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ORRHIZA NETH

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RESEARCH FINDING PAPER

Ectomycorrhizal Fungi: Identification, Multiplication, and Inoculum Production

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At least seven different types of mycorrhizal associations have been recognized, involving different groups of fungi and host plants and distinct morphology patterns. Two main types of mycorrhizae are recognized as the main types, depending on whether the fungus penetrates the root cells or not: ectomycorrhizas and endomycorrhizas. 1. Ectomycorrhizae (ECM); 2. Arbuscular mycorrhizae (AM) (=Vesicular-arbuscular mycorrhizas,VAM); 3. 'Ericoidmycorrhizae; 4. Arbutoid mycorrhizae; 5. Monotropoid mycorrhizae; 6. Orchid mycorrhizae; and 7. Ectoendomycorrhizas. Ectomycorrhizal symbiosis plays an important role in physiology, ecology, resistance, production, and other aspects of life of a single tree, population, and ecosystems of ectotrophic forest tree species, particularly in gymnosperms, woody trees, casuarinas, eucalyptus, and others.

Ectomycorrhizas

The diagnostic feature of ectomycorrhizas (EM) is the presence of hyphae between root cortical cells producing a net-like structure called the Hartig net, after Roben Hartig who is considered as the father of forest biology. Many EM also have a sheath or mantle, of fungal tissue that may completely cover the absorbing root surface externally (usually the fine feeder roots). The mantle can vary widely in thickness, colour, and texture depending upon the particular plant-fungus symbiotic combination. The mantle increases the surface area of absorbing roots and often affects fine-root morphology, resulting in root bifurcation and clustering. Continuous with the mantle are hyphal strands that extend into the soil. Often the hyphal strands aggregate to form rhizomorphs that may be visible to the unaided eye. The internal portion of rhizomorphs can be differentiated into tube-like structures specialized for long-distance transportation of nutrients and water.

In forest establishment and survival, the ectomycorrhizal fungi play an important role. The spore inoculum or the pure culture can be used for inoculation of nursery seedlings. The mycorrhizal seedlings on transfer will establish very well resulting in a good forest formation. Mycorrhizal fungi occupy the roots of plants and enhance the nutrient uptake, including containment of heavy metals, and promote the growth and survival of host plants under nutrient limiting conditions. They exhibit tolerance to high concentrations of heavy metals which normally cause severe toxicity symptoms in higher plants. Revegetation of such metal-contaminated habitats requires plantation of mycorrhizal tree species tolerant to heavy metals and abiotic stress. Mycorrhizal colonizations are often thought to increase stress tolerance of the host plant. The introduction of mycorrhizal fungi onto the sites of unfavourable saline soils improves soil fertility, early plant survival, and growth. The compartmentalized accumulation of salts observed in vacuoles and cell walls of ectomycorrhizal fungi and also in Frankia suggests that the organisms have the capability to compensate for shifts in osmotic potential. Experimental studies with Pinus, Casuarina, and Eucalyptus seedlings, inoculated with specific symbionts, have shown that knowledge of mycorrhizas

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can have practical significance in revegetation of disturbed soils. In the mycorrhizal symbiosis, the mycorrhizal fungi and the host plants are integral components. The beneficial role of mycorrhizal fungi in the survival and growth of plant species in the wastelands and adverse sites could be effectively utilized in the reclamation programmes. Mycorrhizal plants form an important biological resource and their introduction into mine-contaminated habitats and metal contaminated/polluted sites will be highly useful in phytoremediation, revegetation, and forestry.

Identification of ectomycorrhizal fungi

Ectomycorrhizal fungi can be identified by morphotaxonomic parameters, anatomical characteristics and molecular methods (PCR-ITS-RFLP & sequencing). During the study of morphological and anatomical characteristics, the census of fruiting bodies produced by different species are to be taken into account. The soil cores have to be separated and the identification has to be carried out based on the morphology of mycorrhizal roots and Hartig net. Fungal species involved in ectomycorrhizal association with forest trees can be recognized on repeated field observations of sporocarp and host associations. Ectomycorrhizas are mostly formed by fungi belonging to Basidiomycota and Ascomycota. Some of the common fungi forming ectomycorrhiza are Amanita muscaria, Laccaria bicolor, Laccaria laccata, Lycoperdon perlatum, Pisolithus arhizus (=P tinctorius), Rhizopogan luteolus, Russula parazurea, Scleroderma citrinum, Suillus brevlpes, Thelephora terrrestris, etc. Ectomycorrhizas are formed by woody plants, ranging from shrubs to trees but Massicotte et al. (1998) found ECM in Kobresia bellardii (Cyperaceae) and Polygonum viviparum (Polygonaceae) from alpine and arctic regions. The tree species belonging to the families Abeitaceae, Betulaceae, Casuarinaceae, Dipterocarpaceae, Fagaceae, Juglandaceae, Myritaceae, Pinaceae and Salicaceae are associated with ECM fungi. Around 5,000 ECM species interact with 2,000 plant species. Pinus species always require ECM for their survival. A very promising method based on fluorescence is used to distinguish early stages of mycorrhizal formation in plant roots (fungal mantle and hartig net). Fluorescence microscopy appears to be a very powerful tool for the visualization of individual hyphae in plant tissue as well as for the indication of root tip aging. The viability of ectomycorrhizal fungi was assessed with fluorescein diacetate and further numerous histochemical studies of roots have to be performed. The molecular techniques have also been used to know the EM fungal community establishments in soil, using restriction fragment length polymorphism and terminal restriction fragment length polymorphism

(Renske *et al.*, 2003). EM fungi are not a monophyletic group and EM-specific primers therefore do not exist. Primers generally used for identification of EM fungi have enhanced specificity for basidiomycetes and target the ITS regions of the rDNA gene cluster, including the 5.8S rDNA gene. The rDNA gene cluster is represented by one to several hundred copies per genome within the fungal nucleus and therefore provides a good target region. Until now, the basidiomycete-specific ITS1F-ITS4B primer pair has been used to amplify DNA extracted from fruit bodies, fungal cultures, infected plant material, and EM root tips.

Multiplication of ectomycorrhizal fungi

In laboratories, ectomycorrhizal fungi are separated into pure cultures. The culture contains only one type of organism and is suitable for the study of morphological, physiological, and biochemical characteristics. The technique employed for isolation of discrete colonies mainly requires that the number of organisms in the inoculum be reduced. During isolation, individual cells are plated apart on the surface of the agar medium to effect a separation of the different species present. Isolation of ectomycorrhizal (EM) fungal cultures have been critical in performing ectomycorrhizal research. The culture studies provide information about various parametric effects, such as the effect of pH, temperature, moisture, minerals, and various enzymes produced during growth and development of fungal symbionts. Fungal species involved in ectomycorrhizal association with forest trees can be recognized on repeated field observations of sporocarp cum host associations. A factor limiting the successful recovery of the fungal partner directly from mycorrhizas in all these studies is the high incidence of fast growing fungi which emerge despite extensive serial washing and surface sterilization of mycorrhizas prior to plating. Most species of Suillus and Rhizopogon are easily isolated and grown luxuriously in culture. Few species have been successfully isolated in some EcM fungal genera, such as for instance Russula, Gomphidius, etc. Many other species can be isolated from sporocarps, such as Alpova, Amanita, Astraeus, Boletus, Cortinarius, Fuscoboletinus, Hebeloma, Hymenogaster, Hysterangium, Laccaria, Lactarius, Leccinum, Melanogaster, Paxillus, Pisolithus, Rhizopogon, Scleroderma, Suillus, and Tricholoma.

Selection of efficient ectomycorrhizal isolates

Isolates of ectomycorrhizal fungi are generally selected on the basis of their compatibility and efficiency where compatibility means the ability to colonize host plant roots while efficiency means the ability to promote growth of the host plant. The efficiency is usually tested by evaluating parameters, such as the height of the inoculated plant, the diameter of the stem, the overall dry mass of the plant, and the nutrient content of the plant, especially phosphorus and nitrogen. Another important factor that is to be considered, but which at the moment is seldom taken into account in the selection of mycorrhizal isolates, is the growth rate of the fungus when cultivated in large-scale bioreactors. Compared to free-living microorganisms, ectomycorrhizal fungi grow very slowly in laboratory conditions and this slowness of growth may cause operational problems in bioreactors.

Inoculum production

Three main types of ectomycorrhizal inoculants have been used in nurseries during the last decades: Natural inoculum (soil, humus, ectomycorrhizae), basidiospore inoculum, and vegetative (mycelial) inoculum.

Various organic matter, such as soil, litter, variable forms of humus, rotten wood, and ectomycorrhizae, collected from forest plantations or mature stands were exploited, as inocula in an effort to inoculate forest tree seedlings. A major drawback of natural inoculum is that the specificity of ECM fungi in the inoculum cannot be established. This inoculum may also contain harmful microorganisms and weeds in addition to the ECM fungi. Natural inoculum is usually collected from stand or underneath regions of individual tree of species which is already inoculated. Ectomycorrhizae excised from root systems of trees were also used as inoculum on a limited basis in research trials. A great deal of time and care is required to obtain a sufficient quantity of viable ectomycorrhizae. In the earliest studies, a thin layer of soil obtained from natural forests, nurseries, or established plantations was spread on the top of nursery bed and mixed with the soil or planting substrate. This method is still used in many parts of the world, particularly in developing countries. One problem with this type of method is that a large amount of soil is required to inoculate nursery plants, but an even more important problem is the risk of introducing plant pathogens and weeds. Moreover, there is no precise information about the fungal species that are being introduced and their infection potential. Despite these disadvantages, soil inoculant is recommended if no other type of inoculant is available.

Fungal spores, obtained from fruiting bodies harvested in natural forests, old nurseries or established plantations, have also been used in many parts of the world. They are easy to obtain and apply to plants. They can be added as pellets, mixed with the planting substrate, applied directly to seeds or applied as a water suspension through the nursery watering system. Basidiospores can be obtained and stored before application either in dry state or blended with water. Basidiospores can be obtained by crushing and sieving basidiocarps through a mesh screen in a closed plastic bag. Using these techniques, well over 1 kg (fresh weight) of basidiospores of *Pisolithus tinctorius* can be collected in less than 3 h. The dry spores should be stored in either small plastic bags or amber glass bottles in darkness at 5°C for 10 days before use, mixed with vermiculite for seedling inoculation or stored at 4°C for 1–18 months before blended with distilled deionized water (1:10, v:v) for 5s on low speed.

Spore suspensions can be prepared by blending sporocarps with distilled or tap water using a blender at low or high speed until the spores were released. After collection, sporocarps have to be dried at 30 °C-40 °C for 48 hrs before blending and sieving, respectively. Required spore concentrations are prepared by serial dilution of spore suspension with water. Spore concentrations are counted using haemocytometer. The approximate number of spores contained per gram of dried sporocarp tissue are usually 107–1010 (or 1.1 million basidiospores) per milligram. The advantages of using spores for inoculation are that spores require no extended growth phase under aseptic conditions, such as vegetative inoculum, spore inoculum is very light, and spores are able to survive under storage from one season to the next. The major disadvantages are the lack of standard laboratory tests to determine spore viability, insufficiency of sporocarps inoculum in any given time of the year, delay in ectomycorrhizal formation as compared with vegetative inoculum, and lack of genetic definition. This type of inoculant is limited to those fungal species that are able to produce large number of spores and fruiting bodies and there is no certainty of their compatibility and efficiency towards the host plant species that has to be inoculated. As spores are generally collected from multiple fruiting bodies, they tend to present a higher genetic variability than vegetative inoculant. The availability of spores is erratic during the year, hence there is a need to collect and store large numbers of fruiting bodies when they are abundant in nature. According to Marx et al. (1979), root colonization by this type of inoculant is slower than that of vegetative inoculant of the same fungal isolate.

Addition of mycelia obtained from pure cultures of ectomycorrhizal fungi, also called vegetative inoculant, has proven to be the most suitable method. Vegetative inoculants can be prepared from any fungus that is cultivated in pure culture, which allows the use of selected isolates that have been previously tested in terms of their efficiency in promoting plant growth. Pure cultures are generally obtained from fruiting bodies or from mycorrhizal roots and may be maintained indefinitely under laboratory conditions. For commercial production of vegetative inoculant, mycelium has to be grown in solid substrate or in liquid culture medium at several different scales. Vegetative (mycelial) inoculum may be prepared as mycelial suspension (slurry), vermiculite–peat (substrate carrier) inoculum, and alginate-bead inoculum.

Mycelial suspension (slurry) is more often used in a small-scale synthesis of mycorrhizas in controlled conditions than in nursery experiments. Mycelial slurry of Pisolithus tinctorius, Suillus granulatus, and Rhizopogon luteolus can be prepared by blending mycelial mats from liquid cultures with distilled water at high speed for less than 3 s. One litre of sterile water contained 40 g of (fresh weight) mycelium. Mycelial suspension can be prepared in a fermentor. Starter content of living propagules was diluted five times with water and mixed with the substrate in a cement mixer to get a final content of approximately 104 living propagules or 5.68 mg of dried mycelium per seedling. For production of inoculum, mycelium of Suillus plorans was first grown in liquid culture and then in sterile peat moss. Moser and other workers tested various organic materials as the inoculum substrate. However, forest litter, sawdust, grain of cereals, corn, and bark, were not as effective as peat moss.

Vermiculite-Peat (substrate carrier) inoculum is another type of pure culture inoculum. Marx and Bryan (1975) prepared the inoculum in 2-L jars containing the mixture of 1,400 mL of vermiculite, 50 mL of finely divided peat moss, and 750 mL of liquid MMN medium with glucose. The containers were autoclaved for 30 min and each was inoculated with eight mycelium-agar disks of *Pisolithus tinctorius*. After 15 week incubation at room temperature, the vermiculite particles were permeated with mycelium. To prepare mass inoculum for infestation of soil, mycelium was removed from the jars, passed through a 5-mm mesh screen, and held with two layers of cheesecloth while being leached with cool running tap water to remove non-assimilated nutrients. Novel formulations of vermiculite-peat inoculum did not require leaching and drying before use. An important modification of original inoculum formulation was carried out on vermiculite: peat. Marx et al. (1982) reported that vermiculite-peat inoculum mixture produced by solid-substrate fermentation contained

5%–10% peat moss by volume.

Le Tacon et al. (1983) and Mauperin et al. (1987) have shown that mycelium grown in a liquid medium and entrapped in calcium alginate gel is a very efficient inoculum for ECM development and can be used as an alternative to the classical vermiculite-peat mixture. Mycelium in alginate-bead inoculum is better protected, survives longer, and has a long-lasting effect than vermiculite-peat mixture. For production of alginate-bead inoculum, fungul cultures are grown in liquid medium. The mycelial pellets are washed in tap water, homogenized in a blender for 5-10 s, and resuspended in distilled water containing 10 g.l⁻¹ of sodium alginate and 30 g.l⁻¹ of powdered Sphagnum peat. This suspension is pumped through a pipe with 5-mm holes above a 100 g.l⁻¹ CaCl₂ solution, each drop forming a bead of reticulated calcium alginate gel with 3-4 mm in diameter. The beads are cured in CaCl_a for 24 h at room temperature (for ensuring complete reticulation of the gel), washed in tap water to remove NaCl, stored in airtight containers at room temperature to prevent drying, and used in the nursery.

Inoculant efficiency

The evaluation of the efficiency of an ectomycorrhizal isolate in promoting plant nutrition and growth under controlled conditions in a greenhouse is an important first step in selecting a strain for largescale production of host-plant inoculants. However, to justify the investment in the establishment of a large-scale inoculant production plant, it is also necessary to demonstrate the efficiency of the inoculant in promoting plant growth under nursery and field conditions. In the nursery, these studies can be performed up to 6 months for plants such as *Eucalyptus* spp. and *Pinus* spp. The type of ectomycorrhizal material used for inoculation can affect the success of mycorrhizal inoculation programme. The inoculum must remain viable during storage and transport, maintaining its infectivity for several months after its production. Furthermore, the formulated inoculant must be easy to apply and must also be free of contamination by plant pathogens and any free-living microorganisms that could affect inoculant viability. Finally, the cost of the inoculant must be compatible with the financial resources available to the nursery care must be taken to avoid raising seedling prices to uncompetitive levels.

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MorphoTaxonomy of *Glomus desereticola/Septoglomus deserticola* (Accession CMCC/AM-2901)

Varsha and Alok Adholeya*

Most plants in their natural habitat work under the influence of a special group of soil fungi known as arbuscular mycorrhiza fungi (AMF). AMF occurs on a wide spectrum of temperate and tropical plant species and are absent in less than 30 plant families. The fungi frequently increase the supply of plants with nutrients and decrease their sensitivity to different abiotic and biotic stresses. The identification of AMF is done by morphological, molecular, and biochemical analysis.

Continuing the series of morphotaxonomic studies of our successfully isolated and established AMF species, this edition will focus on the wellknown genus that has been successfully maintained in Centre for Mycorrhizal Culture Collection (CMCC) of TERI with the accession number CMCC/AM-2901.

Monosporal Establishment

This culture of accession CMCC/AM-2901 was isolated and raised from the soil of Palwal, Haryana (India). For proliferation of indigenous mycorrhiza, trap cultures were raised in pot conditions with host plant Sorghum bicolour for a period of three months. After an adequate growth cycle, spores propagated in trap culture were checked by wet sieving and decanting method (Gerdeman and Nicholson 1963). First of all the initially weighed soil was suspended in distilled water and was allowed to pass through series of sieves of 60,100, and 300 British Standard Size (BSS) and then were analysed under stereo microscope for AMF diversity and density. Morphologically similar and mature healthy spores were chosen to raise pure culture. Morphological properties of spores and their wall structure were determined by preparing voucher specimen of spores mounted in Polyvinyl lacto glycerol (PVLG) and a mixture of PVLG and Melzer's reagent (1:1, v/v). After the growth period of three to six months,

host roots were examined for root colonization for successful cultures (Figure 1).

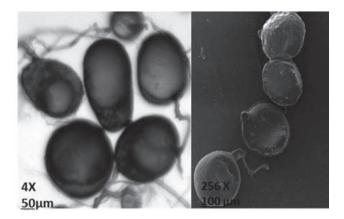


Figure 1 Compound microscopic images (4 X) and SEM (Scanning electron micrographs) of CMCC/AM-2901 showing spore with a subtending hyphae

Spore Morphology and Shape

Spores isolated from successful monosporals are found to arise in soil singly or in loose clusters lacking a peridium. The colour of spores varies from deep yellow to light brown. The shape of spore varied from globose to subglobose, $(90-)120(-160) \mu m$ diam., (Figure 2) sometimes ovoid to pear-shaped, with one subtending hypha which is flared, funnel-shaped, and rarely constricted at the spore base. Scanning Electronic Micrograph (SEM) of the spores showed that the outer surface of the spores is flared with one subtending hypha (Figure 3).

Subcellular Structure of the Spore

These spores showed very less reaction with polyvinyl lacto glycerol: Melzer's reagent but no reaction was seen with polyvinyl lacto glycerol (PVLG). The subcellular structure and wall layer are visible in Figure 4.

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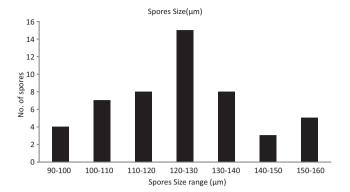


Figure 2 Analysis of spore diameter of 50 healthy and cleaned spores obtained from one-year old monosporal culture of accession CMCC/AM-2901

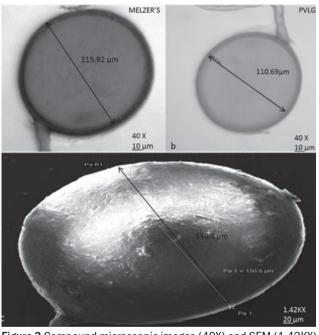


Figure 3 Compound microscopic images (40X) and SEM (1.42KX) of mature spores of accession CMCC/AM-2901 showing globose to subglobose shaped spores with a single subtending hyphae. SEM images reveal rough outer layer of the spore

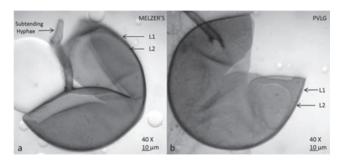


Figure 4 Compound microscopic images of spore wall layers of CMCC/AM-2901 after mounting in PVLG: Melzer's reagent(A) and PVLG reagent(B)

Spore Wall Layer 1 (L1)

This layer forms the spore surface. Layer 1 is evanescent and hyaline. This layer is $0.5-2.3 \mu m$ thick. It is observed frequently that this layer is completely sloughed in mature spores.

• Spore Wall Layer 2 (L2)

This layer is formed when the formation of L1 is completed. Layer 2 is laminate and smooth. This layer is of deep yellow to light brown colour. The thickness of this layer is $1.8-3.8 \mu m$.

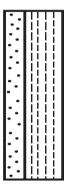


Figure 5 Murograph (Walker 1983) of the mature spore showing two distinct wall layers

Subtending Hyphae

These spores show hypha intact, straight or curved, flared, funnel-shaped, rarely constricted at the spore base. The width of the subtending hyphae ranges $5.0-10.0 \,\mu\text{m}$ at the spore base. Wall of subtending hypha is composed of two layers continuous with spore wall L1 and L2. Width of L1 is $0.8-1.3 \,\mu\text{m}$ thick and L2 (2.5-3.8) μm thick (Figures 5 and 6).

Mycorrhiza

From the monosporal culture of accession CMCC/ AM-2901, the host roots are harvested and stained in ink vinegar. Mycorrhiza structures such as arbuscules, extra and intraradical hypha, are observed. Root colonization assays showed the presence of both extraradical and intraradical hyphae. However, the extraradical hyphae are more abundant in specimens observed in pot cultures. Arbuscules are abundant and are dispersed evenly through the root cortex. Intraradical vesicles are present (Figure 7).

Conclusion and Classification Level

On the basis of this morphotaxonomic analysis of the accession CMCC/AM-2901, many distinguishing features regarding the family, genera, and the species could be derived. The following features were taken into consideration for characterization and identification:

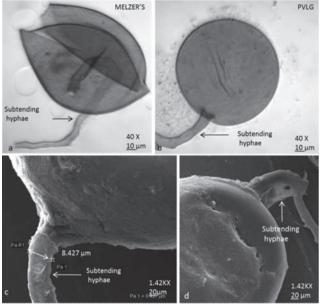


Figure 6 Compound microscopic images of spore of CMCC/AM-2901 showing cylindrical subtending hyphae after mounting it with PVLG: Melzer's reagent (a and b) SEM images of subtending hyphae (c and d).

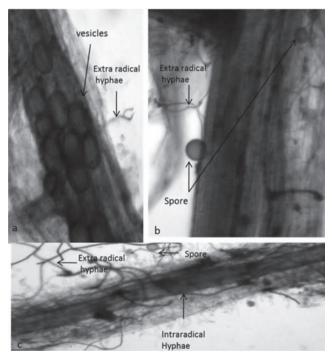


Figure 7 Compound microscopic images of roots of *Sorghum bicolor* stained in ink vinegar and observed for root colonization by CMCC-AM-2901 showing extraradical hypha (a and b) and a) and abundant vesicles in the cortical cells (a). Spores in the extraradical hyphae are also seen (c).

 Globose, asexual spores produced singly with two layered spore walls.

- Spore wall layer is composed of outer hyaline layer followed by one laminate and smooth layer continuous with the subtending hyphal wall.
- Spores are of varying shapes and sizes ranging from globose to sub- subglobose, sometimes ovoid to pear-shaped.
- Formation of both intraradical and extraradical hyphae and abundant vesicles and intracellular arbuscules.

All these features suggest that the culture CMCC/AM-2901 belongs to the family Glomeracea (Walker and Schüßler 2004).

Some of the unique morphotaxonomic features of the accession are enumerated as follows:

- Light yellow to dark brown, globose, asexual spores produced singly with layered spore walls; spores are of varying shapes and sizes ranging from globose to sub-globose. Size ranges 90 – (120) – 160 µm.
- Spore wall layer is composed of outer mucilaginous layer and inner smooth laminate layer.
- Presence of intact, flared, funnel-shaped, and rarely constricted subtending hyphae. The width of the subtending hyphae at the point of attachment at the spore base ranges 8.1–13.3 µm.
- Formation of both intraradical and extraradical hyphae and abundant vesicles and intracellular arbuscules.

The taxonomic feature of the accession CMCC/AM-2901 matches the characters of *Glomus deserticola* (Walker 1983).

Systematic Classification

Glomeromycotina Glomeromycetes Glomerales Glomeraceae Septoglomus Septoglomus Deserticola

Most of the work on AMF focusses on phylogeny, ecology, genetics, taxonomy, and functional properties. Identification of morphological features of an organism has been used as a conventional method for identifying an organism and comparing it with existing reference or type species. Morphological features about AMF spore morphology, wall layers, hyphal attachment, reaction with reagents, etc., help to reveal information about genera and species to which the AMF belongs. However, sometimes this approach can create a problem in identification of the organism at species level. Therefore, using alternative methods have more significant advantages over traditional taxonomic approaches. Characterization techniques, such as biochemical characterization through Fatty acid methyl ester (FAME) profiles and sequencing of ITS region of 18s rDNA (molecular characterization) has become more popular. Analyses of rDNA regions have often confirmed the morphologically defined species. It is therefore advised to our distinguished readers to kindly correlate their morphotaxonomic studies with molecular phylogenetic results.

Acknowledgements

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Role of Mycorrizha in Improving Product Quality

C Manoharachary^a and Anurag Nath^{b*}

The beneficial effects of mycorrhiza are explored and well known from earlier times. There are multiple influential parameters offered by mycorrhiza. One important aspect in this perspective is the improved quality of the products from these plants which are in association with mycorrhiza. Potentially, it has been reported that the majority of the vegetable crops act as a potential host for the mycorrhiza. Their increased surface area and expanded hyphae result in an improved nutrient and water supply to the plants such that they are able to sustain in an adverse environment. Moreover, mycorrhizae have been established to induce and increase the tolerance of the plants towards environmental stress and pathogens. This increases the resistance of the host plant towards the root diseases and pathogens such as the nematodes which attack the

vegetable crops. These pathogens act as a prominent source of pathogen affecting the growth of the plant subsequently inhibiting the growth of these plants and affecting the vegetable yield.

There have been multiple research studies which focus on the beneficial aspects of mycorrhiza towards quality improvement of the products from host plants. The inoculation of the crops with mycorrhiza counter parts could act as a promising solution for the stressful condition faced by the plants especially in temperate conditions. The incorporation of mycorrhiza in the system of vegetable and other economical crops can essentially improve the profit and commercial value of the crops and simultaneously assist in the economic benefit of country.

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Mycorrhiza as a Boon for Plant Growth

C Manoharachary^a and Anurag Nath^{b*}

Most of the terrestrial plants exist in symbiotic associations with fungi which occupy a prominent space in their roots from where they tend to expand themselves supplementing the plant with the required nutrients. Both ecto- and endo mycorrhizae are beneficial for the plants however they differ in their structure and physiological relationship with their host specific organism. Studies have been conducted on the different aspects of symbiotic association amongst the plants and mycorrhizae from earlier times. They have been reported to reduce the root biomass and simultaneously stimulate the plants to increase the uptake of nutrients. They have also been found to be proliferating in the soil pores which tend to be small for even the root hairs to enter. The arbuscular mycorrhiza have been found to show fundamental impacts and maintain a balance in the system of the plants. They enhance the eco-physiology and soil biota when being associated with the host plant. Mycorrhizae have been found to cause a profound beneficial effect in metal contaminated soils as well.

The mycorrhizae act as a source of connection for the flow of energy and nutrients between the soil and the host plants. The symbiosis amongst the mycorrhiza and the plant has been explored for the purpose of devising an efficient means of supporting the plant growth. The mycelial networks of the fungus have been reported to establish a strong connection with the plant system and often these form the largest portion of the biomass found in soil. Their relationship has a profound effect on the ecosystem and the surrounding biotic communities. A study based on the fossil discoveries and the DNA sequences concluded that arbuscular mycorrhizae have been existing on Earth since 400 million years. For example *Rhynia* chert plants. The surface area of the plant roots increases when in association with mycorrhiza so that the host plants can withstand even in extreme and harsh climatic conditions.

Mycorrhizae have the ability to alleviate the capability of the plants to withstand the different anthropogenic stresses including metal contaminated soils and other toxic pollutants. They increase the tolerance level of the host plants to withstand the soil pollutants and simultaneously remediate the soils such that the growth of the plants can be enhanced efficiently.

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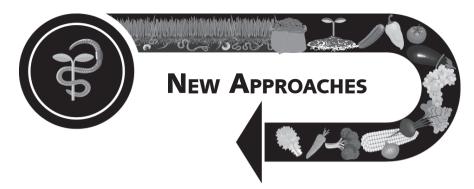
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New Approaches and Techniques

1. Omics for understanding the increased yield and resistance of plants due to mycorrhiza growth

Increased tolerance of plants in the presence of mycorrhiza has been reported multiple times. Often biomass production is increased by the presence of mycorrhizal association with the plants. Understanding the underlying concepts which support this cause is essential for utilizing the same to enhance the tolerance of plants towards biotic and abiotic stresses. Omics is an emerging field which can be used for the identification of such principles such that they can be applied for the increased product yield from crops.

2. Proteomic approach can be employed for identification of symbiotic association of mycorrhiza

Applying proteomics to focus on the fungal endophytes that protect these plants by preventing nutrient deficiency is an essential field which can enhance the ability of the plants towards environmental stresses. Protein characterization based on the qualitative and quantitative aspects of the plant yield can enable better understanding about the signalling pathways and various other principles that can be applied for enhancement of productivity and simultaneously increasing the tolerance of the plants towards the stressful conditions of soil and also increase their efficiency in nutrient uptake.

3. Stimulation of root hair by mycorrhiza during drought

Audio has been found to be associated with the increased root hair development. Further, mycorrhiza have been reported to be increasing the auxin production within the plants such that their root hairs develop thus increasing the surface area by enhancing the nutrient uptake and flow/transport of water across the plant surface. Mycorrhizal association offers multiple benefits for the betterment of plants and increasing their production/productivity.

4. Species-specific inoculation for plant growth

Certain mycorrhiza species have been reported to be associated with some specific plants. The growth of the corresponding host plants has been found to be enhanced with higher product yield. Inoculating species of plants with the specific mycorrhiza species can help in the growth and development of plants by providing better nutrition.

ANNOUNCEMENTS

- 1. In 2016–17, TERI set up the world's biggest facility for mycorrhiza production in Gual Pahari in Gurugram, Haryana.
- 2. Submission to *Mycorrhiza News* in the form of relevant notes; brief write-ups highlighting current research achievements; news/events of common interest to members like seminars/workshops attended, in the field of mycorrhiza are always welcome! Members are requested to provide the MIC (Mycorrhiza Information Center) with copies of articles,papers,reports,reviews,etc., dealing with mycorrhiza for the proper dissemination of mycorrhizal information amongst researchers.
- 3. The Mycorrhiza website www.mycorrhizae.org.in is currently undergoing transformation and will soon be revamped into a better and much more informed website with new added features for better information dissemination.

RECENT REFERENCES

The latest additions to the network's database on mycorrhiza are published here for the members' information. The list consists of papers from the following journals:

- Agricultural Water Management
- Applied Soil Ecology
- Biocatalysis and Agricultural Biotechnology
- Biochemical Systematics and Ecology
- Ecological Engineering
- Ecotoxicology and Environmental Safety
- Environmental and Experimental Botany
- European Journal of Soil Biology
- Fungal Biology
- Fungal Ecology
- Geoderma

- Journal of Arid Environments
- Journal of Environmental Sciences
- Journal of Integrative Agriculture
- Journal of Plant Physiology
- Mycoscience
- *Rhizosphere*
- Science of The Total Environment
- Scientia Horticulturae
- Soil Biology and Biochemistry
- South African Journal of Botany

Copies of papers published by mycorrhizologists during this quarter may please be sent to: **Mr Anurag Nath** (anurag.nath@teri.res.in) for inclusion in the next issue.

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19–24 August 2018	Tel.: + 353 86 860 9818 Email: iapbhome@gmail.com Website: http://iapb2018.com/					
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10–12 September 2018	<i>Email:</i> agri@agriconferences.com <i>Website:</i> https://agricultureconference.wordpress.com/					
London, England 6–7 October 2018	UK Fungus Day, British Mycological Society UK Fungus Day, c/o British Mycological Society, Charles Darwin House,12 Roger St, London WC1N 2JU					
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