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About TERI

A dynamic and flexible organization with a global vision and a local focus, TERI was established in 1974. While in the initial period the focus was mainly on documentation and information dissemination activities, research activities in the fields of energy, environment, and sustainable development were initiated towards the end of 1982. The genesis of these activities lay in TERI's firm belief that efficient utilization of energy, sustainable use of natural resources, large-scale adoption of renewable energy technologies, and reduction of all forms of waste would move the process of development towards the goal of sustainability.

The Bioresources and Biotechnology Division

Focusing on ecological, environmental, and food security issues, the Division's activities include working with a wide variety of living organisms, sophisticated genetic engineering techniques, and, at the grassroots level, with village communities. The Division functions through five areas—Centre for Mycorrhizal Research, Microbial Biotechnology, Plant Molecular Biology, Plant Tissue Culture, and Forestry/Biodiversity. The Division is actively engaged in mycorrhizal research. The Mycorrhiza Network has specifically been created to help scientists across the globe in carrying out research on mycorrhiza.

The Mycorrhiza Network and the Centre for Mycorrhizal Culture Collection

Established in April 1988 at TERI, New Delhi, the Mycorrhiza Network first set up the MIC (Mycorrhiza Information Centre) in the same year, and the CMCC (Centre for Mycorrhizal Culture Collection) – a national germplasm bank of mycorrhizal fungi – in 1993. The general objectives of the Mycorrhiza Network are to strengthen research, encourage participation, promote information exchange, and publish the quarterly newsletter, *Mycorrhiza News*.

The MIC has been primarily responsible for establishing an information network, which facilitates information sharing among the network members and makes the growing literature on mycorrhiza available to researchers. Comprehensive databases on Asian mycorrhizologists and mycorrhizal literature (RIZA) allow information retrieval and supply documents on request.

The main objectives of the CMCC are to procure strains of both ecto and VA mycorrhizal fungi from India and abroad; multiply and maintain these fungi in pure culture; screen, isolate, identify, multiply, and maintain native mycorrhizal fungi; develop a database on cultures maintained, and provide starter cultures on request. Cultures are available on an exchange basis or on specific requests at nominal costs for spore extraction/handling.

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Mycorrhizal dependency, Part 2: determinants of mycorrhizal dependency and intraspecific variation

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Determinants of mycorrhizal dependency

Many soil and plant parameters can indicate whether or not a particular plant is dependent on mycorrhizal association in roots for its better growth. These parameters are briefly discussed.

Phosphorus utilization by plants

One of the most dramatic effects of mycorrhizal infection on the physiology of host plants is an increase in phosphorus absorption, especially in phosphorus-deficient soils. This aspect is adequately analysed for VAM (vesicular–arbuscular mycorrhizal) fungi by Koide (1991). When phosphorus is limiting, the maximum extent to which mycorrhizal infection can improve plant performance is predicted to be a function of the phosphorus deficit of the plant, which is the difference between phosphorus demand and phosphorus supply. Phosphorus demand is defined as the rate of phosphorus absorption under prevailing conditions. Variations among plant taxa in morphological, physiological, or phenological traits that affect either phosphorus demand or phosphorus supply (and thus phosphorus deficit) are predicted to lead to variations in the potential response to mycorrhizal infection. The actual response to mycorrhizal infection is predicted to be a function of the increase in phosphorus uptake due to mycorrhizal infection and the phosphorus utilization efficiency of the plant. However, mycorrhizal infection may alter the phosphorus deficit or phosphorus utilization efficiency independent of its direct effect on phosphorus uptake, making the prediction of response to mycorrhizal infection based on the traits of non-mycorrhizal plants quite difficult. Infection may at times increase the rate of phosphorus accumulation beyond what can be currently utilized in growth, reducing the current phosphorus utilization efficiency. Such momentary 'luxury consumption' of phosphorus may, however, have a storage function and may be utilized subsequently (Koide 1991).

Greenhouse investigations conducted at the Department of Agronomy and Soil Science, University of Hawaii, Honolulu, USA, showed that plant species that were marginally dependent on VAM fungi (e.g. *Sesbania pachycarpa*) tended to have lower phosphorus utilization efficiency, higher phosphorus absorption rate, and higher growth rates compared with moderately (e.g. *Leucaena retusa* and *S. grandiflora*) or highly (e.g. *L. leucocephala*) VAM dependent species when they were not colonized by the mycorrhizal fungus *Glomus aggregatum*. It was also suggested that species differing in growth rates could differ in their mycorrhizal dependency even when they had similar root morphological characteristics (Manjunath and Habte 1991a).

In another study conducted at the above university on selected species of *Leucaena* and *Sesbania*, mycorrhizal dependency was determined at three phosphorus levels on fumigated oxisol, inoculated or not with *Glomus aggregatum*. Under non-mycorrhizal conditions, *L. leucocephala*, a highly VAM dependent species, was observed to have a higher external and lower internal phosphorus requirements than those of *S. pachycarpa*, a marginally VAM dependent species. A sequential harvest experiment indicated that highly dependent plant species have lower growth rates, low

* Compiled from TERI database – Riza

phosphorus uptake rate, and higher phosphorus utilization efficiency than marginally dependent species under non-mycorrhizal conditions. These results illustrate that soil solution phosphorus levels can serve as a basis for categorizing plant species in distinct mycorrhizal dependency groups (Manjunath 1990).

Studies conducted at the Centro Int Agric Tropical Apartado Acreo, Cali, Colombia, on 24 tropical forage legumes and grasses, with or without inoculation with mycorrhizal fungi, showed that mycorrhizal inoculation significantly increased shoot and root dry weights and total uptake of phosphorus, nitrogen, potassium, calcium, and magnesium of all test plants and, with few exceptions, decreased the root/shoot ratio. Non-mycorrhizal plants always contained lower quantities of mineral elements than mycorrhizal plants did. There was no correlation between percentage mycorrhizal infection and plant growth parameters. Total uptake of all elements by non-mycorrhizal legumes and uptake of phosphorus, nitrogen, and potassium by non-mycorrhizal grasses correlated inversely with mycorrhizal dependency. Utilization of soil phosphorus by non-mycorrhizal plants correlated inversely with mycorrhizal dependency (Saif 1987).

Studies conducted at the Kansas State University, Department of Plant Pathology, Manhattan, USA, on 10 wheat cultivars showed that six cultivars responded positively to five mycorrhizal fungi whereas four responded negatively or were non-responsive. Response of an individual cultivar was consistent regardless of the inoculum sources, suggesting that mycorrhizal responsiveness is an inherent trait rather than a response to individual fungi. Mycorrhizal responsiveness decreased with use of phosphorus fertilizes for cultivars that were dependent on the symbiosis but was unaffected in cultivars negatively impacted by mycorrhizae. Mycorrhizal and phosphorus responsiveness of each cultivar were highly correlated ($r = 0.94$), suggesting that phosphorus responsiveness may be a good predictor of the mycorrhizal dependence of selected wheat cultivars. The relationship between wheat biomass production and percentage root colonization was positive for cultivars responding favourably to symbiosis but negative for those responding negatively or being non-responsive to symbiosis. Amendment with phosphorus did not significantly affect these relationships (Hetrick, Wilson, and Todd 1996).

Leaf phosphorus

Studies conducted at the Unite de Recherche in Biologie et Agro-Foresterie, Universite de Tizi-Ouzou, Algeria, on 11 *Eucalyptus* species showed that 20 weeks after VAM inoculation, stem dry weights of *Eucalyptus* seedlings could be increased by up to 49% when compared to non-inoculated control plants. Intensity of root colonization by a given fungus depended on the host species but was not related to plant growth response. Leaf phosphorus concentration of non-inoculated *Eucalyptus* seedlings varied greatly among species. Increases in leaf phosphorus concentration following mycorrhizal infection were not necessarily associated with plant growth stimulation. The most mycorrhiza-dependent *Eucalyptus* species tended to be those having the highest leaf phosphorus concentration in the absence of a fungal symbiont. These mycorrhiza-dependent *Eucalyptus* species seem to have greater phosphorus requirement and, consequently, seem to rely more on the symbiotic association (Adjoud, Plenchette, Halli-Hargas et al. 1996).

Phosphorus level in cuttings

In studies conducted at the Department of Agronomy and Soil Science, University of Hawaii, USA, cassava plants were grown in greenhouse either from small cuttings (2.0 mg phosphorus per cutting) or large cuttings (20.2 mg phosphorus per cutting) in a subsurface oxisol, inoculated or not inoculated with *Glomus aggregatum* at target soil solution phosphorus concentrations of 0.003–0.02 mg/L. VAM colonization levels in excess of 60% were attained on cassava roots irrespective of the size of cutting material used or target soil solution phosphorus status. However, plants started from large cuttings grew better and faster than those started from smaller cuttings. Cassava was found to be very highly dependent on VAM fungi if grown from small cuttings, but only marginally dependent on VAM fungi when started from large cuttings. This appears to be related to the high phosphorus reserve in large cuttings and hence the low requirement of plants for soil phosphorus until the phosphorus reserves in the cuttings are significantly depleted (Habte and Byappanahalli 1994).

Root morphology

In studies conducted at the University of Hawaii, Honolulu, USA, on selected species of *Leucaena* and *Sesbania*, mycorrhizal dependency was determined at established soil phosphorus levels of 0.002–0.02 and 0.2 mg/L in a fumigated oxisol, inoculated or not inoculated with *Glomus aggregatum*. At a soil phosphorus level of 0.02 mg/L, stepwise regression analysis showed that 85% of the variability in mycorrhzial dependency was explained by root dry weight, root hair length, root density, root diameter, and root hair incidence. As all species tested reduced the pH on an agar medium, there appears to be no relationship between mycorrhizal dependency and ability of *Leucaena* and *Sesbania* to reduce rhizosphere pH (Manjunath 1990).

In another study conducted at the above university, *Leucaena diversifolia*, *L. retusa*, *L. trichodes*, *Sesbania formosa*, *S. grandiflora*, *S. pachycarpa*, and *S. sesban* were grown in greenhouses and growth chambers in soil inoculated with *Glomus aggregatum*. Compared with the moderately to very highly mycorrhizal dependent *Leucaena* spp., the

marginally to moderately dependent *Sesbania* spp. were characterized by higher root weight, root density, root surface area and root length, smaller root diameter, a higher percentage of root hair incidence, shoot to root ratio, and total phosphorus uptake. The two groups of species were not consistently different from each other with respect to mycorrhizal colonization level, root hair diameter, root hair length, phosphorus uptake per unit root surface area, and acid production in agar medium. A stepwise regression model in which mycorrhizal dependency was used as a dependent variable and root characteristics as an independent variable suggested that root weight, root hair length, root diameter, root density, and root hair incidence were important determinants of mycorrhizal dependency, with root weight accounting for 65.5% of the variability (Manjunath and Habte 1991b).

Studies conducted at the Department of Plant Pathology, University of Kansas, Manhattan, USA, showed that inoculated warm-season species (*Andropogon gerardii*, *Penicum virgatum*, *Sorghastrum nutans*, and *Bouteloua curtipendula*) and cool-season species (*Koeleria cristata*, *Bromus inermis*, *Festuca arundinacea*, *Lolium perenne*, *Agropyron smithii*, and *Elymus cinereus*) of mycorrhizal grasses had respectively 63–215 and 0.12–14.1 times higher dry weights than uninoculated controls. Non-mycorrhizal warm-season plants did not grow and frequently died whereas cool-season grasses grew moderately well in the absence of mycorrhizal symbiosis. Like warm-season grasses, tall grass prairie forbs (*Liattris aspera*, *Dalea purpurea*, and *Baptisia leucantha*) were highly dependent on mycorrhizal symbiosis even though they are not known to employ the C4 photosynthetic pathway. Thus, phenology may be more critical than the photosynthetic pathway in determining mycorrhizal dependence. Warm-season grasses and forbs had a coarser, less frequently branched root system than cool-season grasses did, supporting the hypothesis that mycorrhizal dependence is related to root morphology. Cool-season grasses may have developed more fibrous root systems because mycorrhizal nutrient uptake was not effective in the cooler temperature environment in which they evolved. In contrast, warm-season plants and dependence on mycorrhizal fungi may have coevolved because both symbionts are of tropical origin (Hetrick, Kitt, and Wilson 1988).

Further studies at the above university showed that cool-season grasses (*Bromus inermis*, *Elymus cinereus*, *Festuca arundinacea*, *Koeleria pyramidala*, and *Lolium perenne*) had significantly more primary and secondary roots than warm season grasses had (*Andropogon gerardii*, *Schizachyrium scoparium*, *Penicum virgatum*, *Bouteloua curtipendula*, and *Sorghastrum nutans*). The diameter of primary, secondary, and tertiary roots of cool-season grasses was significantly smaller than that of warm-season grasses. Soil microorganisms, mycorrhizas (*Glomus etunicatum*), and phosphorus fertilizer application (0.15 or 45 mg/kg soil) did not affect root number or root diameter of the cool-season grasses. Root number of warm-season grasses did respond to mycorrhiza and phosphorus fertilization but not to soil microorganisms. Specific root length of cool-season grasses was not altered by mycorrhizas, soil microorganisms, or phosphorus fertilization and was significantly greater than that of warm-season grasses but had no effect on the rooting strategy of cool-season grasses. In contrast, phosphorus fertilizers did not substantially alter root branching in warm- or cool-season grasses. Apparently, root architecture of the mycorrhiza dependent warm-season grasses is quite plastic, allowing energy expenditure for root development to be conserved. The root architecture of the less mycorrhiza dependent cool-season grasses appeared to be fixed and did not alter to accommodate the symbiosis (Hetrick, Wilson, and Leslie1991).

In studies conducted at the Department of Horticultural Science, Texas A&M University, USA, seeds of 19 cowpea (*Vigna unguiculata*) cultivars with known mycorrhiza dependency levels were inoculated with *Glomus fasciculatum* and *Bradyrhizobium* in seedling trays, and 12-day-old seedlings were transferred to a hydroponic culture system where they were grown for five weeks. Leaf area, length of tap root, total root length, root weight, root abundance, average length of fine roots, number of nodules formed on lateral roots, and total nodules weight differed among cultivars. Less than 5% of the root length was colonized by mycorrhizal fungus in all cultivars. Average length of fine roots negatively correlated with mycorrhizal dependency of the cowpea cultivars. However, only 27% of the variability in mycorrhizal dependency was explained by this variable. Therefore, root morphology did not appear to determine mycorrhizal dependency in cowpea (Rajapakse and Miller 1988).

Studies conducted at the Iowa State University, Department of Agronomy, Ames, USA, on three cultivars of soybean with high, intermediate, and low mycorrhizal dependencies showed that *Glycine soja* PI 964816 ('Soja'), a highly mycorrhiza dependent cultivar, developed an active symbiosis more quickly than low mycorrhizal dependent *G. max* cv. swift (Swift) or intermediate mycorrhiza dependent *G. max* cv. mandarin ('Mandarin'). Non-mycorrhizal 'Soja' root lengths (average 1401 cm) at five weeks of growth were significantly shorter than 'Mandarin' (2303 cm) and 'Swift' (2236 cm) root lengths. The mean diameter of the lateral roots of 'Soja' (0.38 mm) was greater than that of 'Mandarin' or 'Swift' (approx.0.26 mm). Root lengths and root surface area correlated negatively ($p = 0.01$) with mycorrhizal dependency (Khalil, Loynachan, and Tabatabai 1999).

Studies conducted at the INRA, Station D' Agronomie, Cedex, France, on seven cultivars of

banana (*Musa acuminata* AAA group), inoculated with *Glomus mosseae* and *G. macrocarpum*) in greenhouses showed that both root dry weight and root hair length or density of the non-inoculated plants inversely correlated with the relative mycorrhizal dependency values of cultivars (DeClerck, Plenchette, and Strullu 1995).

In studies conducted at the Department of Forestry and Natural Resources, Purdue University, West Lafayette, USA, on four hard woods (*Fraxinus pennsylvanica*, *Liquidambar styraciiflua*, *Liriodendron tulipifera*, *Platanus occidentalis*) inoculated with six VAM fungi, an inverse correlation was observed between lateral root length, root dry weight, root fibrosity of uninoculated control plants, and mycorrhizal dependency of each tree species. Also, mycorrhizal dependency was directly correlated with root/shoot ratio (Pope, Chaney, and Rhodes 1983; Pope, Rhodes, Chaney et al. 1981).

Other parameters

Studies conducted at the Department of Plant Pathology, Kansas State University, Manhattan, USA, on growth of VAM and non-VAM big blue stem in 15 soils showed that there were three different growth-response groups. Group 1, which included the prairie soils, displayed high mycorrhizal dependency and significant suppression of plant growth in non-sterile soil. Group 2 showed no mycorrhizal dependency and plant growth was suppressed on non-sterile soil. Group 3 exhibited no mycorrhizal dependency or suppression in non-sterile soil. Mycorrhizal dependency of big blue stem was not highly correlated with any individual soil parameter. However, concentrations of soil potassium, calcium, copper, and iron were used to successfully classify high or low mycorrhizal dependency. The mechanism of non-sterile soil suppression is not clear; however, the nutrient that predicts suppression may be related to the development of mycorrhizal symbiosis or in demand by plants or other microbes. These nutrients that predict mycorrhizal dependency may be related to those supplied to the plant by symbiosis (Kitt, Hetrick, and Wilson 1988).

Studies conducted at the University of Hawaii, Honolulu, Hawai, USA, showed that species such as *Sesbania pachycarpa* that are marginally dependent on VAM have higher growth rates compared with moderately dependent (*Sesbania grandiflora*, *Leucaena retusa*) and highly dependent (*L. leucocephala*) species when they are not colonized by the VAM fungus *Glomus aggregatum*. It is, therefore, suggested that species differing in growth rates can differ in their mycorrhizal dependency even when they have similar root morphological characteristics (Manjunath and Habte 1991).

A glasshouse study was conducted at the Citrus Research and Education Centre, University of Florida, Lake Alfred, Florida, USA, at low and high phosphorus supply, with five mycorrhizal (M) or non-mycorrhizal (NM) genotypes of Citrus, namely *Citrus volkameriana*, sour orange, *Poncirus trifoliata*, 'Swingle citrumelo', and 'Carrizo citrange'. Lower total incidence of *Glomus intraradices* (FL 208), intensity of vesicle formation, and accumulation of 16:1 sub (W5) cis fungal fatty acid (as measures of root colonization) were related to lower MD (mycorrhizal dependency) in five Citrus genotypes. At high phosphorus supply, when host carbon (C) production was not affected by phosphorus nutrition, less M-dependent genotypes had consistently lower starch concentrations in their root and shoot tissues than did more M-dependent genotypes, irrespective of mycorrhizal inoculation. At low phosphorus supply, M plants were more heavily colonized by *G. intraradices* and had lower starch contents than NM plants supplied with additional phosphorus. At high phosphorus, M plants of more dependent genotypes allocated more C to starch pools relative to NM plants than in the least dependent genotypes. Tissue sucrose concentrations did not vary consistently with the dependency of the genotype or with M inoculation except in high phosphorus NM plants when less M-dependent genotypes had lower root sucrose concentrations than the more dependent species. At high phosphorus supply, sucrose concentrations were lower in colonized roots than in NM roots. Across Citrus genotypes, sucrose in M fibrous roots decreased relative to NM roots, with increase in root colonization suggesting that more sucrose was allocated for growth and maintenance of *G. intraradices* in roots of M-dependent species. The concentrations of reducing sugars in root tissues varied in relation to MD of citrus genotypes in the same way as that of starch and sucrose, but was less responsive to M colonization. Response of total non-structural carbohydrates in tissues indicated that more heavily colonized plants at low phosphorus expended more C to acquire phosphorus than did M plants of similar biomass and phosphorus status grown at high phosphorus supply. Total carbohydrate pools increased with MD of Citrus genotypes, providing evidence that C allocation patterns in the host affect M colonization (Graham, Duncan, and Eissenstat 1997).

Intraspecific variation in mycorrhizal dependency

There may be a lot of variation within a plant species in its various varieties/provenances/cultivars. Such a variation is highlighted below.

Allium cepa

In studies conducted at the Tata Energy Research Institute, New Delhi, India, four onion (*Allium cepa*) varieties, namely Pusa White Flat (PWF), Pusa White Round (PWR), Early Grano (EG) and Pusa Madhvi (PM), were grown in phosphorus-deficient alfisol at two phosphorus levels (25 and

50 kg P/ha) and were inoculated with mixed culture of indigenous VAM fungi. At harvest, all inoculated onion varieties showed higher values of bulb diameter, fresh weight, shoot dry matter, shoot P content, and bulb yield than uninoculated plants. Inoculated plants tended to have greater bulb yield for varieties PM and PWR grown at 25 kg P/ha. On the other hand, PWF and EG plants showed similar responses at 50 kg P/ha. The per cent root length colonized by VAM fungi between both the P levels of inoculated plants did not differ significantly, but the extent of colonization varied among the varieties. EG and PWF plants exhibited maximum dependence on VAM at 50 kg P/ha where as PM and PWR plants exhibited a maximum mycorrhizal dependency at 25 kg P/ha (Sharma and Adholeya 2000).

Capsicum annum

Studies conducted at the Department of Agricultural Microbiology, University of Agricultural Science, Dharwad, India, on six chilli (*Capsicum annum*) cultivars showed that all cultivars responded well to soil inoculation with *Glomus macrocarpum*. Highest percentage root colonization and spore count were observed on the Byadagi cultivar and lowest on the G-3 cultivar. Similar trends were observed for shoot phosphorus concentration, plant dry mass, and yield (Sreenivasa and Gaddagimath 1993).

Citrus spp.

In studies conducted at the Indian Institute of Horticultural Research, Hassarghatta, Bangalore, India, seedlings of root stocks of rough lemon, Rangpur lime (*Citrus limonia*), *Ponicirus trifoliata*, Troyer citrange, Carrizo citrange, Citrumelo and Cleopartra mandarin, with three to four leaves, were inoculated with *Glomus fasciculatum* in pots containing 5 kg of alfisol (pH 6.8, phosphorus 5 ppm, carbon 0.72%). All plants received 100 ppm nitrogen as calcium ammonium nitrate and 50 ppm potassium as potassium chloride. Mycorrhizal dependency was calculated 160 days after planting as the per cent improvement in dry weight due to VAM inoculation. *P. trifoliata* was found most dependent root stock, followed by rough lemon, Troyer citrange, Rangpur lime, and Carrizo citrange. *Coleopatra* mandarin was least dependent (Onkarayya and Sukhada 1993).

In studies conducted at the University of Florida, Citrus Research and Education Centre, Lake Alfred, USA, seedlings of five Citrus root stocks were grown in a low-phosphorus sandy soil and were either inoculated with *Glomus intraradices* or non-inoculated and fertilized with phosphorus or non-inoculated and unfertilized with phosphorus. The order of mycorrhizal dependency in five root stocks was sour orange, Cleopartra mandarin, Swingle citrumelo, Carrizo citrange, and trifoliate orange (Graham and Syvertsen 1985)

Coconut

In studies conducted at the Division of Microbiology, Central Plantation Crops Research Institute, Kasaragod, Kerala, India, in one-year-old seedlings of 17 tall and dwarf cultivars and four hybrids of coconut growing on a sandy loam soil, the proportion of root segments with VAM ranged from 56.8% to 95.2%, being highest in the tall Laccadive ordinary and significantly higher in the tall cultivars than in the dwarfs or hybrids. Similar trends were observed for infection grading (a measure of colonization intensity) and for numbers of spores in the soil around the seedlings (Thomas and Ghai 1987).

Eleusine coracana

Studies conducted at the Department of Microbiology, College of Basic Science and Humarnities, G B Pant University of Agriculture and Technology, Pant Nagar, India, showed that out of five genotypes of *Eleusine coracana* (HR-374, PRS110, Pe-4, PES 4000, and PES-176) tested for mycorrhizal dependency against *Glomus caledonium*, HR-374 gave the highest plant biomass, mycorrhizal efficiency, and root colonization. The mycorrhizal inoculation resulted in increased mineral contents (phosphate, nitrogen, zinc, and copper) and mineral uptake in shoots (Tewari, Johri, and Tandon 1993).

Eucalyptus spp.

Studies conducted at the Research Institute of Tropical Forestry, Chinese Academy of Forestry, Longdong, Guangzhou, China, on four provenances of *Eucalyptus urophylla* showed that there was a marked variation in plant dry weight and phosphorus content between *E. urophylla* provenances at 10 mg P/kg soil, but variations were less at 5 mg P/kg and 20 mg P/kg soils. Inoculation of all the four provenances with *Pisolithus tinctorius* and *Scleroderma cepa* showed increased biomass and phosphorus uptake at 5 mg P/kg soil and reduced or no effect on biomass and phosphorus uptake at 20 mg P/kg soil (Xu DaPing, Malajczuk, and Grove 1995).

Glycine max and G. soja

In studies conducted at the Iowa State University, Department of Agronomy, Ames, USA, on three cultivars of soybean (with high, intermediate, and low mycorrhizal dependencies), shoot weight, phosphorus percentage in shoot tissues, total shoot phosphorus uptake, phosphorus use efficiency, rate of growth, and root acid phosphatase activity were measured at three harvests when plants were grown at three relative phosphorus levels, each with or without the mycorrhizal fungus *Gigaspora margarita* colonization. *Glycine soja* PI468416 ('Soja'), a highly dependent cultivar, developed an active symbiosis more quickly than low mycorrhizal dependent *G. max* cv. swift ('Swift') or intermediate mycorrhizal dependent *G. max* cv. mandarin

('Mandarin'). When averaged across harvests at low phosphorus, mycorrhizal 'Soja' had 7.8 times more total shoot phosphorus than non-mycorrhizal 'Soja' did; comparable values were 2.4 times for 'Mandarin' and 1.5 times for 'Swift'. 'Soja' also showed higher phosphatase activity and a higher percentage increase in phosphatase activity with mycorrhizal colonization than other cultivars did. Non-mycorrhizal 'Soja' root lengths (average 1401 cm) at 5 weeks of growth were significantly shorter than root length of Mandarin (2303 cm) and Swift (2236 cm). The mean diameter of lateral roots of 'Soja' (0.38 mm) was greater than that of 'Mandarin' or 'Swift' (approx. 0.26 mm) (Khalil, Loynachan, and Tabatabai 1999)

Studies conducted at the LW Lambourn and Co., Carolyn House, Croydon, England, on 10 recent selections of promiscuous soybean breeding lines and two herbaceous legumes (*Lablab purpureus*, *Mucuna pruriens*) grown in pots in soil with available phosphorus of 5.33 µg/L soil showed that mycorrhizal colonization differed among promiscuous soybean lines and ranged from 16% to 33%. Mycorrhizal colonization was, on an average, 20% for mucuna and lablab. Three groups of plants were obtained according to their mycorrhizal dependency. These were (1) highly dependent: a mycorrhizal dependency of more than 30% (soybean line 1039 and mucuna), (2) intermediate: mycorrhizal dependency between 10% and 30% (soybean lines 1576 and lablab), and (3) not mycorrhizal dependent: majority of the soybean lines (5 lines) (Nwoko and Sanginga 1999).

Helianthus annuus

In studies conducted at the University of Agricultural Sciences, Dharwad, Karnataka, India, sunflower (*Helianthus annuus*) cvs. Morden and MSFH-8 were grown in summer of 1993 on medium black soil with 38, 56, or 75 kg P_2O_5 per ha and without or with VAM (*Glomus asciculatum*) inoculation (2 cm below the seeds). Mean seed yield was 1853 and 1628 kg/ha with and without VAM inoculation respectively. Seed yield was higher in MSFH-8 than in Morden. Seed yield of inoculated plants given 38 kg P_2O_5 was similar (MSFH-8) or superior (Morden) to yield of uninoculated plants given 75 kg P_2O_5 per ha. Per cent mycorrhizal root colonization was 74.6%–88.1% in inoculated and 40.5%–47.0% in uninoculated plants and in each case decreased with increase in phosphorus (Chandrashekara, Patil, and Sreenivasa 1995).

Hordeum vulgare

In studies conducted at the Department of Soil Science, Waite Institute, University of Adelaide, Glen Osmond, Australia, eight barley (*Hordeum vulgare*) cultivars ('Kaniere', 'yagan', 'O Connor', 'Galeen', 'WI2539', 'WI2737', 'Skiff' and 'Shannon') were grown in pots with sterilized sandy soil having 4 mg P/kg soil. Three rates of phosphorus, i.e. 0, 10, and 20 mg/kg soil, were added as KH_2PO_4. The plants were inoculated or not inoculated with *Glomus etunicatum* and were grown for 42 days with the soil temperature at 15 °C. Significant differences were observed among the barley cultivars in the percentage of root length infected, the range being 1.1–31.7. Phosphorus concentration in plant tissues and percentage of root length infected were not significantly correlated. Agronomic phosphorus efficiency of the barley cultivars was negatively correlated with the response of the plants to both mycorrhiza and phosphorus application. Inefficient cultivars ('Shannon') had high responses to both mycorrhiza colonization and phosphorus addition than efficient cultivars ('Yagan') (Baon, Smith, and Alston 1992).

Ipomoea batatas

Studies conducted at the Department of Plant Pathology, University of Georgia, Athens, USA, on three sweet potato (*Ipomoea batatas*) cultivars inoculated with VAM and grown in different phosphorus fertility regimes showed that all the three cultivars had a low degree of mycorrhizal dependency (Negeve and Roncadori 1985).

Lycopersicum esculentum

In studies conducted at the Department of Plant Pathology, University of Kentucky, Lexington, USA, seeds of two cultivars (cvs. Manapal and Walter) of tomato (*Lycopersicum esculentum*) were inoculated with *Glomus etunicatum* or *G. mosseae* with several inoculum levels. In cv. Manapal, *G. etunicatum* inoculation resulted in significantly greater ($p = 0.001$) shoot dry weights and plant heights than did *G. mosseae* and non-inoculated controls at 6 and 13 weeks. In cv. Walter, however, inoculated plants with *G. mosseae* had significantly greater shoot dry weight and plants heights than did *G. etunicatum* at 6 and 13 weeks (McGraw and Schenck 1991).

Medicago sativa

In experiments conducted at the Department of Botany, University of Guelph, Ontario, Canada, on alfalfa (*Medicago sativa*), 20 half-sib families from within each of two populations (Pioneer 530 and OAC78-101) were assessed for VAM formation with *Glomus versiforme*. Mean percentage colonization was 5.8% and 6.8% for Pioneer 530 and OAC78-101 respectively. Significant ($p = 0.05$) differences in mean percentage colonization were detected among half-sib families derived from Pioneer 530 but not among those derived from OAC78-101. Significant ($p = 0.05$) 'pot and run' effects were detected in the latter experiment, which may have masked differences among families (Lackie, Bowley, and Peterson 1988).

Studies conducted at the Department of Plant Pathology and Agronomy, the Pennsylvania State University, USA, on six clones from each of six alfalfa (*Medicago sativa*) varieties grown with or without mycorrhiza at 0 or 8 ppm phosphorus in a low-phosphorus pasteurized soil showed significant mycorrhiza × variety interactions for shoot dry weight and zinc concentration but not for phosphorus and copper concentrations. Mycorrhiza × clone interactions for shoot dry weight and phosphorus, zinc, and copper concentrations were significant within several varieties. Shoot dry weights inversely correlated with arbuscule frequencies and directly correlated with vesicle frequencies in the low but not in the high phosphorus treatments (Lambart, Cole, and Baker 1979)

Musa acuminata

Studies conducted at the INRA, Station D' Agronomie, Cedex, France, on seven banana cultivars (*Musa acuminata* AAA group) inoculated with *Glomus mosseae* and *G. macrocarpum* in greenhouses showed that cv. Williams had the highest and cv. Poyo had the lowest relative mycorrhizal dependency (DeClerck, Plenchette, and Strullu 1995).

Oryza sativa

Studies conducted at the Division of Plant Pathology, ICAR Research Complex for North East Hill Region, Barapani, Meghalaya, India, on two rice varieties showed that the phosphorus-deficiency-tolerant variety (RCPL101) responded to mycorrhiza up to a soil phosphorus level of 20 kg/ha. A susceptible phosphorus-deficiency variety (RCPL104) responded up to 80 kg P/ha. Thus, the variety requiring high phosphorus depended on VAM even at high soil phosphorus level (Singh and Mishra 1995).

Pennisetum americanum

Studies conducted at the International Crop Research Institute for Semi Arid Tropics, Patancheru, Andhra Pradesh, India, on pearl millet (*Pennisetum americanum*) genotypes showed that for 30 genotypes tested across three field locations, there was a range of mycorrhizal colonization intensity between 25% and 56%. In another experiment with two male sterile lines, restore lines, and their derived crosses grown in pots filled with non-sterilized soil, there were significant differences between genotypes for colonization by mycorrhiza that showed host–genotype dependence for mycorrhizal colonization (Krishna, Shetty, Dart et al. 1985).

Pinus taeda

Studies conducted at the School of Forestry, Auburn University, Auburn, USA, on loblolly pine (*Pinus taeda*) progenies (from an Oklahoma USA tree improvement programme) inoculated with different isolates of *Pisolithus tinctorius* showed genetic variation in ectomycorrhizal development, shoot height, component dry weight, and net assimilation rate. Rapid growing progenies exhibited superior mycorrhizal colonization and a positive correlation between infection and total dry weight (Dixon, Garrett, and Stelzer 1987).

Populus deltoides

Studies conducted at the Biomass Research Centre, National Botanical Research Institute, Lucknow, India, on *Populus deltoides* clones showed that out of nine clones growing at the Banthra Research Station, three formed endomycorrhizal association with *Glomus intraradices* and *Scutellospora gigantea*. A wide variation in the extent of root infection was found between the clones even when abundant spore inoculum was present (Sindhu, Misra, and Behl 1990).

Solanum writti

Studies conducted at the Department of Plant Pathology and Agricultural Microbiology, Mahatma Phule Agricultural University, Rahuri, India, on brinjal (*Solanum writti*) showed that the shoot and root dry weights in mycorrhizal plants of various genotypes were respectively 1.20–3.83 times and 1.03–1.78 times that of non-mycorrhizal plants. The mycorrhizal dependency of the genotypes ranged from 117.20% to 309.91%. An increase in P uptake by 11 genotypes due to mycorrhizal inoculation was 1.27–4.3 times that of comparable controls. *Solanum writti*, and four genotypes (PS-8, Dorli, Pragati and Borgaon 1) were more responsive to inoculation with *Glomus fasciculatum* than the other six genotypes (Vaishali, Manjri, Gota, Annamali, Krishnakathi and P P Long) tested. In general, the results indicated a genotype-dependent variation in dry matter production and phosphorus uptake as influenced by inoculation with a VAM fungus (Indi, Konde, and Sonar 1990).

Sorghum bicolor

Studies conducted at the IARI Regional Station, Rajedranagar, Hyderabad, India, on 47 diverse varieties and exotic germplasms of *Sorghum bicolor* showed that VAM infection ranged from 25% to 95% in different cultivars. Restorer lines of hybrids such as PD-3-1-11, PVR-10, M-148, RS-29, and RS-71 showed maximum VAM colonization and high phosphorus content in shoot and root. Certain genotypes of sorghum were adapted better than others to phosphorus-deficient conditions and showed higher efficiency in their uptake. The data on correlation revealed significant and positive associations of VAM infection with phosphorus content of shoot and root and dry matter production (Subhashini, Rana, and Potty 1988).

Triticum spp.

In studies conducted at the Department of Agronomy and Natural Resources, Institute of Field and Garden Crops, Bel Dagan, Israel, on wild and cultivated primitive and modern wheat (nine

Triticum spp. and four *Aegilops* spp.) showed that mycorrhizal colonization in *T. limupheevii* var. *araraticum* (*T. araraticum*) (AAGG) was higher than that found in other tetrapoloid wheats (AABB). Mycorrhizal dependency was higher in representation of the D genome donor, *A. squarrosa*, than in the representative of A and possibly B genome donors, *T. monococcum* and *A. sharonensis*, *A. longissima* and *A. speltoides* respectively. The nature of response to VAM in hexapoloid wheat was controlled by factors of the A and B genomes, which are epistatic over those located at the D genome. The high mycorrhizal colonization and dependency that were found in *T. limopheevii* var. *araraticum* may indicate special genomic affinity possessed by the G genome of wheat interaction. Based on the study of 27 wheat lines and species, only low correlation between *G. intraradices* colonization and its contribution to plant growth can be suggested (Kapulnik and Kushnir 1991).

In studies conducted at the Soil Microbiology Department, Estacion Experimental del Zaidin, Granada, Spain, wheat cultivars inoculated with *Glomus mosseae* showed different degrees of mycorrhizal infection and mycorrhizal dependency. Mycorrhizal dependency was affected by root and root/shoot ratio and dry weight, but neither mycorrhizal dependency nor mycorrhizal infection levels were directly affected by nitrogen, phosphorus, potassium, calcium, and magnesium concentrations in plant tissues. Absence of mycorrhizal infection in some wheat varieties was associated with lack of sugar exudation from the root rather than the sugar content of the root (Azcon and Ocampo 1981).

In studies conducted at the Department of Plant Pathology and Department of Agronomy, the Pennysylvania State University, USA, two wheat (*Triticum aestivum*) cultivars (Laura, Neepawa) were grown in soil with phosphorus at four levels (0, 5, 10, 20 ppm) and inoculated with *Glomus clarum* NT4 to provide an inoculant:indigenous VAM ratio of 1:100. NT4 inoculant generally had no positive effect on any of the parameters assessed for wheat cv. Laura at any phosphorus level at 48 or 95 days after planting. Similarly, there was no positive effect of NT4 on shoot or root growth or AMF colonization of wheat cv. Neepawa at any phosphorus level at 48 days after planting. However, NT4 inoculation increased the growth yield of Neepawa by 20% ($p = 0.05$) when fertilized with 20 mg P/kg soil. The yield increase was associated with a significant ($p = 0.05$) reduction in root biomass and a significant ($p = 0.05$) increase in the grain phosphorus content of inoculated plants (Xavier and Geimida 1997).

Studies conducted at the Institute fur Planzenbau und Tierhygiene in den Tropen und Subtropen Grisebachstr, Gottingen, Germany, showed that under phosphorus deficiency conditions, the VA mycorrhizal fungal (*Glomus manihotes*) inoculation showed a higher efficiency in growth response with most of the wild wheat forms and landraces than with high yielding varieties (Manske 1990).

Studies conducted at the Station d'Amelioration des Plantes, INRA, Dijon, Cedex, France, on 20 wheat (*Triticum aestivum*) cultivars showed that the responses to inoculation with *Glomus mosseae* varied greatly from important yield increases to significant yield depressions; certain cultivars apparently being unaffected by the infection. Large variation in infection development were also observed, but no correlation existed between these and yield response. However, in cultivars where mycorrhizal infection improved yield, root/shoot ratios were consistently reduced. When infection levels, which were generally low, were increased by changes in culture conditions, mycorrhizal growth responses decreased (Gianinazzi, Bertheau, Gianianazzi-Pearson et al. 1979).

Vasccinium corymbosum

Studies conducted at the Warsaw Agricultural University, Poland, on five highbush blueberry (*Vaccinium corymbosum*) cultivars (Berkeley, Bluecrop, Darrow, Goldtraube and Herbert) at two locations showed that in all the cultivars studied, an average of 19% of cortical root cells were mycorrhizal (14.9% in Location 1 and 23.1% in Location 2). Significant differences in the infection level were observed among cultivars derived from the two soils, and, to a lesser extent, among plants within one cultivar (Czesnik and Eynard 1990).

Vigna unguiculata

In studies conducted at the Agronomy Department, Clemson University, Clemson, USA, cowpea (*Vigna unguiculata*) cultivars were grown on soil fumigated with methyl bromide and with or without inoculation with *Glomus etunicatum*, *G. claroideum*, native VAM fungi, and *Bradyrhizobium* strain 32 HP. At lower phosphorus levels, cultivar Katumane K80 gave better results with *G. etunicatum* than with *G. claroideum* whereas California No.5 Black Eye gave better results with *G. claroideum* than with *G. etunicatum*. At higher phosphorus levels, results of both cultivars were comparable (Obura, Skipper, and Wagner 1987).

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Research findings

Vesicular–arbuscular mycorrhizal association with some medicinal plants growing on alkaline soils of Mainpuri district, Uttar Pradesh

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Introduction

VAM (vesicular–arbuscular mycorrhizal) fungi are a major component of the soil microbial community, forming symbiotic associations with roots of over 90% of the terrestrial plants. VA mycorrhizae have gained significance because of their role in soil fertility, nutrient uptake, biocontrol of plant diseases, and growth of plant species used in afforestation programmes (Bakshi 1974; Byrareddy and Bhagyaraj 1988; Thapar and Khan 1973). Members of Endogonaceae are widely distributed in agricultural and forest soils worldwide. Over 81% of the 89 shrub species in arid and semi-arid areas of the world are endomycorrhizal (Lindsey 1984). VAM fungi are not yet routinely applied in commercial agriculture, forestry, or land reclamation, although commercial development is in progress (Menge and Timmer, 1982). Isolation and multiplication of alkalinity-tolerant efficient strains of VAM fungi are important for reclamation of marginal, eroded, or degraded soils, which are frequently used for rehabilitation and reforestation. VAM fungal isolates tolerate high alkalinity or pH, low nutrient, and irrigation regimes in alkali soils. The present study was conducted to record the percentage of VAM infection on roots of some medicinal plants growing in the Mainpuri District, Uttar Pradesh.

Materials and methods

Experimental site

The Mainpuri district is located at a latitude of 27°14' North and longitude of 79°03' East, and has 8101.4 Mha of waste land in blocks of 37 ha or more. There is a marked diurnal temperature difference: the temperature can be as low as 4.2 °C in January and high as 45 °C in June. The annual rainfall is 50–75 cm. Climatic regions are semi-humid or humid. The alkali soil is composed of a deep layer of stiff, heavy, and poorly aerated clay devoid of humus and containing large amounts of salts, injurious to all types of vegetation; the surface of soil is covered with a hard, compact crust having a dirty-white or dark-brown colour and infertile due to the presence of alkali salts in the subsoil.

Soil analysis

Soil samples were collected randomly from 10- to 30-cm-deep pits from usar land of the Mainpuri district. The soil collected from different sites was analysed for pH, EC (electrical conductivity) texture, SAR (sodium absorption ratio) organic matter and exchangeable sodium (Table 1).

Estimation of VAM infection

Plants were collected along with the rhizosphere soil and roots were washed and cleared in 10% KOH at 90 °C for 1 h. Cleared roots were acidified with 5 N HCl and stained with 0.5% trypan blue (Phillips and Hayman 1970), mounted in lactophenol, and the per cent colonization was calculated. The endogonaceous spores were isolated from the rhizospheric soil of all plants by using the wet sieving and decanting technique described by Gerdemen and Nicolson (1963).

Table 1 Physiochemical characteristics of alkali soil in Mainpuri

Parameter	*Value*
pH	9.0– 0.4
Electrical conductivity (mmhos/cm)	20.1–10.4
Texture (sand: silt: clay)	62:24:14
Exchange sodium (%)	80–88
SAR	830–950
$CaCO_3$ (%)	3
Total porosity (%)	35.9–40.6
Non-capillary porosity (%)	2.4–5.9
Intrinsic permeability (%)	0.01–0.08

Results and discussion

All medicinal plants were grown at high pH and were tolerant to alkali. From Table 2, it is clear that all the medicinal plants under investigation were infected by VAM fungi. However, the degree of infection varied with plant species and place of collection.

It is well known that mycorrhizal infection is heaviest on infertile soil. Low levels of nitrogen or phosphorus can themselves lead to an increased intensity of infection (Mosse 1973). Most plant species are typically mycorrhizal, with approximately four-fifth of all land plants forming VAM (Malloch, Pirozynski, and Raven 1980). In total, in the present studies, 15 plant species were positive to VAM fungi infection. Highly responsive species were *Cassia fistula* (94%), *Cassia alata* (69%), *Leucaena leucophala* (100%), and *Convoluvlus arvensis* (100%). High level of colonization indicated a deep susceptibility towards VAM fungi and the infectiveness of VAM fungi. All plants were observed in the young stage. The effect of VAM fungi could not be reflected in the form of growth responses of the host, but the efficiency of VAM fungi through a high rate of colonization was indicated. Therefore, the study shows that effective VAM fungi are not enough to generalize the endophyte species composition in alkali soils because the endophyte species composition is known to change with time and crop (Schenck and Kinloch 1980). Thus, extensive sampling over a long period is required to determine species diversity (Walker et al. 1982). It is apparent from these results that in alkali soil, it is possible that VAM infection increases the capacity of plants to accumulate phosphate as it is released by other microorganisms.

The present study reveals that all medicinal plants growing on alkali soils showed mycorrhizal infection. Hence, further identification of the natural endophytes and their application after thorough screening would be rewarding to rehabilitate or reclaim alkali soils.

Table 2 Per cent colonization of vesicular–arbuscular mycorrhiza in medicinal plants

Scientific name	*Family*	*Per cent VAM colonization*	*Drugs obtained from*	*Medicinal uses*
Cassia fistula	Caesalpinaceae	94	Leaf	Throat, respiratory, liver and eye diseases
Cassia alata	Caesalpinaceae	69	Leaf	Heart diseases
Cassia occidentalis	Caesalpinaceae	40	Flower, fruit, and seed	Rheumatism, leprosy
Emblica officinalis	Euphorbiaceae	45	Fruit	Good laxative and diuretic
Azadirachta indica	Amaranthaceae	28	Leaf	Ulcers and eczema
Amaranthus spinosus	Amaranthaceae	8.76	Root	Astringent, diuretic
Achyranthus aspera	Amaranthaceae	14	Root	Pyorrhea, cough, and fever
Datura stramorium	Solanaceae	10	Leaf	Asthma and bronchitis
Leucaena leucocephala	Mimosaceae	100	Flower, fruit, and seed	Tonic and anthelmintic
Adhatoda vasica	*Acanthaceae*	*50*	Leaf	Leprosy, tumours, bronchitis, and asthma
Eucalyptus globulus	Myrtaceae	30	Leaf	Removes intestinal parasites and nasal congestion
Aegle marmelos	Rutaceae	14	Fruit	Diarrhoea, dysentry
Calotropis procera	Asclepidaceae	20.3	Leaf	Sore throat, cough, and abdominal colics
Pongamia pinnata	Papilionaceae	50	Flower, fruit, and seed	Abdominal colics
Ipomoea paniculata	Convolvulaceae	45.3	Root	Purgative

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Arbuscular mycorrhizal association of Kashini (*Cichorium intybus* L.) in relation to physico-chemical characters

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Introduction

The ecology of rhizosphere microflora and their effects on plant growth have gained much interest in the recent past. Among the microflora colonizing the rhizosphere, AM (arbuscular mycorrhizal) fungi occupy a unique ecological position as they are partly inside and partly outside the root. The fungi are invariably present in practically all soils in association with a variety of plants of different taxonomic groups (Mosse 1981) and help the plants in the uptake of phosphorus and other nutrients and increase plant growth. The impact of soil factors on AM fungi has been studied earlier from temperate regions (Azcon and Ocampo 1981; Dubey and Negi 1995). However, there is no single report on AM association of Kashini (*Cichorium intybus* L.) in relation to soil characters. Kashini is an important essential medicinal plant commercially cultivated in the Vellore district of Tamil Nadu, India. The cultivated plant is used in Indian medicine as a toxic. The roots and leaves are used to treat stomachic, diuretic, fever, and vomiting, to enrich and purify the blood, and to reduce pain in the joints. Therefore, the present investigation was carried out to study the type of mycorrhizal association and scan the AM fungal population from the rhizosphere of Kashini in relation to soil characters.

Materials and methods

The survey were conducted to collect root and rhizophere soil samples of Kashini (*Cichorium intybus* L.) from 10 localities of the Vellore district of Tamil Nadu, India (Table 1). Initially, attempts were made to detect the type of mycorrhizal association by examining root sections and cleared root samples by adopting the method given by Phillips and Hayman (1970). Per cent root colonization was determined based on the number of root segments colonized by AM fungi. AM fungal propagules were isolated from the rhizosphere soils collected from different sites by using the wet sieving and decanting technique (Gerdemann and Nicolson 1963). The number of AM propagules present in 100 g dry weight of the soil was calculated. The species-level identification of different fungi was done following the keys provided by

Table 1 Status of arbuscular fungi and physico-chemical characteristics of the rhizosphere soils of Kashini[a] from 10 sites

Factor	*Vellore*	*Tirupathur*	*Vaniyambadi*	*Ambur*	*Ramapuram*	*Ranipet*	*Amabalur*	*Nattrampalli*	*Latheri*	*Alangayam*
Number of AM fungal propagules per 100 g rhizosphere soil	382	260	162	73	185	85	145	232	90	210
AM fungal root colonization (%)	92	84	64	42	72	53	58	70	50	72
Soil type	Sandy loam	Black sandy loam	Sandy	Sandy clay loam	Sandy	Clay loam	Sandy loam	Sandy loam	Clay	Sandy loam
Soil pH	7.2	7.4	7.2	6.1	7.4	6.2	8.2	7.2	6.8	7.8
Moisture (%)	8	10	12	15	9	14	8	10	5	9
Organic matter (%)	0.92	0.85	1.53	1.81	1.23	1.68	0.86	1.42	1.32	0.96
Available nitrogen (mg/kg)	460	650	200	620	420	340	640	800	400	350
Available phosphorus (mg/kg)	1.2	1.8	1.6	3.2	1.6	3.6	1.2	1.9	1.6	2.0
Available potassium (mg/kg)	140	150	220	580	310	240	420	340	160	160
Copper (mg/g)	1.1	1.2	1.8	0.8	1.6	2.1	1.1	1.2	1.9	1.2
Zinc (mg/g)	1.2	1.9	1.2	3.1	1.6	3.8	2.2	2.4	2.1	1.8
Manganese (mg/g)	3.2	4.6	3.4	2.6	1.8	4.2	3.2	1.8	2.4	3.2
Iron (mg/g)	60.2	51.5	61.4	72.2	62.1	99.6	32.2	41.2	51.1	61.2

[a] The values represent mean of three replicates

Trappe (1982) and Schneck and Perez (1990). Various physico-chemical characters – soil type, total nitrogen, available phosphorus, copper, and zinc – were estimated following standard methods (Issac and Kerber 1971; Jackson 1973; Piper 1974; Walkley and Black 1934). Soil pH was measured by taking the extract of soil–water mixture (1:5 w/v).

Results and discussion

Table 1 presents the per cent root colonization, soil spore population of AM fungi, and physico-chemical characters of Kashini at 10 different sites. It revealed that the 10 different samples had extensive roots colonization and spores in soil, but were varied in distribution. A higher per cent colonization and spore number of AM fungi were recorded in sandy loam P-deficient soil with neutral to alkaline soil pH (7.2). It was observed that maximum per cent colonization and AM spores were found in the soil samples collected from Vellore (92% and 382) and Tirupathur (84% and 260). The least number of spores and per cent colonization was recorded in the samples collected from Ambur (42% and 73) where the soil was sandy clay loam, high in phosphorus, and had acidic pH (6.1). In the present study, soils were in the pH range of 6.1–8.2. The soils under study, with a phosphorus content between 1.2 and 3.6 mg/kg, have supported more AM fungal propagules and species than have soils with a high phosphorus content and other nutrients. Similar results have been were obtained earlier by Janaki Rani and Manoharachary (1994) and Bhaskaran and Selvaraj (1997).

The frequency distribution of different AM fungi isolated from the rhizosphere of Kashini plants in 10 sites is shown in Table 2. Almost all the soil samples contained AM spores of different species of the genera *Acaulospora*, *Glomus*, *Gigaspora*, *Sclerocystis*, and *Scutellospora*. A total of 11 different species of AM fungi were collected and identified. *Glomus* was the dominant genus and was represented by five species, followed by *Selerocystis* with three species, and *Acaulospora*, *Gigaspora,* and *Scutellospora* each represented by one species (Table 2). *Glomus aggregatum* was found to be the most frequent fungus (100%), followed by *Sclerocystic pakistanika* (80%) and *Gigaspora margarita* (70%). The genera *Acaulospora* and *Scutellospora* were represented by a single species only in acidic soils. Distribution of various AM fungal species may be influenced by soil pH (Mosse 1981). Some *Glomus* species are very common in neutral or alkaline soils but not in acid soils whereas species of *Acaulospora* and *Scutellospora* are usually reported in acid soils. In the present study, a similar relationship was found between soil types, pH, and a particular endophyte species. Further, the AM fungi in the present study lacked host specificity. The Kashini plant is found to harbour spores of different AM fungi such as *Glomus*, *Acaulospora*, *Gigaspsora*, *Sclerocystis*, and *Scutellospora*.

Earlier studies by the authors have shown variation in per cent colonization in roots and AM spores in the rhizosphere soils of various medicinal plants under different sites/soil conditions (Gupta and Janardhanan 1991). The intensity of AM colonization in roots and spores in the rhizosphere of Kashini in the 10 sites varied according to the physico-chemical characters of the site, including soil pH, soil type, soil nutrients, and other environmental conditions. *Glomus aggregatum*, *Sclerocystis pakistanica*, and *Gigaspora margarita* are widespread and consistent, which makes them most favourable for mass multiplication. The application of these AM fungi can be useful to increase the production

Table 2 Frequency distribution of AM fungi in rhizophere soil samples of Kashini from 10 sites

Fungus	*Vellore*	*Tirupathur*	*Vaniyambadi*	*Ambur*	*Ramapuram*	*Ranipet*	*Amabalur*	*Nattrampalli*	*Latheri*	*Alangayam*	*Site frequency*
Acaulospora scrobiculata											
Trappe	–	–	–	+	–	+	–	–	–	–	20
Gigaspora margarita											
Becker and Hall	+	+	+	–	+	–	+	+	–	+	70
Glomus aggregatum											
Schenck and Smith	+	+	+	+	+	+	+	+	+	+	100
Glomus botryoides											
Rothwell and Victor	–	+	+	–	+	–	–	–	–	–	30
Glomus deserticola											
Trappe, Bloss and Menge	+	+	–	–	+	–	+	–	–	–	40
Glomus geosporum											
(Nicol & Gerd) Walker	+	+	+	-	–	–	+	–	–	–	40
Glomus mosseae											
(Nico & Gerd) Gerd and Trappe	+	+	+	+	–	–	+	–	–	–	50
Sclerocystis pachycaulis											
Wu and Chen	+	+	+	–	+	–	–	–	+	–	50
Sclerocystis pakistanica											
Iqbal and Bushra	+	+	+	–	+	–	+	+	+	+	80
Sclerocystis sinuosa											
Gerd and Bakshi	+	–	+	–	–	–	–	+	–	–	30
Scutellospora calospora (Nicol & Gerde)											
Walker and Sanders	–	–	–	+	–	+	–	–	–	–	20

of this essential medicinal plant. The present observation is also significant for economics of commercial cultivation of medicinal plants.

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Occurrence of arbuscular mycorrhizae on wasteland weeds in eastern Uttar Pradesh: a preliminary survey report

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Introduction

More than 10 million hectares of land in India is salt-affected. The physical, chemical, and biological properties of such soils have converted them into wastelands. There is a vast stretch of such lands spreading to various districts of eastern Uttar Pradesh. These soils have been recorded to have high pH (9.2–10.9), extremely high ESP (exchangeable sodium percentage) (45–90), low permeability, and high electrical conductivity (5–15 dSm^{-1}) (data from Department of Soil, Narendra Dev University of Agriculture and Technology). Although wastelands are unsuitable for agriculture, a variety of weeds have adapted themselves to this condition. *Argemone mexicana*, *Calotropis procera*, *Croton sparsoflorus*, *Cynodon dactylon*, *Datura stramonium*, *Imperata cylindrica*, *Ipomoea cornea*, *Opuntia* sp., *Parthenium hysterosphorous*, *Saccharum spontaneum*, and *Xanthium strumarium* are some of the commonly growing weeds in the wastelands of Allahabad, Faizabad, Jaunpuri, Pratapgarh, Rae-Bareilly, and Sultanpur districts of eastern Uttar Pradesh. In view of shrinking of agricultural lands and growth of human population at an alarming rate, it is being emphasized to reclaim such wastelands for making them useful for forestry and agriculture.

Mycorrhiza play an important role in establishment and survival of plant communities in hostile soils of such lands (Smith and Read 1997). Elucidation of a predominant AM (arbuscular mycorrhizal) fungi, if any, associated with these weeds might be helpful in devising an indigenous reclamation strategy based on microbes (mycorrhizal); till recently, reclamation strategies have been solely chemical (gypsum/pyrite) based. There is little information available on the nature and extent of occurrence of AM on wasteland weeds in eastern Uttar Pradesh. Hence, a survey of weeds in some of the salt-affected districts of eastern Uttar Pradesh was carried out, and the degree of mycorrhization, occurrence, and spore density of AM fungi is reported here.

Materials and methods

Soil and root samples were collected from 11 commonly occurring weeds in six districts of eastern Uttar Pradesh. Roots were properly washed in running tap water and cleared in a near-boiling 10% aqueous solution of KOH for 48 h. Roots were stained in Trypan blue following several washings in distilled water to drain out the KOH (Phillips and Hayman 1970). The stained roots were cut into 1-cm segments, and 80–100 such segments were picked up and examined under the microscope. AM colonization was determined by Nicoloson's formula (1995) as follows.

$$\text{Percent colonization} = \frac{\text{Number of root segments colonized}}{\text{Total number of segments examined}} \times 100$$

The spore density of AM fungi was quantified in 1 kg of soil following the wet sieving and decanting technique of Gerdemann and Nicolson (1963) and the frequency of occurrence was calculated by Norton's formula (1978) as follows.

$$\text{Absolute frequency} = \frac{\text{Number of samples containing a species}}{\text{Total number of segments examined}} \times 100$$

$$\text{Relative frequency} = \frac{\text{Frequency of a species}}{\text{Sum of frequencies of all the species}} \times 100$$

The identification of AM fungi was based on taxonomic keys prepared by Hall (1984), Schenck and Perez (1990), and Mehrotra and Baijal (1994).

Results and discussion

The weeds surveyed in the wastelands of Allahabad, Faizabad, Jaunpur, Pratapgarh, Rae-Bareilly, and Sultanpur districts were found to be colonized by AM fungi to a varying extent (0%–80%). (Table 1). The degree of mycorrhization appears to be influenced by the weed species rather than the location. The highest degree of mycorrhization occurred on *Opuntia* sp. (50%–61%), followed by *Saccharum spontaneum* (35%–59%), *Cynodon dactylon* (30%–46%), and *Ipomoea cornea* (30%–49%). *Croton sparsiflorus* was the least colonized (17%–33%). The decreased amount of mycorrhizal colonization of roots of various plants has been correlated with increased soil salinity (Duke, Johnson, and Koch 1986; Poss, Pond, Menge et al. 1985). The growth and establishment of *Aster tripolium* and *Parthenium argentatum* was correlated with decreased concentration of Na and increased concentration of phosphate ions in mycorrizal plants (Pfeiffer and Bloss 1988; Rozema, Arp, Diggelen et al. 1986).

The spore density of mycorrhizal fungi varied from 0 to 510 per 100 g of soil; however, average values ranged from 66 to 405 around various weeds in different districts (Table 1). The spore density, in general, around *Opuntia* sp., *Cynodon dactylon*,

Table 1 Degree of mycorrhization and soil spore density of arbuscular mycorrhizal fungi on some wasteland weeds in six districts of eastern Uttar Pradesh

		Districts					
Weed species	*Factors*	*Allahabad*	*Faizabad*	*Jaunpur*	*Pratapgarh*	*Rae-Bareilly*	*Sultanpur*
Argemone mexicana	Mycorrhizal colonization (%)						
	Range	8–38	10–57	7–53	10–80	8–65	0–60
	Mean + SD	22 + 11	31 + 61	37 + 15	36 + 22	33 + 17	32 + 21
	Spore density (number/100 g soil)						
	Range	80–360	50–250	50–300	30–301	40–285	0–350
	Mean + SD	220 + 99	149 + 65	189 + 78	162 + 84	154 + 73	172 + 105
	Mycorrhizal fungus	Gf, Gm	Gig, G, Gc, Gm	A, Gim, Gm	Gim, G, Gm	G1, Gim, Gm	Gig, Gf, Gm
	N	8	10	12	12	15	12
Calotropis procera	Mycorrhizal colonization (%)						
	Range	8–59	8–60	20–75	0–49	10–60	10–55
	Mean + SD	33 + 16	37 + 19	43 + 18	26 + 16	34 + 17	32 + 16
	Spore density (number/100 g soil)						
	Range	180–395	50–350	80–400	58–302	65–300	55–264
	Mean + SD	282 + 74	187 + 97	232 + 101	158 + 78	194 + 73	185 + 84
	Mycorrhizal fungus	Gi, Gim	Gim, G, Gf	Gig, G	Gim, Gc, Gf	As, Gig, Gc	G, Gc, S
	N	10	10	9	10	12	10
Cronton sparsiflorus	Mycorrhizal colonization (%)						
	Range	0–36	15–50	14–56	15–44	10–40	18–50
	Mean + SD	17 + 13	27 + 13	33 + 19	31 + 11	24 + 10	30 + 10
	Spore density (number/100 g soil)						
	Range	115–320	80–290	0–270	50–137	55–228	55–280
	Mean + SD	204 + 73	140 + 73	129 + 84	90 + 32	131 + 70	135 + 76
	Mycorrhizal fungus	Ga,Gc	Ga, Gm	G, Gm	A, Ga, Gm	Gig	Gig, Gm
	N	7	9	10	7	7	8
Cynodon dactylon	Mycorrhizal colonization (%)						
	Range	10–66	18–68	12–75	10–75	28–77	8–55
	Mean + SD	36 + 17	41 + 14	47 + 17	39 + 20	46 + 17	30 + 15
	Spore density (number/ 100 g soil)						
	Range	185–500	85–380	77–333	48–250	55–321	58–506
	Mean + SD	356 + 106	219 + 89	183 + 85	139 + 81	190 + 83	222 + 138
	Mycorrhizal fungus	Gf, Gm, Gig, Gim	Ga, Gm	Gim, Gf	Gig, G	Gig, Gm	Gig, Gm
	N	15	16	15	15	13	16
Datura stramonium	Mycorrhizal colonization (%)						
	Range	7–37	8–48	0–60	10–70	8–50	8–62
	Mean + SD	21 + 10	27 + 13	32 + 20	34 + 18	30 + 14	33 + 17
	Spore density (number/ 100 g soil)						
	Range	120+360	5–300	50–210	30–210	87–240	50–210
	Mean + SD	214 + 77	155 + 103	139 + 60	110 + 54	151 + 55	143 + 55
	Mycorrhizal fungus	As, Gf, Gig	A, Gm	Gi, Gig	Gm	As, Gm, S	A, Gf, S
	N	10	11	15	12	8	10
Impereta cylindrica	Mycorrhizal colonization (%)						
	Range	18–39	35–68	12–44	8–43	10–50	22–62
	Mean + SD	27 + 9	51 + 13	39 + 17	29 + 15	23 + 15	44 + 18
	Spore density (number/ 100 g soil)						
	Range	80–460	90–450	60–280	59–265	95–280	80–280
	Mean + SD	306 + 158	296 + 127	163 + 86	162 + 68	157 + 77	194 + 85
	Mycorrhizal fungus	Gig, S	Gi, Gim, Gf	As, Gf	Gf, S	Gig, Gm	Gim, Gm, S
	N	5	6	5	8	6	5
Ipomoea cornea	Mycorrhizal colonization (%)						
	Range	19–44	22–77	18–68	16–57	8–50	18–50
	Mean + SD	31 **+ 10**	49 + 20	39 + 17	39 + 14	33 + 15	30 + 11
	Spore density (number/ 100 g soil)						
	Range	100–510	77–315	55–317	39–148	58–239	70–320
	Mean + SD	292 + 161	198 + 95	196 + 104	76 + 41	141 + 71	182 + 79
	Mycorrhizal fungus	Gim, Gig	Gm, S	Gim,Gm, S	Gig, Gf	A, Gm	A, G, Gc
	N	6	8	8	8	7	8

Table 1 continued

Weed species	Factors	Districts					
		Allahabad	*Faizabad*	*Jaunpur*	*Pratapgarh*	*Rae-Bareilly*	*Sultanpur*
Opuntia sp.	Mycorrhizal colonization (%)						
	Range	21–78	21–80	27–77	30–70	30–75	47–85
	Mean + SD	50 + 22	59 + 19	53 + 19	55 + 15	55 + 12	61 + 10
	Spore density (umber/ 100 g soil)						
	Range	210–510	90–430	90–425	90–450	70–310	155–465
	Mean + SD	405 + 110	248 + 112	235 + 123	260 + 159	168 + 90	290 + 119
	Mycorrhizal fungus	Gf, Gig, S	Gim, Gf	Gig, Gf	Gc, Gf	Gim, Gf, S	A, Gim, Gf
	N	7	8	7	5	8	8
Parthenium hysterophorus	Mycorrhizal colonization (%)						
	Range	0–62	0–50	0–58	0–55	8–60	8–55
	Mean + SD	25 + 19	27 + 15	29+16	30 + 16	23 + 15	28 + 16
	Spore density (number/100 g soil)						
	Range	80–330	50–300	0–300	0–175	80–305	62–315
	Mean + SD	200 + 87	177 + 89	169 + 89	66 + 53	172 + 73	187 + 86
	Mycorrhizal fungus	G, Gm, Gf, Gim	Gim, Gf	Gf, S	Gim, Gm, S	Gi, Gim, Gm	Gig, G, Gm
	N	10	13	12	16	10	9
Saccharum spontaneum	Mycorrhizal colonization (%)						
	Range	18–77	30–75	32–80	16–60	15–70	18–60
	Mean + SD	50 + 21	59 + 14	53 + 15	43 + 15	44 + 20	35 + 16
	Spore density (number/100 g soil)						
	Range	200–515	90–500	68–376	8–204	70–330	66–275
	Mean + SD	369 + 101	309 + 156	202 + 106	87 + 75	218 + 98	154 + 77
	Mycorrhizal fungus	Gi, Gig, Gm	Gig, Gm	Gig, Gm	Gf, S	Gig, Gf, Gm	A, Gig, Gm
	N	10	8	9	8	8	8
Xanthium strucmarium	Mycorrhizal colonization (%)						
	Range	8–38	0–50	10–58	8–56	10–52	14–62
	Mean + SD	23 + 11	27 + 14	33 + 14	27 + 15	31 + 15	31 + 17
	Spore density (number/100 g soil)						
	Range	110–300	70–300	75–345	16–280	58–300	28–260
	Mean + SD	201 + 69	175 + 81	175 + 90	113 + 82	175 + 68	129 + 77
	Mycorrhizal fungus	A, Gig, Gf	As, Gi, Gig, Gf	Gim, G, Gf	Gim, G, Gm	As, G, Gf	As, G, Gf
	N	8	10	15	10	12	8

Note SD: standard deviation; *N*: number of samples analysed; A: *Acaulospora* sp.; As: *Acaulospora spinosa*; Gi: *Gigaspora* species; Gig: *Gigaspora gigantea*; Gim: *Gigaspora margarita*; G: *Glomus* sp.; Ga: *Glomus aggregatum*; Gf: *Glomus fasciculatum*; Gc: *Glomus constrictum*; Gm: *Glomus mosseae*

and *Saccharum spontaneum* was higher than that around other weed species. In the absence of data generated under controlled conditions that simulate the physico-chemical characteristics of wasteland soils of these districts, it is difficult to correlate spore density with any soil factor. However, alkalinity and the higher proportion of Na ions have been implicated for reduced colonization and low spore count (Juniper and Abbott 1993, Smith and Read 1997).

The survey revealed that *Acaulospora* sp. (*A. spinosa)*, *Gigaspora* spp. (*G. margarita*, *G gigante*, *Glomus* spp. (*G. aggregatum*, *G. constrictum*, *G fasciculatum*, *G. mosseae*) and *Sclerocystis* sp. were the AM fungi occurring in these wastelands (Table 1). *Glomus* appeared to be the most dominant genus followed by *Gigaspora*, *Acaulospora*, and *Sclerocystis* (Table 2). Of various species belonging to these genera, *Acaulospora spinosa*, *Gigaspora margarita,* and *Glomus fasciculatum* occurred more frequently than other species (Table 3).

Mycorrhizal fungi have been shown to occur naturally in saline soils by several workers (Ho 1987; Khan 1974; Rozema, Arp, Diggelen et al. 1986). From the preliminary results of the present investigation, it is evident that *Glomus fasciculatum* is well adapted to the wastelands of the districts under study, and this can be used to develop a long-term reclamation protocol for such soils.

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Table 2 Relative frequency (%) of occurrence of various mycorrhizal fungus genera in wastelands of six districts of eastern Uttar Pradesh

Mycorrhizal fungus	*Allahabad*	*Faizabad*	*Jaunpur*	*Pratapgarh*	*Rae-Bareilly*	*Sultanpur*
Acaulospora spp.	9.2	9.7	8.4	8.9	9.2	11.4
Gigaspora spp.	33.9	30.7	35.9	36.3	33.1	34.9
Glomous spp.	53.2	53.0	50.3	51.1	52.3	48.6
Sclerocystis spp.	3.7	6.6	5.3	3.7	5.3	5.1

Table 3 Absolute frequency of occurrence of various arbuscular mycorrhizal fungi in wastelands of six districts of eastern Uttar Pradesh

AM fungi	*Allahabad*	*Faizabad*	*Jaunpur*	*Pratapgarh*	*Rae-Bareilly*	*Sultanpur*
Acaulospora spp.	15.0	20.0	15.0	17.5	20.0	26.0
	(5.5)[a]	(5.3)	(4.6)	(5.2)	(5.3)	(7.4)
A. spineas	10.0	16.7	12.5	12.5	15.0	14.0
	(3.7)	(4.4)	(3.8)	(3.7)	(3.9)	(4.0)
Gigaspora spp.	17.5	25.0	22.5	22.5	30.0	22.0
	(6.4)	(6.6)	(6.9)	(6.7)	(7.9)	(6.3)
G. margarita	42.5	50.0	55.0	62.5	60.0	58.0
	(15.6)	(13.2)	(16.8)	(18.5)	(15.9)	(16.6)
G. gigantea	32.5	41.7	40.0	37.5	35.0	42.0
	(11.9)	(10.9)	(12.2)	(11.1)	(9.3)	(12.0)
Glomus spp.	20.0	33.3	15.0	20.0	27.5	20.0
	(7.3)	(8.8)	(4.6)	(5.9)	(7.3)	(5.7)
G. aggregatum	22.5	26.7	20.0	25.0	20.0	16.0
	(8.3)	(7.0)	(6.1)	(7.4)	(5.3)	(4.6)
G. constrictum	12.5	16.7	25.0	20.0	22.5	22.0
	(4.6)	(4.4)	(7.6)	(5.9)	(7.3)	(6.3)
G. fascicultaum	50.0	58.3	62.5	47.5	52.5	42.0
	(18.3)	(15.3)	(19.1)	(14.1)	(13.9)	(12.0)
G. mosseae	40.0	66.7	42.5	60.0	75.0	70.0
	(14.7)	(17.5)	(12.9)	(17.8)	(19.9)	(20.0)
Sclerocystic spp.	10.0	25.0	17.5	12.5	20.0	18.0
	(3.7)	(6.6)	(5.3)	(3.7)	(5.3)	(5.1)
Number of samples analysed	40	60	40	40	40	50

[a] Values in parenthesis are relative frequencies

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VAM infection in *Brassica oleracea* var. *capitata* – a member of Cruciferae

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Introduction

VAM (vesicular–arbuscular mycorrhizal) association is geographically ubiquitous and occurs over a broad ecological range (Gerdeman 1968). In natural communities, approximately 70% of the higher plants are obligatorily dependent on fungal associates, 12% facultatively dependent, and 18% typically non-mycorrhizal (Trappe 1987; Mohan and Subashini 1995). A lot of interest has been developed in the study of VAM in the recent years because of their role in improving plant growth, controlling root pathogens, increasing in biological nitrogen fixation, and improving the ability of plants to withstand water stress, salinity, and heavy metal toxicity (Harley and Smith 1983; Verma and Jamaluddin 1994). Families such as Chenopodiaceae, Fumariaceae, Polygonaceae, Proteaceae, Cyperaceae, Urticeae, Cruciferae, and Commelinacae are widely thought to be non-mycorrhizal (Harley and Harley 1987; Ozinga, van Andel and McDonnell 1997). The present work reports for the first time the occurrence of VAM on *Brassica oleracea* var. *capitata* cv. Hercules of the family Cruciferae, a crop of commercial importance for subsistence farmers in the KwaZulu Natal region, South Africa. It also describes the growth-promoting activity of VAM fungus on the seedlings of the host as a preliminary observation.

Materials and methods

Three VAM species, namely *Glomus aggregatum, G. fasciculatum* and *G. mosseae*, were maintained on onion (*Allium cepa* L.) to prepare pot cultures following the procedure of Krishna, Shetty, Dart et al. (1985). Once the pot cultures were established, the soil from these pot cultures along with the roots of onion was used as a source of inoculum. Seeds of *Brassica oleracea* var. *capitata* cv. Hercules were surface-sterilized in 70% ethanol for 2 min, followed by 2 min in 0.6% mercuric chloride, rinsed three times in sterile distilled water, and sown in plastic pots containing potting mix and vermiculite (2:1). One week after germination, the seedlings were transplanted to plastic pots of diameter 13 cm containing 1 g of mycorrhizal inoculum (500 spores per gram of soil) of *G. aggregatum*, *G. fasciculatum*, or *G. mosseae*, distributed in one layer, 5 cm below the soil surface. The seedlings (24 per pot) were placed 2.5 cm above the mycorrhizal inoculum. Potted soil with no mycorrhizal inoculum served as control. All pots were arranged randomly in growth chambers, with a light intensity of 300 $\mu E \cdot m^{-2} s^{-2}$ (16th day) provided by cool-white fluorescent tubes at 16 °C/14 °C (day/night). Seedlings were watered with distilled water and fertilized every week with 50 ml of 10% Hoagland's nutrient solution (Hoagland and Arnon 1950) without phosphorus, beginning 30 days after transplanting. The experiment was conducted in triplicate.

Seedlings were raised for 60 days, following which VAM inoculated seedlings (three from each pot) were collected and the establishment of VAM in the roots were ensured (Phillips and Hayman 1970). Roots were also freeze-dried in liquid N_2, squashed, sputter-coated with gold particles, and observed under JEOL SEM (scanning electron

microscope) to confirm the establishment of VAM. The percentage of colonization was determined (Giovanetti and Morse 1980). The following growth parameters of both the VAM-inoculated and control seedlings were recorded: stem thickness, shoot and root length, and dry weight of shoots and roots. Phosphorus content of the roots and shoots was determined colourimetrically by the vandomolybdate method (Jackson 1971).

Results and discussion

Observations of the root squash and sections confirmed the establishment of VAM (Figure 1). Plant growth, biomass, and phosphorus uptake were enhanced by all the three species of VAM fungi. Maximum VAM colonization was observed with *G. fasciculatum* treatment (Table 1). However, there was difference in the growth-promoting efficiency of different VAM fungi. *G. fasciculatum* and *G. mosseae* were found to be more effective than *G. aggregatum* in increasing the growth rate, total biomass production, and phosphorus uptake. These differences can be attributed to the mechanism of mycorrhizal infection and development (Sanders, Tinker, and Black 1977), the physiological differences in rate of nutrient uptake between VAM endophytes, and the soil environment (Mosse 1973).

The increase in the phosphorus content in the VAM-inoculated seedlings may be due to the additional absorbing surface provided by the external hyphae of endomycorrhizal fungi, which enhances the ability of the plants to retrieve phosphorus (Perry, Bell, and Amaranthus 1992; Marschner and Dell 1994). A similar increase has also been reported earlier in several agricultural and forestry species (Reena and Bagyaraj 1990; Antones and Cardou 1991; Durga and Gupta 1995).

The effect of VAM fungi (*G. fasciculatum*, *G. aggregatum*, and *G. mosseae*) on the initial growth and establishment of *Brassica oleracea* var. *capitata* was studied. On the basis of the various growth parameters studied, it is concluded that the VAM-inoculated seedlings showed a greater growth rate than the uninoculated seedlings. Of the different species tested, *G. fasciculatum* was found to be more effective for improving plant biomass and seedling establishment, followed by *G. mosseae*. As cabbage cultivation is mainly done by the rural poor farmers in soils that are deficient in phosphorus, VAM symbiosis may be of particular significance in coping with phosphorus deficiency stress in such systems (Mc Arthur and Knowles 1993).

Table 1 Effect of different vesicular–arbuscular mycorrhizal fungi on plant growth, biomass, and phosphorus uptake in *Brassica oleracea* var. *capitata* cv. Hercules[a]

Treatment	VAM colonization (%)	Average stem thickness (mm)	Shoot length (cm)	Root length (cm)	Plant biomass (dry wt mg/plant)			Phosphorus content (mg/g dry wt)	
					Root	Shoot	Total	Root	Shoot
Control	0[a]	6 (2)[a]	8.6 (2.5)[a]	4.3 (1.6)[a]	83.5 (1.8)[a]	277.7 (2.8)[a]	361.2 (2.4)[a]	9.4 (2.5)[a]	19.6 (2.2)[a]
Glomus aggregatum	40.2 (0.57)[b]	9 (3)[b]	12.4 (2.2)[b]	5.4 (1.2)[ad]	113.6 (2.4)[b]	317.0 (2.1)[b]	430.6 (2.2)[b]	12.5 (1.9)[b]	24.7 (1.8)[b]
G. fasciculatum	74.2 (1.1)[c]	13 (3)[c]	19.4 (1.6)[c]	8.8 (2.1)[c]	160.4 (4.4)[c]	418.0 (3.2)[c]	578.4 (2.7)[c]	23.6 (3.6)[c]	51.2 (4.3)[c]
G. mosseae	46.1 (1.3)[c]	10 (2)[b]	14.3 (1.8)[b]	6.2 (1.4)[d]	142.0 (7.3)[d]	370.8 (1.7)[d]	512.8 (2.1)[d]	22.3 (2.0)[c]	39.4 (3.6)[d]

[a] Each value is a mean of five replicates. For each variate, any two values without common letters in their superscripts are significantly different by the Tukey–Kramer multiple range test ($p = 0.05$). Values in parenthesis indicate SE (standard error).

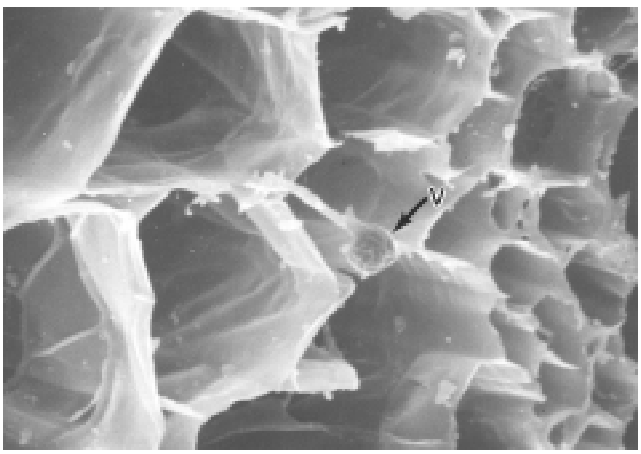

Figure 1 Scanning electron micrograph of the squashed root of *Brassica oleracea* var. *capitata* cv. Hercules, showing the vesicle (v) of *Glomus fasciculatum* in the cortex (× 650)

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New approaches

Quantification of AM fungal activity by glomalin concentration in hyphal traps

Strips of horticultural film (16–32 cm^2) were used by Wright S F and Upadhyaya A (1999) to trap extraradical hyphae emanating from roots of Sudan grass (*Sorghum sudanense*) enclosed in 40-mm mesh bags and colonized by *Gigaspora rosea*, *Glomus intraradice*, or *G. caledonium* (*Mycorrhiza* 8(5): 283–285). Strips of the film were placed at the opposite sides of 17–21 replicate sand culture pots for each isolate and were removed after 12–14 weeks of plant growth. To extract glomalin, a strip was cut into small pieces and submerged in 2 ml of 20-mM citrate, pH 7.0, and then autoclaved for 60 min. A quantitative ELISA (enzyme-linked immunosorbent assay) detected 0.005–0.04 mg glomalin in the volume of extract tested. The Bradford protein assay detected 1.25–5 mg protein in the volume of extract tested. Both assays gave results ranging from 5 mg to 40 mg glomalin/cm^2 of the film. Protein assay values were correlated with ELISA values ($r = 0.6091$, $p = 0.001$, $n = 118$). Analysis of variance indicated that isolates differed in Bradford protein values ($p = 0.001$) but not ELISA values ($p = 0.154$). Spatial variability of glomalin deposition ca. 7 cm from roots on opposite side of pots was indicated by a significant paired *T*-test ($p = 0.05$) for protein values for each of three isolates. ELISA *T*-test values were significant ($p = 0.003$) except for *G. caledonium* ($p = 0.13$). These results indicate that hyphal traps, Bradford protein assay, and ELISA are useful to assess hyphal activity over a growing season.

News from members

Dr Seema Bhadauria, Senior Lecturer, Department of Botany, RBS College, Agra has been honoured with the ASEA Excellence Award for her outstanding performance and lasting contribution in the field of environment by ASEA, Rishikesh, in the *National Conference on Status of Indian Environment* held at Rishikesh on 27–28 May 2001. The award included a memento, citation, and a cash award. She chaired a technical session and presented an invited lecture at the conference. A fellow award, FASEA, was also presented to her. A young scientist award (for researchers under 30 years of age) was awarded to Ms Ekta Bhadauria, Department of Botany, RBS College, Agra, for the best paper presentation. In the 30–34 years category, a gold medal for the best paper presentation was awarded to Dr Archana Singh, RBS College, Agra.

Centre for Mycorrhizal Culture Collection

The CMCC (Centre for Mycorrhizal Culture Collection) maintains a collection of mycorrhizal isolates obtained from different agro-ecological zones in India and different countries. The largest of its kind in Asia, the facility is well equipped to preserve the germplasm available in the country. The scientific community and industrial sectors can obtain high-quality, contaminant-free mycorrhizal cultures from this facility. The mycorrhizal cultures can be obtained on a nominal charge or through exchange. The major goal and mission of this programme is to maintain an efficient supply of cultures through increasing the diversity of its collection and continuous quality control. The inoculum supplied contains spores and other propagules unless specified. Most of these cultures are scientifically tested and screened for performance in different field conditions ranging from agricultural lands to disturbed soils where microbial activity is very low. In addition, the germplasm facility offers services such as characterization of strains and cultures on a payment basis. These include identification using image analysis programmes. The facility also encourages the deposit of cultures from the scientific community and others, which will be subsequently subjected to analysis and incorporated into the germplasm collection for circulation.

Recent references

Latest additions to the Network's database on mycorrhiza are published here for information of the members. The Mycorrhiza Network will be pleased to supply any of the available documents to bonafide researchers at a nominal charge.
This list consists of papers from the following journals, which are arranged in alphabetical order.

- *Mycologia*
- *Mycological Research*
- *Mycorrhiza*

Copies of papers published by mycorrhizologists during this quarter may please be sent to the Network for inclusion in the next issue.

Name of the author(s) and year of publication	Title of the article, name of the journal, volume no., issue no., page nos [*address of the corresponding author]
Aarple I M van *, Olsson P A, and Soderstrom B. 2001	**Microscopic detection of phosphate activity of saprophytic and arbuscular mycorrhizal fungi using a fluorogenic substrate** *Mycologia* **93**(1): 17–24 [*Department of Microbial Ecology, Ecology Building, Lund University, SE-223 62 Lund, Sweden]
Haeksworth David L *. 2001	**Mycological research news** *Mycological Research* **105**(2): 129–131 [*MycoNova, 114 Finchley Lane, Hendon, London, NW4 1DG, UK]
Ubalijoro E *, Hamel C, McClung C R, Smith D L. 2001	**Detection of chitin synthase class I and II type sequences in six different arbuscular mycorrhizal fungi and gene expression in *Glomus intraradices*** *Mycological Research* **105**(4): 470–476 [*Plant Science Department, McGill University, 21,111 Lakeshore, Ste-Anne-de-Bellevue, Qc, H9X 3V9, Canada]
Arocena J M*, Glowa K R, and Massicotte H B. 2001	**Calcium-rich hypha encrustations on *Piloderma*** *Mycorrhiza* **10**(5): 209–216 [*Forestry Program, Faculty of Natural Resources and Environmental Studies, University of Northern British Columbia, Prince George, BC , Canada V2N 4Z9]
Kernaghan G*. 2001	**Ectomycorrhizal fungi at tree line in the Canadian Rockies II. Identification of ectomycorrhizae by anatomy and PCR** *Mycorrhiza* **10**(5): 217–230 [*Groupe de recherche en ecologie forestiere, Universite du Quebec a Montreal, CP 8888 succ. A, Montreal, PQ, Canada H3C 3P8]
Fidelibus M W*, Martin C A, and Stutz J C. 2001	**Geographic isolates of *Glomus* increase root growth and whole-plant transpiration of citrus seedlings grown with high phosphorus** *Mycorrhiza* **10**(5): 231–236 [*Department of Plant Biology, Arizona State University, PO Box 871601, Tempe, AZ 85287-1601, USA]
Tagu D*, Rampant, PF, Lapeyrie F, Frey-Klett P, Vion P, Villar M. 2001	**Variation in the ability to form ectomycorrhizas in the F1 progeny of an interspecific poplar (*Populus* spp.) cross** *Mycorrhiza* **10**(5): 237–240 [*INRA -Nancy, Microbiologie Forestiere, 54280 Champenoux, France]
Oliveira R S, Dodd J C*, and Castro P M L. 2001	**The mycorrhizal status of *Phragmites australis* in several polluted soils and sediments of an industrialized region of Northern Portugal** *Mycorrhiza* **10**(5): 241–248 [*International Institute of Biotechnology, Biotechnology MIRECEN, 1/13 Innovation Buildings, Sittingbourne Research Centre, Sittingbourne, Kent ME9 8HL, UK]
McGonigle T P*. 2001	**On the use of non-linear regression with the logistic equation for changes with time of percentage root length colonized by arbuscular mycorrhizal fungi** *Mycorrhiza* **10**(5): 249–254 [*Department of Biological Sciences, Idaho State University, Campus Box 8044, Pocatello, ID 83209, USA]

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Allen M F*. 2001	**Modeling arbuscular mycorrhizal infection: is % infection an appropriate variable?** *Mycorrhiza* **10**(5): 255–258 [*Centre for Conservation Biology, University of California, Riverside, CA 92521-0334, USA]
Millner P D*, Mulbry W W, and Reynolds S L. 2001	**Taxon-specific oligonucleotide primers for detection of *Glomus etunicatum*** *Mycorrhiza* **10**(6): 259–266 [*US Department of Agriculture, Agricultural Research Service, Beltsville Agricultural Research Centre, Soil Microbial Systems Laboratory, 10300 Baltimore Blvd., Beltsville, MD 20705-2350, USA]
Nakano A, Takahashi K, Koide R T, Kimura M*. 2001	**Determination of the nitrogen source for arbuscular mycorrhizal fungi by N application to soil and plants** *Mycorrhiza* **10**(6): 267–274 [*Laboratory of Soil Biology and Chemistry, School of Bioagricultural Sciences, Nagoya University, Chikusa, Nagoya 464-8601, Japan]
Berreck M* and Haselwandter K. 2001	**Effect of the arbuscular mycorrhizal symbiosis upon uptake of cesium and other cations by plants** *Mycorrhiza* **10**(6): 275–280 [*Department of Microbiology, University of Innsbruck, Technikerstrasse 25, 6020 Innsbruck, Austria]
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Anderson I C*, Chambers S M, and Cairney J W G. 2001	**Variation in nitrogen source utilisation by *Pisolithus* isolates maintained in axenic culture** ***Mycorrhiza* 11(1): 53–56** **[*Mycorrhiza Research Group, School of Science, Food and Horticulture, University of Western Sydney, Parramatta Campus, Locked Bag 1797, Penrith South DC, NSW 1797, Australia]**
Yamada A*, Ogura T, and Ohmasa M. 2001	**Cultivation of mushrooms of edible ectomycorrhizal fungi associated with *Pinus densiflora* by in vitro mycorrhizal synthesis I. Primordium and basidiocarp formation in open-pot culture** *Mycorrhiza* **11**(2): 59–66 [* Ibaraki Prefectural Forestry Center, 4692 Toh, Naka machi, Naka gun, Ibaraki 311-0122, Japan]
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Tehran, **Iran**
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First and Second Announcement, Contact: Dr D Ershad Mailto

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E-mail ripinza@valdivia.uca.uach.cl

Xalapa, **Mexico**
13–17 May 2002

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