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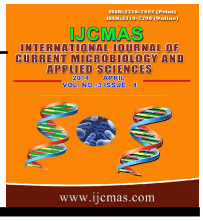

Review Article

Arbuscular Mycorrhizal Fungi (AMF) as a Biofertilizer- a Review

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A B S T R A C T

Biofertilizers are the organisms (Bacteria, fungi, cyanobacteria, etc.) that enrich the nutrient quality of soil. Plants have a number of beneficial relationships with such organisms. Among these AM-Fungi are ubiquitous and form a mutuality relationship with roots of most plant species. Generally, the distribution of AM spores in rhizosphere soil is governed by edaphic and certain climatic factors. Soil-based pot culture is applied as a common method for production of AM –Fungal inoculum. The importance of these fungi to agricultural and forestry resides in their role in plant growth and nutrition. Dual inoculation of such fungi with a *Rhizobium* and other bacterium on plant enhanced the growth and other beneficial effects viz., resistance to disease and tolerance to adverse soil and climatic conditions. This article reviews the information on Arbuscular mycorrhizae in different fields.

Introduction

Bio-fertilizers are the organic substances which make use of microorganisms to increase the fertility of soil. These fertilizers are not harmful to crops or other plants like the chemical fertilizers. They are actually taken from the animal wastes along with the microbial mixtures. Microorganisms are used to increase the level of nutrients in the plants. They let the plants grow in a healthy environment. They are also environment friendly and do not cause the pollution of any sort. Use of bio fertilizers in the soil, makes the plants healthy as well as protect them from getting any diseases.

The main sources of bio- fertilizers are bacteria, fungi, cyanobacteria, etc. Such bio-fertilizers are cultured and are used for inoculating seed or soil or both under ideal conditions to increase the availability of plant nutrients. Among this mycorrhiza is an important one in agriculture field for the cultivation of many crops.

AM-Fungi

A mycorrhiza is a symbiotic association between a fungus and the roots of a vascular plant. In this association, the fungus colonizes the host plant's roots,

either intracellular as in arbuscular mycorrhizal fungi or extracellularly as in ecto mycorrhizal fungi. They are an important component of soil life and soil chemistry.

In the symbiotic associations of plant and fungi, arbuscular mycorrhiza, which is formed between plants and Glomeromycota fungi, has the widest distribution in the nature. AM fungi inhabit a variety of ecosystems including agricultural lands, forests, grasslands and many stressed environments, and colonize the roots of most plants, including bryophyte, pteridophyte, gymnosperms and angiosperms.

Mycorrhiza are commonly divided into ecto mycorrhiza (the hypha of fungi do not penetrate individual cells with in the root) and endomycorrhiza (the hypha of fungi penetrate the cell wall and invaginate the cell membrane). Endomycorrhiza are variable and are further classified as arbuscular, ericoid, arbutoid, monotropoid and orchid mycorhizae. Arbuscular mycorrhizal (AM) fungi are ubiquitous in soil habitats and form beneficial symbiosis with the roots of angiosperms and other plants (Gerdemann,1968).This AM fungi belong to the family Endogonaceae, of the order Muccorales, of the class Zygomycetes (Gerdemann and Trappe,1974).The AM forming genera of the family includes *Acaulospora, Entrophospora, Gigaspora, Glomus, Sclerocystis* and *Scutellospora*.

A wide range of AM Fungi distribution is found in India. Bakshi (1974) was the first to give an account of 14 spore types: *Glomus macrocarpum Tul and Tul var., G.geosporum, G.mosseae, Glomus sp., Sclerocystis coremioides, Sclerocystis sp.,Gigaspora, Calospora, Acaulospora sp., Endogone gigantea, E. microcarpum, Endogone 1, Endogone 2, Endogone 3*.Gerdemann and Bakshi (1976) reported two new species viz., *Glomus multicauli* and *Sclerocystis sinuosa*.

In general, the distribution of AM spores in rhizosphere soil is governed by edaphic and certain climatic factors. According to Khaliel (1988), pH is the only edaphic factor which determines the abundance of AM fungi. However, pH did not influence the mycorrhizal spore density and frequency (Bergan and Koske, 1981).High soil P and N content caused a reduction in infection and number of AM spores (Mosse, 1981; Azcon-Aquilar and Barea, 1982) as well as decreasing the dependency of the plant on the fungal association (Ojala *et al*., 1983).

The importance of AM fungi to agricultural and forest plant species resides in its role in plant growth and nutrition. In tropical forests incidence of mycorrhizae profoundly influence soil fertility and thus the growth and development of plants (Bagyaraj, 1989).The understanding of mycorrhizal associations in tropical forest tree species is necessary for proper exploitation of these habitats.

The bulk of known AMF species belongs to the family Glomaceae (Pirozysnki and Dalpe, 1989) which includes the genera *Glomus* and Sclerocystis. Arbuscular mycorrhizal (AM) fungi are associated with about 80% of the plant families in the world (Giovannetti and Sbrana, 1998).

AMF have been separated from the Zygomycota and placed in a new monophyletic phylum, Glomeromycota composed of four orders with seven families and ten different genera (Schubler *et al.,* 2001). There are more than 180

species of AMF (Morton and Redecker, 2002). Morton and Redecker (2002) discovered two ancestral classes of AM fungal species from deeply divergent ribosomal DNA sequences and is classified into two new families Archaeosporaceae and Paraglomaceae.

Zhang *et al.,* (2004) surveyed 44 taxa of AM fungi of AM fungi (total species richness) in the deforested and natural forest in sub tropical region of Dujiangyan. They also reported that *Acaulospora* and *Glomus* were the dominant genera in the study sites.

AM fungi-root colonization

Bisleski (1973) reported that AM fungi may increase the effectiveness of absorbing capability of surface host root as much as ten times. Ions such as P, Zn and Cu do not diffuse readily through soil. Because of this poor diffusion, roots deplete these immobile soil nutrients from the zone immediately surrounding the root. The increase in plant growth resulting from AM symbiosis is usually associated with increased nutrient uptake by the hyphae from the soil (Harley and Smith, 1983). It is widely accepted that a hyphal network associated with the roots of a living plant is capable of infecting the roots of other plants growing in its vicinity (Chiariello et al., 1983; Franschis and Read, 1984; Newman, 1988).

Mycorrhizal fungal hyphae extend into the soil, penetrating into nutrient depletion zone and increased the effectiveness of immobile elements by as much as sixty times. Direct benefits are usually related to the enhancement of phosphate uptake by the plant, however, in some soils Zinc, Copper and ammonium are also important (Stribley, 1987). Indirect benefits may include increased soil aggregation or stabilization of soil associated with hyphae formed in the soil.

Mycorrhizae are known to increase the root growth of infected plants (Graham *et al.*, 1987) and are also known to increase (Huang et al., 1988) or cause no change in xylem pressure. Species of AM fungi that can either directly or indirectly increase plant growth by improving soil conditions (Kapoor and Mukerji, 1990).

Root colonization by VAM fungi can provide protection from parasitic fungi and nematodes (Duchesne *et al.,* 1989, Grandmaison *et al.,* 1993, Newsham *et al.,* 1995, Little and Maun 1996, Cordier *et al.,* 1998, Morin *et al.,* 1999). Mycorrhizal benefits can include greater yield, nutrient accumulation and reproductive success (Lewis and Koide, 1990, Stanley *et al.,* 1993).Network of hyphae supported by dominant trees may help seedlings become established or contribute to the growth of shaded under storey plants (Hogberg *et al.,* 1999, Horton *et al.,* 1999).

The main advantage of mycorrhiza is its greater soil exploration and increasing uptake of N, P, K, Zn, Cu, S, Fe, Ca, Mg and Mn supply to the host roots (Li *et al.,* 1991; Champawat and Pathak, 1993; Marschner and Dell, 1994; Smith et al., 1994; Abdul Malik, 2000). The *Glomus etunicatum* inoculated maize plants in sandy loam soil, under water stressed conditions, absorbed more phosphorus than non-mycorrhizal plants (Muller and Hofwer, 1991). Michelsen and Rosendahl(1990) and Osonubi *et al.,* (1992) observed that the AM fungi contribution to drought tolerance is minimal in *Acacia nilotica*.

The fact that colonized plants are better able to obtain their nourishment from the soil and resist environmental stresses gives

fungal symbionts a biofertilizing and crop protection role. In agriculture by the increased uptake of soil minerals by colonized plants, it is possible to consider substantially reducing applications of fertilizers and pesticides and at the same time obtain equivalent or even higher crop yields (Abbott and Robson, 1991a).Through appropriate management of mycorrhizae in agriculture, it is also possible to maintain soil quality and sustainability thereby protecting the environment over long term and also reducing cost of production.

The advantages and benefits of adopting mycorrhizae in agriculture, allows us to better visualize the scope of this phenomenon at the crop level and , intern, the impact of its long- term adoption on the quality of life in improvement in nutrition, tolerance to water stress, resistant to low temperature, transformation of root architecture, diversity of microbes in soil development of resistance against pathogens increased synthesis of primary or secondary metabolites and improvement in the quality and quantity of agricultural products(Abbott and Robson, 1991b).

Mycorrhizal fungi contribute to carbon storage in soil by altering the quality and quantity of soil organic matter (Ryglewicz and Andersen, 1994). AM fungi alter the kinetic properties of the root, thereby enhancing its nutrient uptake abilities. Hence it is clear that mycorrhizal fungi play a vital role in nutrient cycling and productivity of crops (Smith and Read, 1997).

AM fungi are known to colonize a number of tropical plants including vegetables. AM association is known to help in the growth of various crops including horticultural plants like carrot or tomato (Sassal, 1991).The occurrence of AMF in a natural forest was recorded in the old Delhi ridge, Saraswathi Range of Haryana (Thapar and Uniyal, 1996), forest soils of Andhra Pradesh (Manoharachary and Rao, 1991), coastal tropical forest of Tamil Nadu (Ragupathy and Mahadevan, 1991; Bhaskaran, 1997). Shervaroyan hills of Tamil Nadu (Raman and Nagarajan, 1995), Tea plantations soils of Nilgiris of Tamil Nadu (Rajeshkumar, 2002; Rajeshkumar and Selvaraj,2006) and black pepper grown in the forest soils of Kerala(Lekha et al., 1995).

The diversity of AM fungi also studied in the soils of cultivated cereal crops and medicinal plants in TamilNadu(Selvaraj,1989;Mahesh,2002; Murugan,2002;Sankar,2002; Suresh and Selvaraj, 2006); and in the coastal regions of Kongan and Shervaroyan hills of Tamil Nadu (Gopinathan *et al.,* 1991) south east coast of Tamil Nadu(Nirmala and Selvaraj,2005),West Coast of India (Beena *et al.,* 2000) and Western Ghats of Goa (Khade and Rodrigues,2003).

The diversity of AMF in agricultural fields were also reported *Leucaena leucocephala* in Bangalore(Nalini *et al;*1986), tobacco in the cultivated fields of Tamil Nadu(Abdul Malik,2000), cultivated medicinal plants in Tamil Nadu viz., *Wedelia Chinensis* (Sankar,2002),*Cichorium intybus* (Murugan,2002), *Gloriosa superba* (Elango,2004); tea plantations in Nilgiris, (Rajesh kumar,2002; Kumaran and Santhanakrishnan,1995), ornamental and cultivated plants at Allahabad and adjoining areas (Kehri *et al*.,1987), in the fields of Pearl millet, maize, pigeon pea and chick pea in Gwalior (Singh and Pandya,1995), Tamil Nadu (Suresh and selvaraj,2006) and different agro climatic

regions of India (Singh and Adholeya,2002).

The distribution of AM fungi in stressed ecosystems has also been reported from coal, lignite and calcite mine soils of India(Mehrotra,1995), Kothagundam coal mine site, Andhra Pradesh(Rani *et al.*,1991), heavy metal polluted soils of Tamil Nadu(Sambandan et al.,1991; Mahesh,2002), petro-effluent irrigated fields, soils polluted with industrial and sewage effluents(Reddy *et al.*,1995;Mahesh,2002), tannery effluent polluted soils of Tamil Nadu(Raman *et al.*, 1995;Mahesh,2002).

Kandaswamy *et al.*, (1988), carried out an intensive survey for the prevalence of AM fungi in forest tree species occurring at different altitudes in Western Ghats of Nilgiris district, Tamil Nadu. Mohankumar and Mahadevan (1988) carried out ecological studies on AM fungal association with plant species from Kalakad reserve area located in the Western ghats, Tamil Nadu. They investigated the influence of edaphic factors and seasonal variation on the distribution of AM fungi in seven well defined ecosystems viz., evergreen, semi-evergreen, mixed deciduous, teak forests, scrub jungle and grasslands of high and low altitudes.

Rodrigues and Jaiswal (2001) recorded arbuscular mycorrhizal(AM) association in six plant species growing on sand dune vegetation of Goa. They reported the presence of three AM fungal genera viz., *Acaulospora*, *Glomus* and *Sclerocystis*. Khade and Rodrigues (2002) recorded AM fungal association in commonly occurring pteridophytes from two sites located in Western Ghats region of Goa. They recorded a total of 18 AM fungi belonging to 5 genera viz., *Acaulospora, Gigaspora, Glomus, Sclerocystis and Scutellospora.*

Rhizosphere soil is an imperative one, to assess the AMF diversity in the roots of host plant and also associated with a great variety of plants of different taxonomic groups (Jeffries, 1987).The rhizosphere is highly dynamic, plant driven micro environment, which is characterized by interaction between plant root process, soil characteristics and associated microbial population (Wenzel, *et al.,* 1999).

Soil moisture plays a significant role on mycorrhizal development and colonization (Singh, 2001).The occurrence of spores of Vesicular Arbuscular Mycorrhizas(VAM) in the rhizosphere, and the extent of root infection by VAM are reported for six Acacia species- *A. leucophloea*, *A. chundra*, *A. farnesiana*, *A. planiferons*, *A. auriculiformis* and *A. nilotica* in the Madurai district of Tamil Nadu. Altogether 19 species of VAM fungi belonging to 6 genera viz. *Acaulospora, Entrophospora, Gigaspora, Glomus, Sclerocystis and Scutellospora* were recorded (Santhaguru and Sadhana, 2000). The extent of VAM infection ranged from 56% (*A.auriculiformis*) to 80% (*A.leucophloea*).

Muthukumar and Udaiyan (2000) surveyed the arbuscular mycorrhizal (AM) status of plants growing in Western Ghats region of Southern India. They recorded 174 species of 329 plant species in such areas were mycorrhizal and 35 species belonging to *Acaulospora, Gigaspora, Glomus, Sclerocystis and Scutellospora.* The Vesicular Arbuscular Mycorrhizal status was examined in Pteridophytes of Western Ghats of Southern India by Muthukumar and Udaiyan (2000). According to them, sixty of the seventy

one species of Pteridophytes showed VAM association and the substratum strongly influenced the presence of mycotrophy; 100% of terricolous, 62% of the epiphytic and 79% of the lithophytic species had VAM infection.

Eriksen *et al.,* (2002) observed four species exhibited AM colonization, among the 82 plant species growing in traditionally managed grasslands in three different locations of Norway. The regeneration pattern and species diversity of Vesicular Arbuscular Mycorrhizal (VAM) fungi in shifting cultivated abandoned land (Jhum fallow) and natural forest soils of Arunachal Pradesh, North-eastern India was assessed (Suresh Kumar Singh *et al.,*2003). A total of 44 VAM species belonging to six genera namely *Acaulospora, Entrophospora, Gigaspora, Glomus, Sclerocystis and Scutellospora* were recorded from soils of Jhum fallow and natural forest sites.

AM fungal species richness, composition, spore density and diversity indices were evaluated in the *Phoenix sp*. area at Arizona (Cousins *et al.,* 2003). Vandenkoornhuyse *et al.,* (2003) assessed the diversity of AM fungi in 89 root samples of *Agrostis capillaries*, *Festuca rubra* and *Poa pratensis* using T-RFLP strategy. Camargo-Ricalde *et al.,* (2003) investigated the mycorrhizal status of perennial xeric plant species occurring in arid tropical scrub and ecotone of tropical deciduous forest communities in south central Mexico and found that 45 species were mycorrhizal among the 50 species sampled.

Most terrestrial plants associate with root colonizing mycorrhizal fungi, which improve the fitness of both the fungal and plant associates. The role of mycorrhizal fungi on sedge growth and nutrient uptake or non-nutritional benefits has yet to be fully ascertained. Muthukumar *et al.,* (2004) reviewed the current information available on the incidence of mycorrhiza in sedges and the possible reasons for low mycotrophy observed in this family.

Fifteen arbuscular mycorrhizal (AM) fungi are reported in the rhizosphere soil of three solanaceous vegetable namely, tomato, chilli and brinjal collected from five different locations (Reddy *et al.,* 2006). The genus Glomus is the most dominant fungus followed by *Acaulospora*, *Sclerocystis, Gigaspora* and *Entrophospora*.

Studies conducted on the microbial associates of Seabuckthorn (*Hippophae rhamnoides*) revealed the presence of 26 fungal species in its rhizosphere. Three fungal endophytes (*Mortierella minutissima* and a sterile mycelium) and four species of VAM spores (ie. *Glomus albidum, Glomus fasciculatum, Glomus macrocarpum and Gigaspora margarita)* has also been isolated from different plant parts (root, stem, leaves and bark) and soil samples (Shivkumar and Anand sagar, 2007).

A total of 122 AM fungi species with in 11 genera, including 8 new species, have been reported in various environments in China since 1980s.Advances in the last 20 years in AM fungal biodiversity and host diversity are reviewed by Fa Yuan Wang and Zhao Yong Shi (2008). Graminous plants establish strong symbiosis with arbuscular mycorrhizal fungi can improve the uptake of P from soil. *Sorghum bicolar* is asugar-rich multipurpose fodder crop well suited to dry farming. Deepadevi *et al.,* (2010) studied the response of *Sorghum bicolor (L.) Monech* to dual

inoculation with *Glomus fasciculatum* and *Herbaspirillum seropedicae*.

Mass production of AM fungi

The AM fungi are not host specific, any plant species can be infected by an AM fungal species but the degree of AM infection and its effect can differ with different host endophyte combinations.

Cultures of AM fungi on plants growing in disinfected soil have been frequently used technique to increase propagule numbers. A highly susceptible host plant should be used. It should produce abundant roots quickly and tolerate the high-light conditions required for the fungus to reproduce rapidly. Trap plants should be screened to ensure that maximum levels of inoculums were achieved (Bagyaraj and Manjunath, 1980).

Large quantities of the inoculum can be produced by pot culture technique (Wood, 1984). Plants with mycorrhizal associations predominate in most natural eco systems, so inoculum of mycorrhizal fungi is present in most soils. The quantity of inoculum of AM fungi were compatible with a host plant in soils can be measured by bioassay experiments. In these experiments, seedlings were grown in intact soil cores or mixed soil samples for sufficient time to allow mycorrhizas to form, and then roots were sampled, processed and assessed to measure mycorrhiza formation (Perry et al., 1982, Parke et al, Mc A Fee and Fortin 1986, Warcup, 1988, Brundrett and Abbott, 1995). Nelson and Safir (1982) investigated high level of root colonization in drought stressed plants.

Attempts have been made to use the genetic variability in fungal efficiency and host response to select AM fungal isolates to improve plant production (Trouvelot *et al.,* 1986).Variation in the effect of AM colonization has also been linked with genotype of host plant (Krishna *et al*., 1985; Lackie *et al.,* 1987).

Several host plants including Sudan Grass (*Sorghum bicolor var*. *suddanase*), bahia grass (*Paspalum notatum*), Cenchrus grass (*Cenchrus ciliaris*), Clover (*Trifolium subterraneum*), Strawberry (*Fragaria sp.*), Sorghum (*Sorghum vulgare*), Maize (*Zea mays*), Onion (*Allium cepa*) and Coleus (*Coleus sp*.) have been used for their suitability to multiply AM fungal inoculam. Sreenivasa and Bagyaraj (1988 b) reported that Rhodes grass (*Chloris gayana*) is the best host for mass multiplication of *Glomus fasciculatum*.

Bahia grass (*Paspalum notatum*) and industrial sweet potato (*Ipomea batatas*) colonized by *Glomus deserticola, G. etunicatum and G.intraradices* were grown in aeroponic cultures(Hung and Sylvia,1988).Infectivity studies comparing *G.etunicatum* spores from soil and aeroponic culture indicated no biological differences between the spore sources. Aeroponically produced *Glomus deserticola and G.etunicatum* inoculam retained their infectivity after cold storage ($4^{o}C$) in either sterile water or moist vermiculite for at least 4 and 9 months, respectively.

The traditional and most widely used approach has been applied to grow the fungus with the host plant in solid growth medium individually or a combination of the solid growth medium such as soil as soil, sand, peat, Vermiculite, perlite, clay or various types of composted barks(Tiwari and Adholeya,2002). However, there are some reports in which

AM colonization has been detected in Chenopods in the field and pot culture (Aquilera *et al.,* 1998, He *et al.,* 2002 and Johnson *et al.,* 1995).

Chourasia and Khare (2005) examined the four plant species viz. *Hordeum vulgare, Triticum aestivum, Phaseolus vulgaris and Phaseolus mungo* for mass production of consortium of AM fungi present in the rhizosphere soil. Such mass production of AM fungi was observed in terms of (%) AM colonization, AM consortia was recorded in terms of height and dry weight of inoculated and uninoculated plants. They observed that the *Hordeum vulgare* showed the highest colonization (92%) and 74 spores per 25 g soil.

Christine *et al.,* (2006) investigated that, invasion of AMF in grass lands of California. AM fungi colonizing roots were characterized with PCR amplifications of the ITS (Integrated sequence) region, cloning and sequencing. They also observed the significant effect of the presence of exotic grasses on the diversity of mycorrhizal fungi colonizing native plant roots.

Louis *et al.,* (2006) observed that the decreased level of colonizing at high concentration of copper in *Glomus* inoculated plants of *Aster tripolium L*. AM fungal colonizing significantly increases plant phosphorus uptake, but decreases soil phosphorus level in AMF inoculated plants of *Trifolium subterraneum*.

Effect of AM on host plant

Many species of fungi exhibit variation in enzyme banding patterns both in terms of total buffer soluble proteins (Seviour *et al.,* 1985) and specific enzymes (Alfenas *et al.,* 1984; Backhaus *et al.,* 1984 Maghrabi and Kish, 1985), it is essential that there should be similarities between isolates of the same species for identification. AM fungi can be identified within roots by differences in the mobility of specific fungal enzymes during polyacrylamide gel electrophoresis (Hepper *et al.,* 1986).

Subba Rao(1993) reported that the *Glomus mosseae* enhanced the accumulation of enzymes and amino acids resulting development of resistance in the host plants. In 1995, he also suggested that *Glomus mosseae* is known to enhance the enzyme activity and arginine accumulation, which develops resistance in host plants.

Cliquet and Stewart, (1993) reported that, the AM fungi are well known to bring about physiological changes in plants by means of increasing enzymatic activity. Panwar (1991) reported that nitrate reductase (NR) activity enhanced significantly in the mycorrhizal inoculated plants.

Dumas *et al.,* (1994) observed higher protein content in AM fungi inoculated *Nicotiana tabacum* and *Allium cepa* roots than the un-inoculated plants. Fatty acids derived from abundant phospholipids of AM fungi (located in membrane structures) and neutral lipids (located in storage structures) are potentially useful for estimating the biomass of infective AM propagules (Olsson et al., 1995). Thakur and Panwar (1995) reported that the AMF inoculation increased the root, shoot and total dry matter production in Mungbean.

Udaiyan and Sugavanam (1996) reported that inoculation of *Glomus fasciculatum* with plants of *Casuarina equisetifolia*,

results in the higher growth and biomass. Mudalagiriyappan et al., (1997) analyzed that AMF inoculation significantly increased in dry matter production, improved growth rate and net assimilation rate. Sitaramaiah *et al.,* (1998) reported that AMF inoculated plants increased vegetative growth, total chlorophyll content and uptake of nutrients like nitrogen, phosphorus, potassium, calcium and magnesium in maize plants.

The proposed model for carbon movement in the AM symbiosis include translocation of fungal lipids as a central route of carbon flow during both symbiotic and spore germination phases of the fungal life cycle(Bago *et al.,* 1999a;Bago *et al.,* 2000;Lammers *et al.,* 2001). The amount of neutral lipids is usually higher than that of phospholipids in AM fungi, since these fungi store a large proportion of their energy carbon as neutral lipids (Olsson and Johansen, 2000).

Ramesh *et al.,* (2000) reported that individual inoculation of AM fungi in *Pennisetum pedicellatum* enhanced the activity of alkaline phosphatase, super oxide dismutase (SOD), Chitinase and acid-β-glycerophosphatase. Bago *et al.,* (2000) suggested that the glyoxylate cycle may be central to the flow of carbon in the AM symbiosis. He carried out a study in the analysis of storage and structural carbohydrates labeling after $^{13}C_3$ glucose provided to AM roots and concluded that glycogen performs four roles in AM symbiosis viz.,sequestration of hexose taken from host. Long term storage in spores, translocation from intra-radical mycelium to extra-radical mycelium and buffering of intracellular hexose levels throughout the life cycle.

Chinnamuthhu and Venkata Krishnan (2001) reported that combined application of vermicompost with AMF significantly increased the yield of Sunflower. Madan *et al.,* (2002) reported the fatty acid methyl ester analysis in the spores of four AM fungi *Glomus coronatum, G. mosseae, Gigaspora margarita* and *Scutellospora calospora* and found out that 16:1 ω 5c to be the dominant fatty acid. Samanta *et al.,* (2003) reported the increasing plant height, number of leaves in combined application of phosphate solubilizer with AMF.

Ergosterol has been used as a biomass indicator to compare the growth of AMF (Hart and Reader, 2002a and b). Most members of the newly identified phylum Glomeromycota (Schubler *et al.,* 2001) fungal obligate symbionts forming arbuscular mycorrhizas contain sterol other than ergosterol. Fujiyuoshi *et al.,* (2000) found that the mycelium collected around roots colonized by *Gigaspora margarita* has 0.63 mg ergosterol in soil and roots (Hart and Reader, 2002a and b).

Van Aarle and Olsson (2003) compared the occuurence fatty acid with that of arbuscules and vesicles (lipid storage organs). The fatty acid 16:1, ω5 was used as an indicator for both AM fungal phospholipids and neutral lipids in roots and in soil. The formation of arbuscules and accumulation of AM fungal phospholipids in intraradical mycelium closely related. n contrast, the neutral lipids of *G. intraradices* increased continuously in the intraradical mycelium and it decreases in the vesicles after initial rapid root colonization by the AM fungus. The inoculation with exotic AM fungi increased the root and shoot NR activity(about 188% and 38% respectively) than the un-inoculated plants of *Juniperus oxycedrus* even under stressed conditions(Alguacil,2006).

Dual Effect of Biofertilizers

Mukherjee and Rai (2000) reported that the highest grain yield in *Triticum aestivum* at levels of phosphorus by inoculation of *Pseudomonas striata* followed by *Glomus fasciculatum*. The interactive effect of seed inoculation (with *Rhizobium*) and phosphorus application on growth and yield of Chickpea was studied at Agronomic Research Area, University of Agriculture, Faisalabad (Hakoomat Ali *et al.,* 2004). The results revealed that higher 1000- seed weight, seed yield and biological yield (256.10g, 3088.21 and 7496.99 kg ha^{-1}, respectively) were obtained with seed inoculation and 90kg $P_2O_5ha^{-1}$application (Inoc.1xP_3).

The effect of biofertilizers like *Rhizobium*, AM seedling production of *Acacia nilotica* was studied by Rajendran and Jayasree (2007). They showed that the total length of seedlings and biomass were significantly increased in all the treatments when compared to control. Among the treatments maximum growth and biomass were recorded in *Rhizobium* +AM + *Azospirillum* combination and it was 156.8% more than the control.

The efficacy of microbial bioagents for the control of collar rot disease in chick pea was studied (Ashraf Zahid *et al.,* 2007). Maximum reduction in seedling mortality was obtained in combination of *Rhizobium* and VAM compared to control (100% seedling mortality). It was inferred that both the amendments used have effectively improved various yield parameters by controlling the disease and reducing seedling mortality.

Inoculation effect of single and dual Vesicular Arbuscular Mycorrhizal with *Gigaspora rosea, Glomus intraradices* + *Gigaspora rosea* and *Glomus etunicatum* + *Glomus intraradices* on the growth and nutrients uptake (NPK) on *Medicago sativa* were examined and showed the significant increase in dry weight of shoot and root (Khan *et al.,*2008). The dual soil application of *Rhizobium* and VAM showed synergistic effect on all mung bean cultivars. Among form cultivars tested variety "Vaibhav" was found host responsive to root nodulation, growth parameters and grain yield (Manke *et al.*, 2008).

Effects of phosphate solubilizing microorganisms (*Glomus intraradices*, *Pseudomonas putida, P.aeruginosa*(Pa28), *P.alcaligenes, A. awamori)* and *Rhizobium sp*. Was examined on the growth, nodulation yield and root-rot disease complex of chickpea under field condition(Mohd. Sayeed Akhtar and Siddiqui,2009). The result showed that the number of nodules per root system was significantly higher in plants inoculated with Rhizobium sp. compared to control.

Bhat *et al.,* (2010) studied the effect of *Rhizobium* and Vesicular Arbuscular Mycorrhizae fungi on Green gram (*Vigna radiata L.Wilczek*) under temperate conditions. There was a significant effect of *Rhizobium* and VAM on nitrogen levels on nodulation, yield parameters, yield, NPK content in grain and straw and crude protein content in grain.

Rhizobium inoculation is a promising fertilizer because it is cheap, easy to handle and improves plant growth and seed quality. The growth and yield response of Chickpea (*Cicer arietinum*) to seed inoculation with *Rhizobium sp*. was studied (Giri *et al*., 2010). Such result revealed that, the bacterized seeds showed 14.06% in total length, 10.83% in total

weight and 9.0% on germination over control in pot experiment.

Arbuscular mycorrhizal fungi are ubiquitous in soil habitats and form beneficial symbiosis with the roots of angiosperms and other plants. Most terrestrial plants associate with root colonizing mycorrhizal fungi, which improve the fitness of both the fungal and plant associates. Ubiquitous occurrence and importance of AM fungi for plant growth is now a well established fact. Distribution and abundance of AM fungi vary greatly among different sites i.e. natural and manmade ecosystems (Chaurasia, 2001 and Gianinazzi-Pearson *et al.,* 1985).Natural soil offers consortium of indigenous mycorrhizal fungi and often used as source of inoculam. AM fungi can be produced on a large scale by pot culture technique. The beneficial use of AM inoculam in agriculture and raising nurseries has been reported (Muthukumar *et al.,* 2001; Smith and Read, 1997).From the above review, AM fungi plays a key role in crop field management with biofertilizers. They are also environment friendly fertilizers and do not cause the pollution of any sort.

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