Martino E, Perotto S. Model systems to unravel the molecular mechanisms of heavy metal tolerance in the ericoid mycorrhizal symbiosis. *Mycorrhiza*. 2016;**26**:263-274. DOI: 10.1007/s00572-015-0675-y

[123] Krpata D, Fitz W, Peintner U, Langer I, Schweiger P. Bioconcentration of zinc and cadmium in ectomycorrhizal fungi and associated aspen trees as affected by level of pollution. *Environmental Pollution*. 2009;**157**:280-286. DOI: 10.1016/j.envpol.2008.06.038