

Effect of edaphic factors and seasonal variation on spore density and root colonization of arbuscular mycorrhizal fungi in sugarcane fields

Nallusamy Sivakumar

Received: 7 December 2011 / Accepted: 14 March 2012 / Published online: 17 April 2012
© Springer-Verlag and the University of Milan 2012

Abstract Sugarcane fields in 14 different study sites were analyzed for the presence of different arbuscular mycorrhizal fungal (AMF) spores. A total of 23 AMF species representing four genera were identified, among which *Glomus fasciculatum* and *G. mosseae* were the dominant species. The mean spore density in the root-zone soils of sugarcane plants varied from 119 to 583 per 100 g of soil, and the mean percentage root colonization varied from 60 to 89 %. A study of the effect of edaphic factors on AMF spore density and percentage root colonization revealed a positive correlation between pH and AMF spore density and root colonization and a negative correlation between electrical conductivity, nitrogen, and phosphorus. A positive correlation was observed between AMF spore density and root colonization. Season was also found to play a vital role in determining AMF spore density and percentage root colonization, with high spore density and root colonization observed during the summer season and lower spore densities and root colonization during the winter season.

Keywords Edaphic factors · Seasonal variation · Arbuscular mycorrhizal fungi · Sugarcane

Introduction

Arbuscular mycorrhizal fungi (AMF) are a main component of the soil microbiota in most agroecosystems. These are obligate symbiotic soil fungi which colonize the roots of the

majority of plants, regulating community and ecosystem functioning. These fungi biotrophically colonize in the root cortex and develop as extramatrical mycelium that helps the plant to acquire mineral nutrients from soil (Harley and Smith 1983). In nutrient-poor soil of humid tropical regions, many plants are dependent on AMF (Gemma et al. 2002), while as obligatory symbionts, AMF are believed to be dependent upon the host plant for carbon fixation. In turn, the plant receives a variety of benefits which include increased nutrient uptake, notably of immobile nutrients such as phosphorus (P) and zinc (Zn) (Bolan 1991; Burkert and Robson 1994). AMF can also enhance resistance to root pathogens (Borowicz 2001) and abiotic stresses, such as drought and metal toxicity (Meharg and Cairney 2000), and they may also play a role in the formation of soil aggregates, building up a macrocarpous structure of soil that allows the penetration of water and air and prevents soil erosion (Miller and Justrow 1992).

Seasonal fungal patterns are closely related to host phenology and climatic variations (Rosendahl and Rosendahl 1992; Allen 1996). Seasonal variation in the diversity of AMF has been studied mainly in sand dune systems, and very little information is currently available on other habitats (Lugo and Cabello 2002). Seasonal variation in the activity of AMF in tropical soil is poorly understood and generally based on very few observations (Muthukumar et al. 2006). A common difficulty in detecting statistically significant seasonal trends results from an aggregated spore distribution of AMF in soil. An accurate mathematical study of seasonal spore abundance is therefore essential to gaining an understanding of the seasonal dynamics of AMF. The potential for increasing plant growth by the effective management of AM strains reinforces the need to determine species composition in cultivated soils of different locations. The density of AMF spores varies with the kind of

N. Sivakumar (✉)
Department of Biology, College of Science,
Sultan Qaboos University,
P.O. Box 36, 123 Muscat, Sultanate of Oman
e-mail: apnsiva@squ.edu.om

cultivated crop (Isobe et al. 2007), indicating that soil from specific crop fields must be examined to clarify the differences in AMF spore density in each region. India boasts the largest surface area under sugarcane cultivation, a crop that grows best in a well-drained productive soil, a hot and humid climate, sufficient nitrogen (N), P, potassium (K) and other nutrients for growth. Therefore, the association between AMF and sugarcane is of paramount importance. However, information on the occurrence of AMF populations with this cash crop is still quite limited. The aim of the study reported here was to conduct a detailed examination of the AMF status of sugarcane cultivated fields, the relationship between edaphic factors, AMF spore density and root colonization, and the influence of different seasons on AMF. Fourteen different study sites in Pudukkottai district, India, were selected for this study. This is the first comprehensive study of AMF in these regions.

Materials and methods

Selection of sampling areas

To assess the diversity of AM fungi in rhizosphere soils and its association with roots of sugarcane, root samples together with rhizosphere soils were collected from 14 different regions in Pudukkottai district, India (Alathur, Avudaiyar kovil, Budalur, Gandarvakkottai, Illupur, Kavinad, Madukkur, Mudukulam, Mullur, Perungalur, Poram, Vayalagam, Vadavalam, and Visalur).

Tropical details of the study areas

Pudukkottai district has a total geographical area of 4.645 lakhs ha, 87.8 m a.s.l. with a tropical climate. All of the study sites have an annual atmospheric temperature of 21 °C with a mean annual temperature of 29.5 °C. The mean annual rain fall was 940 mm, with the major precipitation occurring during the months of October to December. The study sites have four different seasons, i.e., winter (WIN: January–March), summer (SUM: April–June), south-west monsoon period (SWM: July–September), and north-east monsoon (NEM: October–December).

Sample collection

Five different fields continuously cultivated with sugarcane were selected from each study site, and an area of about 500 m² was taken for sampling in each field. Ten healthy plants were selected from each field, and their roots and rhizosphere soil samples were collected down to a soil depth of 30 cm. The samples were collected in triplicates and brought to the laboratory in sealed plastic bags where they

were stored at 5–10 °C (Koske and Halvorson 1981). The root samples were washed thoroughly to removed attached soil particles, cut into several small segments (length 1 cm), and fixed in formalin, acetic acid, and alcohol (FAA) in 5:5:90 ratios (Phillips and Hayman 1970). Soil samples collected from each site was mixed thoroughly, and a portion of each soil sample was analyzed for soil texture, pH, electrical conductivity (EC), percentage organic matter (OM), available N, P, K, Zn, copper (Cu), manganese (Mn), and iron (Fe). From the remainder each soil sample, 100 g was taken to estimate AM fungal spore number.

Analysis of physicochemical characteristics of soil samples

All samples were air dried, ground into powder, and passed through a 1-mm-pore sieve. The sieved powdered samples were collected in plastic containers, sealed, and stored at 4 °C until further use. For the pH measurement, 20 g of soil was added to 40 ml of distilled water and stirred thoroughly with a glass rod; after standing undisturbed for 15 min, the pH was measured. For the measurement of EC, 20 g soil was dissolved in 100 ml distilled water and agitated for 1 h in a shaker; conductivity was then measured using an electronic digital conductivity meter (Elico, Hyderabad, India). Soil OM content (Walkey and Black 1934), available N and K (Sankaran 1966), available P (Olsen et al. 1954), and available micronutrients (Lindsey and Norwell 1978) were also analyzed.

Isolation and enumeration of AMF spore

Spore and sporocarp numbers were determined by the wet sieving and decanting method (Gerdemann and Nicolson 1963) followed by sucrose density centrifugation (Ianson and Allen 1986). In brief, 100 g of soil was dispersed in 1 l of water and centrifuged. After centrifugation, the suspension was carefully decanted through a descending series of sieves (mesh size:500 to 40 μm) and a 40 % sucrose solution added. Residues were filtered through gridded filter papers, and all intact spores were counted under a stereoscopic microscope at 40× magnification. Sporocarps and spore clusters were considered as one unit. For the taxonomic identification, AMF spores were mounted onto slides using polyvinyl alcohol lactoglycerol (Omar et al. 1979) with or without Melzer reagent (Mortan 1988) and identified based on the synoptic keys of Walker and Sanders (1986) and Schenck and Perez (1990).

Assessment and quantification of AM fungi in roots

The FAA-fixed root samples were washed, cleared in 2.5 % KOH at 90 °C (Koske and Gemma 1989), acidified with 5 N HCl, and stained with trypan blue (0.05 %) in lactoglycerol

(Phillips and Hayman 1970). Fungal colonization was quantified by the glass slide method, in which 50 randomly selected 1-cm-long root segment units were examined microscopically (Giovannetti and Mosse 1980). Only fine roots were considered for quantification because larger roots were not colonized or not easy to observe. Total colonization was expressed as the percentage of AM colonization.

Statistical analysis

Pearson product moment correlation was followed to examine the relationship among AMF spore abundance, AM percentage colonization, and physicochemical characteristics of the soil. The mean values and standard deviations were calculated for each site from the data obtained from five different fields (in triplicate). To determine significant variations in AMF spore density and percentage root colonization during different seasons, analysis of variance (ANOVA) was performed by one-way ANOVA procedures followed by Tukey HSD post hoc tests using SPSS ver. 11.5 (SPSS, Chicago, IL). Statistical differences at $p < 0.05$ were considered to be significant.

Results

Physicochemical characteristics of soil samples

Three different soil types were found in the study sites. Sandy loam soil was found in Alathur, Gandarvakkottai,

Madukkur, Kavinad, and Vadavalam; clay loam soil was found in Avudaiyarkovil, Perungalur, and Visalur; sandy clay loamy soil was found in Budalur, Illuppur, Mudukulam, Mullur, Porum, and Vialogam. Over all of the study sites, the pH ranged from 6.1 to 7.8, the EC ranged from 0.11 to 0.20, and the OM ranged from 1.18 to 1.40%. Macronutrients such as N, P, and K ranged from 71.4 to 81.9, from 5 to 11, and from 187 to 262 kg acre⁻¹, respectively. Micronutrients such as Cu, Zn, Fe, and Mn ranged from 2.18 to 3.62, from 0.76 to 1.1, from 5.18 to 7.2, and from 3.16 to 4.16 kg acre⁻¹, respectively (Table 1).

Correlation between AMF spore density and root colonization

To examine whether the physicochemical characteristics of soil could have any influence on AMF, a correlation study was carried out to determine the relationship between root-zone soil parameters, such as pH, EC, OM, available N, P, K, Cu, Zn, Mn, and Fe and AMF spore density and percentage AM root colonization. The mean spore density was negatively correlated with EC, OM, N, P, Cu, Zn, and Fe and positively correlated with pH, K, and Mn. The percentage root colonization showed a negative correlation with EC, N, P, and K and a positive correlation with pH, OM, Cu, Fe, and Mn. A significant positive correlation was found between Cu and AMF root colonization. No correlation was found between root infection and Zn (Table 2). A positive correlation was observed between AM spore density and percentage root colonization.

Table 1 Physicochemical characteristics of soil samples isolated from the 14 different study sites

Study site	Soil type ^a	pH	EC	OM (%)	Macronutrient (kg acre ⁻¹)			Micronutrient (kg acre ⁻¹)			
					N	P	K	Cu	Zn	Fe	Mn
Alathur	SL	6.1	0.13	1.27	89.9	5.0	187	2.52	0.76	5.18	3.66
Avudaiyar kovil	CL	7.8	0.17	1.37	76.7	6.5	206	2.65	0.81	6.23	3.44
Budalur	SCL	6.6	0.12	1.23	80.0	7.4	220	2.76	0.84	6.05	3.52
Gandarvakkottai	SL	7.0	0.19	1.16	86.4	6.3	245	2.56	0.92	5.86	3.68
Illuppur	SCL	7.6	0.11	1.34	74.7	6.6	193	3.34	1.06	5.47	3.20
Kavinad	SL	7.3	0.15	1.25	71.4	5.8	235	2.81	0.72	6.37	4.16
Madukkur	SL	7.2	0.17	1.18	77.3	8.9	255	2.37	0.95	7.20	3.72
Mudukulam	SCL	6.7	0.19	1.26	87.5	10.3	247	2.93	0.83	6.40	3.48
Mullur	SCL	7.1	0.20	1.32	83.6	11.0	195	3.10	1.10	5.72	2.97
Perungalur	CL	7.5	0.14	1.19	79.4	9.2	262	2.18	0.94	5.94	3.63
Porum	SCL	7.4	0.13	1.40	81.2	7.4	237	2.47	0.79	6.14	3.94
Vadavalam	SL	7.3	0.16	1.29	78.7	7.6	210	3.62	0.85	7.03	4.07
Vyalogam	SCL	6.6	0.16	1.36	73.8	9.4	245	2.39	1.07	6.63	3.54
Visalur	CL	6.8	0.17	1.21	84.3	8.2	226	2.51	0.92	5.62	3.16

EC, Electrical conductivity; OM, organic matter

^a CL, Clay soil; SCL, sandy clay loamy soil; SL, sandy loam soil

Table 2 Correlation between arbuscular mycorrhizal fungal variables and physico-chemical characteristics of soils in the study sites

Variable	Colonization (%)	pH	EC	OM	N	P	K	Cu	Zn	Fe	Mn
Spore number	0.40	0.47	-0.05	-0.06	-0.30	-0.43	0.26	-0.01	-0.27	-0.09	0.31
Colonization (%)	-	0.43	-0.08	0.02	-0.49	-0.23	-0.17	0.64*	0.00	0.17	0.36

* Correlation is significant at the 0.05 level

AM diversity and species richness

Altogether, 23 AM fungal species were isolated in the study sites, representing four genera, namely, *Acaulospora*, *Gigaspora*, *Glomus*, and *Scutellospora*. Of these four genera, *Glomus* was the most prevalent, and both *G. mosseae* and *G. fasciculatum* were found as dominant species with a distribution frequency of 92.8 (Table 3). The mean spore density in the root-zone soils of sugarcane plants varied fivefold from 119 to 583 per 100 g of soil. The minimum spore number was 119 and was observed in Budalur soil, while soil at the Kavinad site contained the maximum number of spores (583). The spore density at Illuppur, Gandarvakkottai, and Kavinad was significantly higher than that at other study sites. In comparison, the lowest mean spore density was found at Mullar (170.15 spores per 100 g of soil) (Fig. 1).

Effect of seasonal variation on AMF spore density

The highest mean spore density of 378.79 spores per 100 g soil was observed during the summer (Fig. 2) and was significantly higher than that during all other seasons. Increases in spore density were observed during the SWM and NEM, with a mean spore density of 329.27 and 291.29 spores per 100 g soil, respectively; however, the difference between these two seasons is insignificant (Fig. 3). The mean spore density was significantly lower during the winter than during the other seasons (222 spores/100 g soil). Spore density was highest during summer (dry season) and lowest in winter (wet season), with intermediate values in the SWM and NEM.

Effect of seasonal variation on AMF root colonization

Almost all the root samples of sugarcane plants were positive for AMF colonization. The highest mean percentage AMF colonization was observed in Illuppur, Kavinad, and Vadavalam, while the lowest was observed at Visalur (Fig. 4). Seasonal variations also influence the percentage root colonization of AM in the study sites (Fig. 5). The highest percentage of colonization was observed during SUM and SWM, but it was decreased significantly during NEM. The lowest infection percentage infection was observed during WIN (Fig. 6).

Discussion

The study reported here was undertaken to make a detailed examination of the AM status of sugarcane cultivated fields. The physicochemical characteristics of all the 14 study sites were analyzed. Most of these were sandy clay loam or sandy loam soils. Soil texture may affect plant growth as well as mycorrhizal efficiency in various ways, such as through drainage, aeration, and limitations in nutrient availability (Joshi and Singh 1995). There were three sites with clay loam soil. The pH over all sites ranged from 6.1 to 7.8.

Effect of soil pH on AMF

Little information is currently available on the correlation between edaphic characters and spore populations under Indian conditions. In this study, AMF spores were found in both slightly acidic and neutral to slightly alkaline soils. Some AMF species occur either in acid or in alkaline soils and others occur in both (Young et al. 1985; Robson and Abbott 1989). Spore germination and root colonization of AMF are suppressed in acidic and alkaline soils (Isobe et al. 2007). Variations in soil pH may alter the concentration of many nutrients and toxic ions in the soil and thereby affect the development and function of AM fungi. Consequently, the density of AMF spores is assumed to be low in acidic and alkaline soils. However, in the present study, a positive correlation was observed for soil pH with AM spore density and root colonization, possibly due to the narrow range of pH (6.1–7.8) in the soils observed.

Effect of OM, N, and P on AMF

The OM showed a positive correlation with both AMF spore density and root colonization, thereby corroborating previously reported results (Boddington and Dodd 2000; Khanam et al. 2006). However, a negative correlation found between N and AMF spore density, and N and AMF root colonization. N can either stimulate or suppress root colonization and spore production by AM through modifications of soil pH (Sylvia and Neal 1990). It has been shown that AM spore density in fields reaches a maximum when the P status in the soil is less than that required for maximum plant

Table 3 Diversity of arbuscular mycorrhizal fungal flora in rhizosphere soil of 14 different study sites

AMF spores	Site 1	Site 2	Site 3	Site 4	Site 5	Site 6	Site 7	Site 8	Site 9	Site 10	Site 11	Site 12	Site 13	Site 14	Distribution frequency (%)
<i>Acaulospora elegans</i>	+	+	-	+	+	+	-	+	+	+	-	+	-	+	71.4
<i>A. foveata</i>	+	-	-	+	+	+	-	+	-	+	+	-	+	-	57.1
<i>A. laevis</i>	+	+	-	+	+	+	+	-	-	+	+	-	+	+	71.4
<i>A. morrowae</i>	-	+	+	-	+	+	+	+	+	-	-	+	-	+	57.1
<i>Gigaspora candida</i>	-	+	-	+	-	+	+	-	+	+	+	+	-	+	64.3
<i>G. decipiens</i>	-	+	-	+	+	+	+	-	+	+	+	+	-	+	64.3
<i>G. margarita</i>	+	+	+	+	+	+	+	+	+	+	+	-	+	-	78.6
<i>Glomus aggregatum</i>	-	+	+	+	+	+	+	+	+	+	+	-	+	+	78.6
<i>G. clarum</i>	-	-	+	+	-	+	+	-	+	-	+	-	+	+	57.1
<i>G. constrictum</i>	+	+	-	+	+	+	+	-	-	+	-	+	+	-	64.3
<i>G. deserticola</i>	-	+	-	+	+	+	+	-	-	+	+	-	-	+	57.1
<i>G. fasciculatum</i>	+	+	+	+	+	+	+	+	+	-	+	+	+	+	92.8
<i>G. geosporum</i>	-	+	-	+	+	+	+	-	+	-	+	+	+	-	64.3
<i>G. intradices</i>	+	-	+	+	+	+	-	+	-	+	+	-	+	+	71.4
<i>G. macrocarpum</i>	+	+	-	+	-	+	+	+	-	+	-	+	-	+	64.3
<i>G. microcorpum</i>	+	+	-	+	+	+	+	-	+	+	+	+	-	-	71.4
<i>G. mosseae</i>	+	-	+	+	+	+	+	+	+	+	+	+	+	+	92.8
<i>G. multisubtensum</i>	-	-	+	-	+	-	+	+	-	-	+	+	+	-	50.0
<i>G. occultum</i>	+	-	-	+	-	+	+	+	+	+	-	+	-	+	57.1
<i>G. radiatum</i>	-	+	-	+	+	+	+	+	-	+	+	-	+	-	57.1
<i>G. scintillans</i>	+	-	+	-	+	+	+	-	+	+	+	-	-	+	64.3
<i>Scutellospora calospora</i>	+	+	-	+	+	+	+	+	-	+	+	-	+	-	64.3
<i>S. nigra</i>	+	+	+	+	+	+	+	+	+	+	+	+	+	-	78.6
Species richness (%)	60.9	69.6	43.5	86.9	78.3	95.6	52.1	65.2	60.9	78.3	78.3	56.5	56.5	60.9	

+, Presence of arbuscular mycorrhizal fungi (AMF); -, absence of AMF

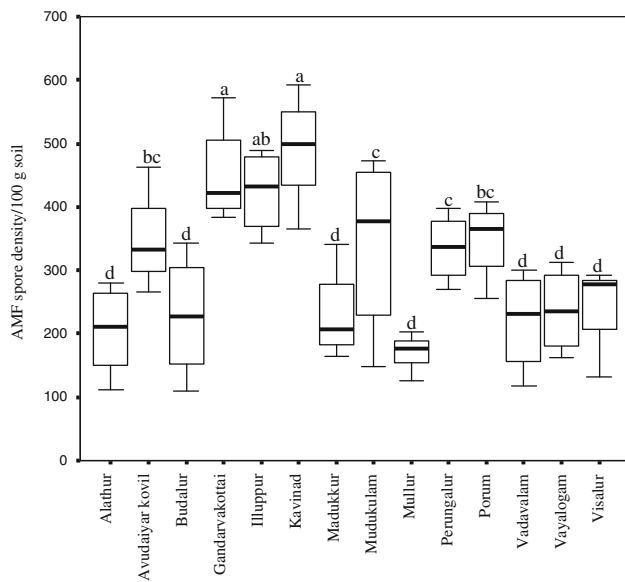


Fig. 1 Box-and-whisker plots showing the mean, range, and significant variation in arbuscular mycorrhizal fungal (*AMF*) spore density in different study sites. Different lowercase letters above the boxes indicate a significant difference at the 0.05 level

growth (Abbott and Robson 1991). Although the correlation in the present study was not statistically significant, negative correlation coefficients of -0.43 and -0.23 were observed between spore density and P, and root colonization and P, respectively. The customary agricultural practice at the sugarcane fields of the study sites was to apply chemical fertilizers. Increasing the application of P

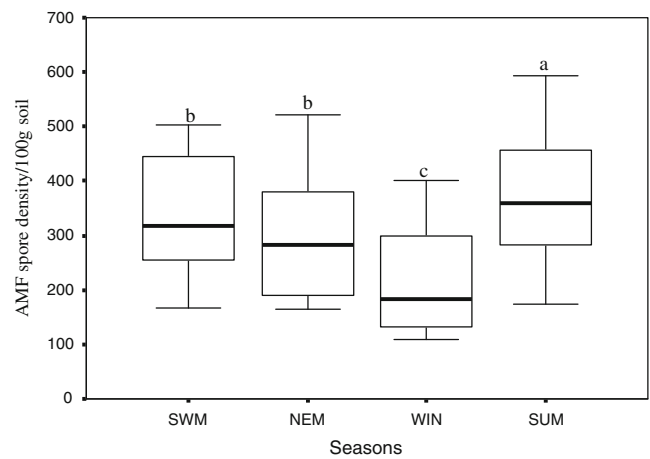


Fig. 3 Box-and-whisker plots of the variation in *AMF* spore density during four seasons. Different lowercase letters above the boxes indicate a significant difference at the 0.05 level

fertilizers would increase the amount of available P in the soil, resulting in the soil having a low P adsorption coefficient. The application of P fertilizer decreases the rate of infection and the density of *AMF* in soil (Isobe and Tsuboki 1998; Mohammed et al. 2004; Lekberg and Koide 2005). Hence, if the aim is to increase the density of *AMF* spores, the excessive application of P fertilizers should be avoided. The positive correlation of soil K on spore density shows that the slow diffusion of K ions in the soil may favor spore germination (Haselwandter and Bowen 1996).

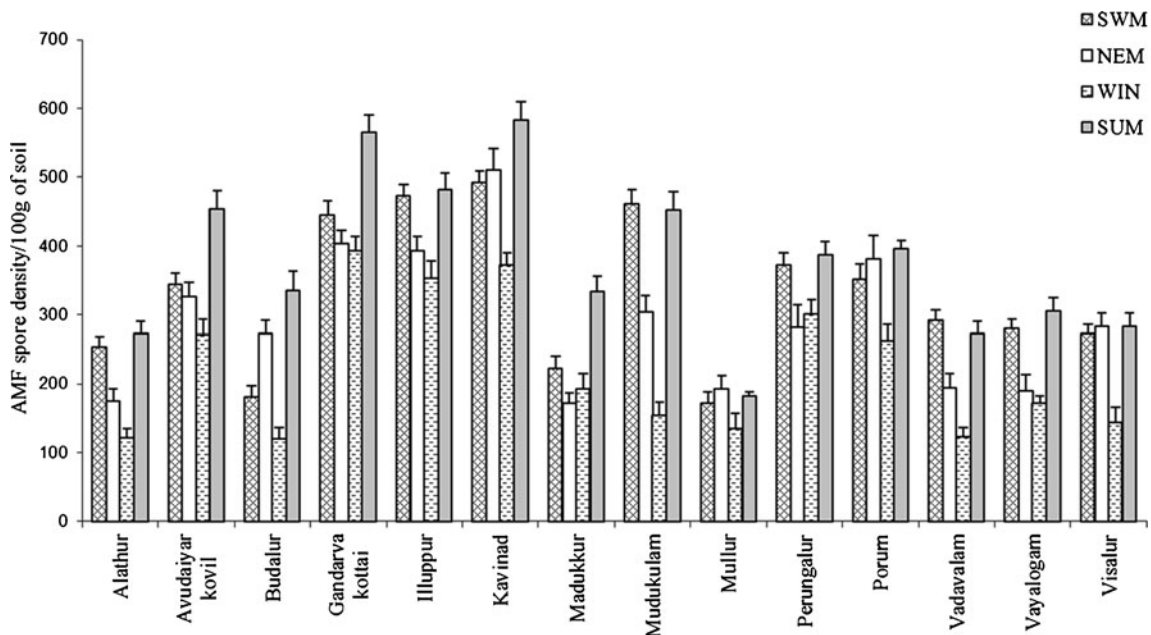


Fig. 2 Influence of seasonal variation on the spore density of *AMF* in 14 different study sites. *SWM* South-west monsoon, *NEM* north-east monsoon, *WIN* winter, *SUM* summer

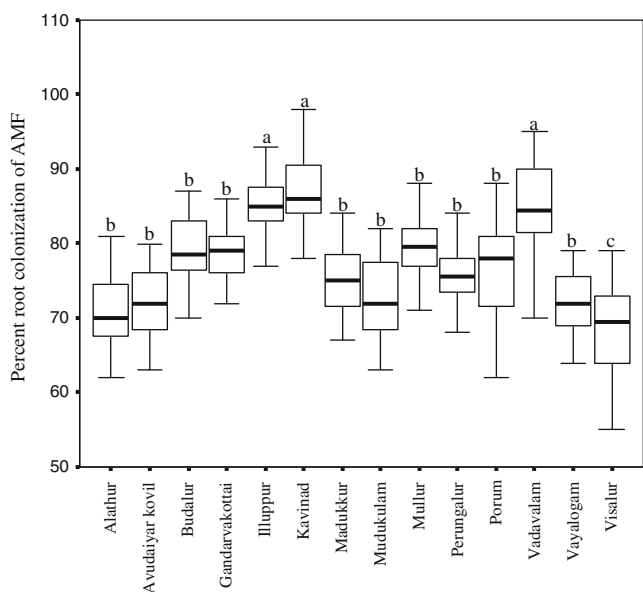


Fig. 4 Box-and-whisker plots showing the mean percentage AMF root colonization and its significant variation in different study sites. Different lowercase letters above the boxes indicate a significant difference at the 0.05 level

Correlation between AMF spore density and root colonization

AM spore density is positively correlated to the percentage of root colonization in sugarcane (Wuen et al. 2002; Isobe et al. 2008). The number of spores has also been correlated with the phenological stage of the host plant. It has also been suggested

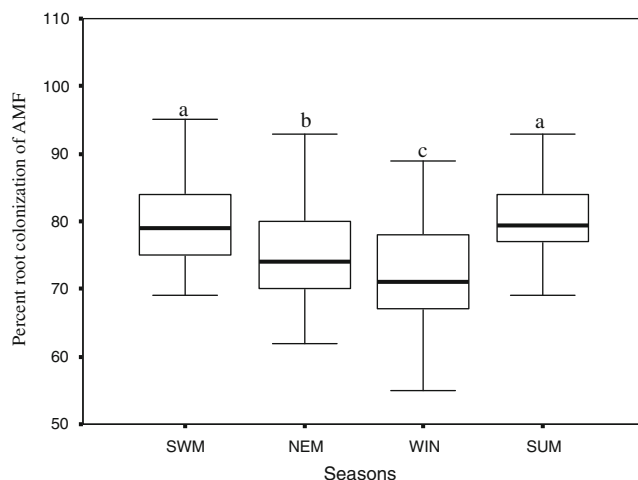


Fig. 6 Variations in percentage AMF root colonization during four seasons. Different lowercase letters above the boxes indicate a significant difference at the 0.05 level

that AMF show a different behavior in perennial plants, without a condensed sporulation at the end of the life cycle, as in annuals (Gemma and Koske 1988). In situations where the spore density is positively correlated with the extent of root colonization, both aspects may increase during the growing season of annual plants. However, this relationship does not always hold; Louis and Lim (1987) observed an inverse relationship between spore density and colonization in four perennial trees from a lowland tropical rain forest. The relationship of spore density and root colonization with different soil characteristics is a result of the interactions between them and could be specific for each case.

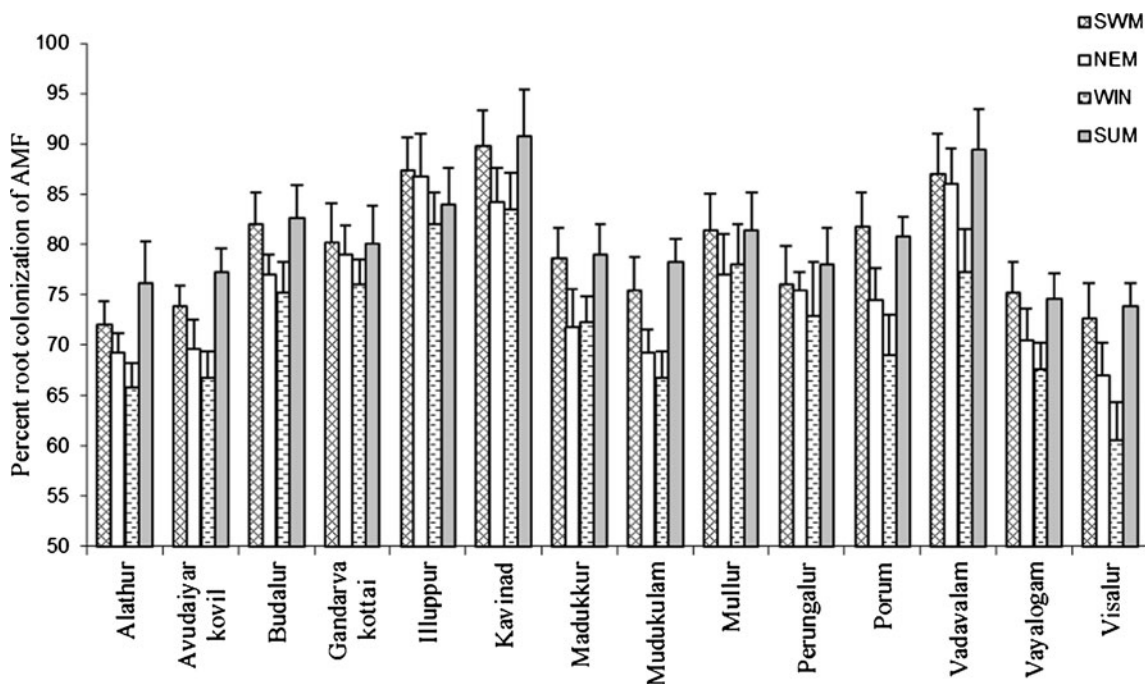


Fig. 5 Influence of seasonal variation on the percentage root colonization of AMF in 14 different study sites

AM diversity and species richness

A total of 23 AM fungal species, belonging to the genera *Acaulospora*, *Gigaspora*, *Glomus*, and *Scutellospora*, were found in the study sites. Because of edaphic factors and climatic changes, the spores and activities of AM fungi may not distribute equally in different soils. Moreover, the density of AM fungal spores generally varies significantly with the season and growing stage of the host crop.

In general, the presence of more than one AMF species is quite common in the perennial rhizosphere (Thaper and Khan 1985). The occurrence of several AM fungi in soil or within the roots suggests the possibility of an intra-specific competition between different species. The spore abundance of AMF is determined by the host plant species and the environmental variables rather than by the AM fungal species itself (Koske and Halvorson 1981). Edaphic characters, such as soil type (Joshi and Singh 1995), soil pH (Sindhu and Behl 1997), and soil fertility (Abbott and Robson 1991), have been reported to influence AM sporulation. It is unlikely that a single property would determine AM fungal dynamics (Walker et al. 1982). The relationship between spore density, species richness, and the distribution frequency of AMF with different soil characteristics is a result of the interactions between all of these factors and could be specific for each case.

Effect of seasonal variation on AMF spore density

Seasonal variation plays a remarkable role on the occurrence of AM fungi (Mallesha and Bagyaraj 1991). Spore density was highest during the summer (dry season) and lowest during the winter (wet season), with intermediate values in the SWM and NEM. This seasonal pattern in spore density has been observed in salt marsh soils (Carvalho et al. 2001) and temperate grasslands (Escudero and Mendoza 2005). In xeric Mediterranean grasslands, the variation in total spore density appears to be controlled mainly by dry and wet periods (Lugo and Cabello 2002). Mago and Mukerji (1994) observed that AM colonization was the lowest during the winter and highest during the late summer in some members of the Lamiaceae. Spore density reflects the net effect of sporulation against spore disappearance due to germination, dispersal, leaching, predation, mortality, and other factors. The crop roots that can be infected by AM fungi are another important factor for the difference in spore density because AM fungi do not form spores without infection. The variation in spore density is a consequence of many interacting factors, such as plant communities, soil characteristics, sporulating nature of fungi, growing season of host plant, and climate. It is necessary to understand the relationship between spore production and the type of crop.

Effect of seasonal variation on AMF root colonization

Almost all of the root samples of the collected sugarcane plants had an association with AM fungi. The highest percentage of colonization was observed during SUM and SWM, but this decreased significantly during the NEM. The lowest percentage infection of AM was observed during the WIN. There are several explanations for the seasonal variation in root colonization, including exudation of toxic metabolites (Iqbal and Queorshi 1986) and the production of easily oxidizable compounds (St. John and Coleman 1983). Although these factors play a decisive role in colonization, several edaphic and climatic factors may also influence the process (Giovannetti 1985). It has also been reported that the community of AM fungi may determine host plant community's association and production (Van Der Heijden et al. 1998). Bever (2002) demonstrated that each endophyte multiplied quite differently on different host plants and that the infection ratio differed with the species of AM fungi. In the present study sugarcane plants collected from 14 different sites showed more than 72 % colonization in all seasons, with a mean percentage colonization for all seasons of 76.9 %. Such a high degree of colonization may be due to the fact that the plants in the study sites were essentially P-deficient.

Conclusion

Species diversity was apparent in all study sites, but *G. mosseae* and *G. fasciculatum* were found in almost all of the soils. The marked difference observed in the composition of AM fungi in the study sites may be attributable to the influence of edaphic factors and seasonal differences. Variations in climate also influence the selection of AMF as climate regulates the incidence of specific fungal strains in the soil. Factors such as cultural practices and vegetation in the study sites may also contribute to determining the dominance of a particular species. The measured indices, such as species richness, spore density, distribution frequency, and percentage root colonization, varied in the different regions. The infection ratio of AMF might be affected by the difference in AMF flora. Therefore, it is necessary to determine the population density of each AMF species and the respective infection ratio with sugarcane plants that would favor growth and production of the host plant.

Acknowledgements The author would like to thank the management of J. J. College of Arts and Science, Pudukkottai, India, for its support.

References

- Abbott L, Robson A (1991) Factors influencing the occurrence of vesicular—arbuscular mycorrhizas. *Agric Ecosyst Environ* 35:121–150
- Allen MF (1996) The ecology of arbuscular mycorrhizas: a look back into the 20th century and a peak into the 21st. *Mycol Res* 100:769–782
- Bever J (2002) Host-specificity of AM fungal population growth rates can generate feedback on plant growth. *Plant Soil* 244:281–290
- Boddington CL, Dodd JC (2000) The effect of agricultural practices on the development of indigenous arbuscular mycorrhizal fungi. *Plant Soil* 218:145–157
- Bolan NS (1991) A critical review of the role of mycorrhizal fungi in the uptake of phosphorus by plants. *Plant Soil* 134:189–208
- Borowicz VA (2001) Do arbuscular mycorrhizal fungi alter plant-pathogen reactions? *Ecology* 82:3057–3068
- Burkert B, Robson A (1994) ⁶⁵Zn uptake in subterranean clover (*Trifolium subterraneum* L.) by three vesicular-arbuscular mycorrhizal fungi in a root-free sandy soil. *Soil Biol Biochem* 26:1117–1124
- Carvalho LM, Cacados I, Martiris-Loucao MA (2001) Temporal and spatial variation of arbuscular mycorrhizas in salt marsh plants of Tagus estuary (Portugal). *Mycorrhiza* 11:303–309
- Escudero V, Mendoza R (2005) Seasonal variation of arbuscular mycorrhizal fungi in temperate grasslands along a wide hydrologic gradient. *Mycorrhiza* 15:291–299
- Gemma JK, Koske RE (1988) Seasonal variation in spore abundance and dormancy of *Gigaspora gigantea* and in mycorrhizal inoculum potential of a dune soil. *Mycologia* 80:211–216
- Gemma JN, Koske RE, Habte M (2002) Mycorrhizal dependency of some endemic and endangered Hawaiian plant species. *Am J Bot* 89:337–345
- Gerdemann JW, Nicolson TH (1963) Spores of mycorrhizal endogone species extracted from soil by wet sieving and decanting. *Trans Br Mycol Soc* 46:235–244
- Giovannetti M (1985) Seasonal variations of vesicular arbuscular mycorrhizas and endogonaceous spores in a maritime and sand dune. *Trans Br Mycol Soc* 84:679–684
- Giovannetti M, Mosse B (1980) An evaluation of technique for measuring vesicular—arbuscular mycorrhizae infection in roots. *New Phytol* 84:489–500
- Harley JL, Smith SE (1983) *Mycorrhizal symbiosis*. Academic Press, London
- Haselwandter K, Bowen GD (1996) Mycorrhizal relations in trees for agroforestry and land rehabilitation. *For Ecol Manag* 81:1–17
- Ianson DC, Allen MF (1986) The effects of soil texture on extraction of vesicular-arbuscular mycorrhizal spores from arid soils. *Mycologia* 78:164–168
- Iqbal SH, Queorshi KS (1986) The influence of mixed showing (cereals and crucifers) and crop rotation on the development of mycorrhiza and subsequent growth of crops under field conditions. *Biologia* 22:287–298
- Isobe K, Tsuboki Y (1998) Relationship between the amount of root exudates and the infection rate of arbuscular mycorrhizal fungi in graminous and leguminous crops. *Plant Prod Sci* 1:37–38
- Isobe K, Aizawa E, Iguchi Y, Ishii R (2007) Distribution of arbuscular mycorrhizal fungi in upland field soil of Japan. 1. Relationship between spore density and the soil environment factor. *Plant Prod Sci* 10:122–128
- Isobe K, Sugimura H, Maeshima T, Ishii R (2008) Distribution of arbuscular mycorrhizal fungi in upland field soil of Japan. 2. Spore density of Arbuscular mycorrhizal fungi and infection ratio in soybean and maize fields. *Plant Prod Sci* 11:171–177
- Joshi KC, Singh HP (1995) Inter relationships among vesicular-arbuscular mycorrhizal population, soil properties and root colonization capacity of soil. *J Ind Soc Soil Sci* 43:204–207
- Khanam D, Mridha MAU, Solaiman ARM, Hossain T (2006) Effect of edaphic factors on root colonization and spore population of arbuscular mycorrhizal fungi. *Bull Inst Trop Agr Kyushu Univ* 29:97–104
- Koske RE, Gemma JN (1989) A modified procedure for staining roots to detect VA mycorrhizas. *Mycol Res* 92:486–488
- Koske RE, Halvorson WL (1981) A preliminary study of interactions between species of vesicular arbuscular mycorrhizal fungi in sand dunes. *Trans Br Mycol Soc* 76:411–416
- Lekberg Y, Koide RT (2005) Arbuscular mycorrhizal fungi, rhizobia, available soil P and nodulation of ground nut (*Arachis hypogaea*) in Zimbabwe. *Agric Ecosyst Environ* 110:143–48
- Lindsey DL, Norwell WA (1978) Development of a DTPA soil test for zinc, iron and Manganese and copper. *Soil Sci Soc Am J* 42:421–428
- Louis L, Lim G (1987) Spore density and root colonization of vesicular arbuscular mycorrhizas in tropical soil. *Trans Br Mycol Soc* 88:207–212
- Lugo MA, Cabello MN (2002) Native arbuscular mycorrhizal fungi (AMF) from mountain grassland (Cordoba, Argentina) I. Seasonal variation of fungal spore diversity. *Mycologia* 94:579–586
- Mago P, Mukerji KG (1994) Vesicular arbuscular mycorrhizae in Lamiaceae: I. Seasonal variation in some members. *Phytomorphology* 44:83–88
- Mallesha BC, Bagyaraj DJ (1991) Season favouring sporulation of VA-mycorrhizal fungi in cardamom plantations. *J Soil Soil Ecol* 11:75–78
- Meharg AA, Cairney JWG (2000) Co-evolution of mycorrhizal symbionts and their hosts to metal-contaminated environments. *Adv Ecol Res* 30:69–112
- Miller RM, Justrow JD (1992) The application of VA mycorrhizae to ecosystem restoration and reclamation. In: Allen MF (ed) *Mycorrhizal functioning*. Chapman and Hill, London, pp 438–467
- Mohammed A, Mitra B, Khan AG (2004) Effects of sheared root inoculum of *Glomus intraradices* on wheat grown at different phosphorus levels in the field. *Agric Ecosyst Environ* 103:245–249
- Mortan JB (1988) Taxonomy of VA mycorrhizal fungi: classification, nomenclature and identification. *Mycotaxon* 32:267–324
- Muthukumar T, Senthilkumar M, Rajangam M, Udayan K (2006) Arbuscular mycorrhizal morphology and dark septate fungal associations in medicinal and aromatic plants of Western Ghats, Southern India. *Mycorrhiza* 17:11–24
- Olsen SR, Cole CV, Watanabe FS, Decan LA (1954) Estimation of available phosphorus in soil by extraction with sodium bicarbonate. *USDA Agric Circulation* 939:1–1
- Omar MB, Bolland L, Heather WA (1979) P.V.A. (Polyvinyl alcohol) A permanent mounting medium for fungi. *Bull Br Mycol Soc* 13:31–32
- Phillips RC, Hayman DS (1970) Improved procedures for clearing root parasitic and staining vesicular—arbuscular mycorrhizal fungi for rapid assessment of infection. *Trans Br Mycol Soc* 55:158–160
- Robson AD, Abbott LK (1989) The effect of soil acidity on microbial activity in soils. In: Robson AD (ed) *Soil acidity and plant growth*. Academic Press, Sydney, pp 139–165
- Rosendahl S, Rosendahl CN (1992) Seasonal variation in occurrence of VA mycorrhizal infection types in a Danish Grassland community. In: Read DJ, Lewis DH, Fitter AH, Alexander IJ (eds) *Mycorrhizas in ecosystems*. CABI, Cambridge, p 400
- Sankaran A (1966) *A laboratory manual for agricultural chemistry*. Asia Publishing House, New Delhi
- Schenck NC, Perez Y (1990) *Manual for the identification of VA mycorrhizal fungi*. Synergistic Publications, Gainesville

- Sindhu OP, Behl HM (1997) Response of three *Glomus* species on growth of *Prosopis juliflora* Swartz. at high pH levels. *Symbiosis* 23:23–24
- St. John TV, Coleman DC (1983) The role of mycorrhizae in plant ecology. *Can J Bot* 61:1005–1014
- Sylvia DM, Neal LH (1990) Nitrogen affects the phosphate response of VA mycorrhiza. *New Phytol* 115:303–310
- Thaper HS, Khan SN (1985) Distribution of VA mycorrhizal fungi in forest soil of India. *Indian J For* 8:5–7
- Van Der Heijden MGA, Klironomos JN, Ursic M, Moutoglis P, Streitwolf-Engel R, Boller T, Wiemken A, Sanders IR (1998) Mycorrhizal fungal diversity determines plant biodiversity, ecosystem variability and productivity. *Nature* 396:69–72
- Walker C, Sanders FE (1986) Taxonomic concepts in the Endogonaceae: III. The separation of *Scutellospora* Gen. Nov. from *Gigaspora* Gerd and Trappe. *Mycotaxon* 27:168–182
- Walker C, Mize CW, McNabb JH (1982) Populations of endogonaceous fungi at two locations in central Iowa. *Can J Bot* 60:2518–2529
- Walkey A, Black TA (1934) An examination of the Degtiareff and method for determining soil organic matter and proposed modification of the chronic acid titration. *Soil Sci* 37:29–38
- Wuen K, Saito K, Sato S, Sugawara K (2002) Arbuscular mycorrhizal colonization and sporulation in rhizosphere of common species on native and sown grasslands. *Grassl Sci* 48:248–253
- Young JL, Davis EA, Rose SL (1985) Endomycorrhizal fungi in breeder wheats and *Triticale* cultivars field grown on a fertile soil. *Agron J* 77:219–224