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RESEARCH ARTICLE

INTERACTION OF AM FUNGI AND SUGARCANE (*Saccharum officinarum* L.)

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ABSTRACT

AM fungi is the most abundant kind of mycorrhizae and develops association with most of the plant species in the earth and enhances the growth and development of associated plants through the transport of required nutrients from the non-rhizosphere soil particularly phosphorus. The plants like sugarcane needs association of AM fungi to overcome 'P' deficiency due to its long standing periods in the same rhizosphere soil and leads to depletion of 'P' in the rhizosphere soil. In the present investigation eight isolates were obtained from eight different locations and identified as five different AM fungal species based on screening studies for acid phosphatase, alkaline phosphatase, efficiency to colonize sugarcane roots and interaction with sugarcane in the form of mycorrhizal inoculation effect (MIE) and relative mycorrhizal dependency, the *Glomus fasciculatum* was found to be a best AM fungal species for sugarcane and used in the pot culture studies. In the present research *G. fasciculatum* with recommended dose of farm yard manure significantly enhanced the plant height, cane girth, root colonization and spore number of sugarcane compared with other organic amendments.

INTRODUCTION

Sugarcane, being the long duration crop producing huge quantum of crop biomass, demands for nutrient inputs in large quantities. It is estimated that on an average the sugarcane removes around 100:60:225 kg of N, P_2O_5 and K_2O respectively for the production of 100 tonnes of cane yield (Sundra, 2002). Unless adequate supply of these nutrients is ensured, the sugarcane yield tends to decline even on most fertile soils. However, the indiscriminate prescription of inorganic fertilizer alone in the long run deteriorates soil health resulting in drastic reduction of cane yield. Hence, there is a permanent need to preserve the soil fertility with balanced proportion of major, secondary and micronutrients for sustainable crop productivity. The approach of integrated nutrient management through biofertilizers and organic manures with inorganic fertilizers in judicious proportions will go a long way to augment the important strategy of enhancing the soil fertility for increased crop productivity especially in a long duration crop like sugarcane. The association between plants and mycorrhizal fungi is wide spread, occurring in 80% of the plant species. Arbuscular mycorrhizal (AM) association is the common and prevalent among the different types of mycorrhizae. AM fungi is the most abundant kind of mycorrhizae found in association with every taxonomic group of plants and the list of species not infected is probably far shorter than the infected ones, these fungal associations are beneficial to crop plants in many ways, including enhancing the nutrient availability especially phosphorus, enhance water uptake and induces resistant against diseases and increase the yield (Lekberg and Koids, 2005). Based on the above traits the present investigation was undertaken to exploit the potentiality of AM fungi in the colonization, growth and development of sugarcane.

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MATERIALS AND METHODS

The laboratory experiments were carried out in the Department of Microbiology, Annamalai University, Annamalainagar, Chidambaram. The pot culture experiment was carried out at the pot culture house of the Department of Microbiology, Annamalai University, Tamilnadu. The survey of AM (*Arbuscular mycorrhizae*) fungal occurrence was conducted from different locations of Cuddalore District of Tamilnadu, India, where sugarcane (*Saccharum officinarum*) is grown as a commercial crop. Sugarcane setts (variety CoSi 98171) for the pot culture experiment were obtained from Department of Agronomy, Annamalai University. The names of the different locations selected for the survey of AM fungi in Cuddalore District of Tamilnadu, India are stated as Annamalainagar, Vilagam, Arasur, Cuddalore, Vallampadugai, Bhuvanagiri, Mutlur, Orathur, Palur, Parangipettai, Pichavaram, Puduchatram and Veeramudianatham. From all the locations soil samples were collected and analysed for the soil texture, soil pH, EC, organic carbon content, available phosphorus. In each location

the sugarcane root rhizosphere soil samples (20-30 core of 2.5 cm dia × 15-20 cm length) were collected. The roots and surrounding soil were excavated to a depth of 15-20 cm. Samples were then transferred to polythene bags for the isolation of AM fungal spores. The percentage colonization was calculated by clearing and staining the roots (Phillips and Hayman, 1970) followed by the determination of per cent root colonization by Krishna and Dart (1984).

Isolation, characterization and population of AM fungal spores

Different soil samples were examined for the presence of AM fungal spore by wet sieving (1000 – 45 µm) and decanting method described by Gerdemann and Nicolson (1963). These spores were cleaned of soil particles by sucrose density gradient centrifugation method and washed with distilled water (Mertz *et al.*, 1979). This spores suspension were counted with stereozoom microscope (× 47). During counting, morphologically similar spores were separated into groups, mounted and identified. The soil texture and the types of the spores in them were counted. Based on the taxonomic key of Gerdemann and Trappe (1974) the spores of *G. fasciculatum, Glomus mosseae, Glomus versiforme, Aculospora laevis* and *Gigaspora margarita* were identified.

Screening AM fungal cultures for efficiency to colonize the roots of sugarcane

The isolated five different AM fungal cultures viz., *G. mosseae, G. fasciculatum, G. versiforme, A. laevis* and *G. margarita* were studied for their comparative efficiency to colonize the roots of sugarcane. The 20 kg capacity cement pots were filled with sand: soil mix (1:1) fumigated by using 2 per cent formaldehyde and grown with single budded sugarcane setts at two setts pot^{-1}. The pots were inoculated with different cultures of AM fungi (soil inoculation at 50 g pot^{-1}). The following observations were taken on 60^{th}, 90^{th} and 120^{th} days after planting (DAP). (1) Relatively effective colonization in the sugarcane as evidenced by percent root infection (Phillips and Hayman, 1970). (2) Number of spores present in the 100g of rhizosphere soil of sugarcane (Gerdemann and Nicolson, 1963). (3) The acid and alkaline phosphatase enzyme activities of sugarcane (Morton, 1952).

Studies on the interaction of sugarcane with arbuscular mycorrhizal fungi (pot experiment)

Sugarcane (CoSi 98071) was used to study its interaction with AM fungi in unsterile soil. The relative mycorrhizal dependency (RMD) was measured and Mycorrhizal Inoculation Effects (MIE) were measured for five AM fungi *viz., G. mosseae, G. fasciculatum, G. versiforme, A. laevis* and *Gi. margarita*. Cement pots (60 × 30 × 60 cm) were filled with 20 kg of clay loam soil with pH 7.2 and native mycorrhizal population of 30 spores per 100 g of soil.

Treatments:

1. Sterilized soil
2. Unsterilized soil
3. Unsterilized soil + *G. mosseae*
4. Unsterilized soil + *G. fasciculatum*
5. Unsterilized soil + *G. versiforme*
6. Unsterilized soil + *A. laevis*
7. Unsterilized soil + *Gi. Margarita*

Each treatment was replicated three times. For sterilized soil, the pots filled with soil were sterilized with 2% formaldehyde. The AM fungal inoculation was done by placing the inoculums (50 g inoculums with 150 infective propagules per g) 2 cm below the soil surface. The single budded sugarcane setts were planted as two setts per pot. Irrigation was given as required. Plants were harvested 150 DAP and dry weight of the plants were recorded after drying to a constant weight.

Mycorrhizal inoculation effects

Mycorrhizal inoculation effect (MIE), a measure of growth response of crop plants to mycorrhizal inoculation was measured for *G. mosseae, G. fasciculatum, G. versiforme, A. laevis* and *Gi. margarita* in sugarcane crop. MIE was calculated using formula proposed by Bagyaraj *et al.* (1988).

$$\text{MIE} = \frac{\text{Dry weight of inoculated plant} - \text{Dry weight of uninoculated plant}}{\text{Dry weight of inoculated plant}} \times 100$$

Phosphatase activity of roots (Morton, 1952)

The phosphatase activity was measured in roots. The enzyme phosphatase hydrolysed para-nitrophenyl phosphate. The released p-nitrophenol was yellow in colour in alkaline medium and was measured at 725 nm. The optimum pH for acid phosphatase was 4.5 and for alkaline phosphatase were 8.5.

Enzyme extract

AM fungi inoculated 10 g of sugarcane roots were ground thoroughly with acid washed sand in a pre-chilled pestle and mortar in grinding medium containing 20 ml of 0.2 M acetate buffer (pH 4.5) for acid phosphatase or 0.2 M borate buffer (pH 8.5) for alkaline phosphatase. The homogenate was passed through four layers of cheese cloth and the filterate was centrifuged at 3000 rpm for five minutes. Supernatant was used as enzyme source.

Estimation of acid phosphatase

The substrate P-nitrophenyl phosphate of 1.0 g was dissolved in 100 ml distilled water. One ml of substrate was pipette out into a test tube and two ml of enzyme extract and five ml of 0.2 M acetate buffer (pH 4.5) were added. This was incubated for 24 h and one drop of 10 per cent TCA was added and centrifuged. From this one ml of clear supernatant was taken in a test tube. To this supernatant, one ml of folin ciocalteau reagent and 2ml of 20 per cent sodium carbonate were added and boiled for one minute (at 100°C). Then the test tube was cooled and the volume was made upto 10 ml with distilled water. The colour intensity was read at 725 nm (red filter). Standard curve using P-nitrophenol was drawn and from this the activity was calculated.

Estimation of alkaline phosphatase

Alkaline phosphatase activity was measured by adopting the procedure described for acid phosphatase. Except that here the borate buffer (0.2 M pH 8.5) was used instead of acid buffer.

RESULTS

Survey of AM fungal occurrence in sugarcane fields of Cuddalore District of Tamilnadu

The AM fungal colonization and spore load in the rhizosphere soil of sugarcane collected from Annamalainagar, Vilagam, Arasur, Cuddalore, Mutlur, Orathur, Pichavaram and Vallampadugai, Cuddalore District, Tamilnadu, India were recorded and presented in table 1. Among the locations Vallampadugai recorded maximum root colonization percentage and spore number and lowest values were recorded by Pichavaram

days after planting (DAP). Based on the percentage of root colonization, spore number 100 g^{-1} of rhizosphere soil, acid phosphatase and alkaline phosphatase activity, the efficient AM fungus was selected. The results are presented in Table 4. The sugarcane root colonization by AM fungi increased with the age of crop from 60^{th} to 120^{th} DAP. The per cent root colonization, spore number $100g^{-1}$ of rhizosphere soil, the acid and alkaline phosphatase enzyme activities were highest in *G. fasciculatum* inoculated plants compared with *G. mosseae, G. versiforme, A. laevis* and *Gi. margarita* inoculated plants. The highest per cent root colonization and spore number of sugarcane were recorded in *G. fasciculatum* inoculation (76.28, 181.48) followed by *G. mosseae* (62.23, 176.20),

Table 1. Survey of AM fungal occurrence in sugarcane fields of Cuddalore District of Tamilnadu

S. No.	Place of the sample	Soil texture	pH	EC mmhos cm^{-1}	Organic carbon	Available phosphorus content (kg ha^{-1})	Percent root colonization	AM spore population 100 g^{-1} of rhizosphere soil
1	Annamalainagar	Clay loam	8.1	0.43	0.51	17.48	48.0	98.0
2	Vilagam	Clay	7.5	0.41	0.46	16.71	50.5	109.0
3	Arasur	Clay loam	7.7	0.47	0.60	16.69	51.0	100.5
4	Cuddalore	Clay loam	8.0	0.50	0.63	11.18	40.5	80.0
5	Vallampadugai	Clay	7.5	0.41	0.59	18.59	62.8	128.0
6	Mutlur	Clay loam	7.4	0.36	0.44	20.04	40.0	84.4
7	Orathur	Sandy clay	7.5	0.34	0.54	18.48	48.0	100.5
8	Pichavaram	Sandy clay	8.5	0.53	0.76	19.38	34.8	67.0

Table 2. Characterization of different AM fungal isolates from sugarcane rhizosphere soil

S.No	Characters	*Glomus mosseae*	*Glomus fasciculatum*	*Glomus versiforme*	*Acaulospora laevis*	*Gigaspora margarita*
1	Size of spore	120 µm	100 – 120 µm	125 – 150 µm	400 µm	200 – 300 µm
2	Spore shape	Globose	Globose hypogeous	Globose	Globose	Ectocarpic
3	Colour of spore	Yellow to brown	Yellow to reddish brown	Yellow to brown	Outer wall – brown Inner wall – Hyaline Ellipsoid	White when young and slightly yellowish at maturity
4	Sporocarp	Present	Present	Present	Present	Absent
5	Thickness of spore wall	3 – 4 µm	4 – 14 µm	3 – 4 µm	4 – 8 µm	> 20µm
6	Subtending hyphae	Cylindric flared	Absent	Cylindric or flared	Not observable	Bulbous (30 – 50 µm)

Table 3. Influence of soil types on spore population of AM fungi

S. No.	Soil texture	Total AM fungal spore population per 100 g of soil in each soil types	Types of AM fungi				
			Glomus mosseae	*Glomus fasciculatum*	*Glomus versiforme*	*Acaulospora laevis*	*Gigaspora margarita*
1	Sandy Clay	75.0	15.0	35.0	5.0	8.0	12.0
2	Sandy loam	68.0	15.0	28.0	5.0	8.0	12.0
3	Clay loam	100.0	20.0	50.0	6.0	9.0	15.0
4	Clay	75.0	15.0	35.0	5.0	8.0	12.0

SE = 0.06; CD (p = 0.05) = 0.14

Influence of soil types on mycorrhizal spore population

Among the four different soil types, clay loam soil, recorded the highest spore population (100 in 100 g^{-1} of soil) followed by clay soil, sandy clay (75.0 in $100g^{-1}$ of soil) and sandy loam (68.0 in $100g^{-1}$ of soil) types. Among the five different AM fungal isolates *G. fasciculatum* was the predominant species in all the soil types followed by *G. mosseae*, *Gi. margarita, A. laevis* and *G. versiforme* (Table 2 and 3).

Screening of five different AM fungi in sugarcane (CoSi 98071)

Pot culture experiments were carried out to study the effect of the five isolates of AM fungi *viz., G. mosseae, G. fasciculatum, G. versiforme, A. laevis* and *Gi. margarita* on the root colonization of sugarcane var. CoSi 98071 on 60, 90 and 120

G. versiforme (47.24, 164.48), *A. laevis* (56.22, 170.93) and *Gi. margarita* (52.21, 166.75). The highest acid and alkaline phosphatase activities were recorded in *G. fasciculatum* inoculated roots (28.78, 27.38 µg 24 hrs^{-1} 10 g^{-1} of root) followed by *G. mosseae* (28.58 and 26.60) and others between 25.50 to 28.78 and 22.25 to 27.38 respectively for acid and alkaline phosphatase activity. In general the acid phosphatase activity was more, compared to alkaline phosphatase activity. The values obtained in *G. mosseae* and *Gi. margarita* and *G. versiforme* were on par.

Studies on the interaction of sugarcane (CoSi 98071) and AM fungi

In general, the sugarcane var. CoSi 98071 grew better in unsterilized soil compared to sterilized soil. The relative mycorrhizal dependency (RMD) for sugarcane var. CoSi

Table 4. Screening of different AM fungal species in Sugarcane var CoSi 98071 under pot culture study

S.No.	AM fungal inoculation	Percent root colonization			Spore number /100g of rhizosphere soil			Acid phosphatase activity (µg/ 24 hrs^{-1}. 10g^{-1} of root)			Alkaline phosphatase activity (µg/ 24 hrs^{-1}. 10g^{-1} of root)		
		Sampling period in days			Sampling period in days			Sampling period in days			Sampling period in days		
		60	90	120	60	90	120	60	90	120	60	90	120
1	*Glomus mosseae*	40.25	55.43	62.23	143.23	165.98	176.20	25.58	26.63	28.58	24.34	25.62	26.60
2	*Glomus fasciculatum*	52.23	62.30	76.28	152.20	169.45	181.48	26.50	27.78	28.78	25.73	27.01	27.38
3	*Glomus versiforme*	25.30	37.25	47.24	126.75	157.01	164.48	22.45	24.25	25.50	21.22	21.90	22.25
4	*Acaulospora laevis*	36.57	49.33	56.22	138.53	162.95	170.93	24.38	25.45	27.28	23.01	24.14	24.48
5	*Gigaspora margarita*	29.28	42.40	52.21	131.48	157.98	166.75	23.55	25.03	26.28	21.60	22.70	22.67
	SE	2.59	2.12	2.02	2.27	0.52	1.14	0.31	0.17	0.33	0.45	0.47	0.61
	CD (p = 0.05)	7.24	6.03	5.75	6.49	1.50	3.28	0.89	0.51	0.95	1.31	1.35	1.76

Table 5. Studies on the interaction between sugarcane (CoSi 98071) and AM fungi

S.No.	Treatments		Plant dry weight on 150 DAP
1	Sterilized soil		7.57
2	Unsterilized soil		10.98
3	Unsterilized soil + *G. mosseae*		13.51
4	Unsterilized soil + *G. fasciculatum*		13.80
5	Unsterilized soil + *G. versiforme*		12.89
6	Unsterilized soil + *A. laevis*		13.34
7	Unsterilized soil + *Gi. margarita*		13.13
	SE		0.03
	CD (p = 0.05)		0.09
a	Relative mycorrhizal dependency (%)		31.80
b	Mycorrhizal inoculation effect of	*G. mosseae* (%)	18.88
c	Mycorrhizal inoculation effect of	*G. fasciculatum* (%)	20.58
d	Mycorrhizal inoculation effect of	*G. versiforme* (%)	14.89
e	Mycorrhizal inoculation effect of	*A. laevis* (%)	17.70
f	Mycorrhizal inoculation effect of	*Gi. margarita* (%)	16.46

Table 6. Effect of different organic amendments on the mycorrhizal colonization (*G. fasciculatum*) and growth of sugarcane (CoSi 98071) in pot culture

S.No.	Organic amendments	Percent root colonization			Spore number/100 g soil			Height of the cane (cm)			Girth of the cane (cm)		
		Sampling period in days			Sampling period in days			Sampling period in days			Sampling period in days		
		60	120	180	60	120	180	60	120	180	60	120	180
1	Control	12.21	31.06	40.19	115.19	125.85	137.80	42.10	89.30	109.74	4.56	7.25	9.74
2	Farm yard manure	15.28	58.89	58.94	142.14	149.29	157.81	69.44	110.51	137.32	6.86	9.52	10.87
3	Groundnut cake	15.00	36.38	48.20	127.82	135.84	144.82	51.43	96.54	125.31	5.73	7.84	10.07
4	Neem cake	15.00	36.18	48.11	127.47	135.23	143.13	50.04	96.35	123.93	5.54	7.86	10.00
5	Press mud	15.18	48.18	52.21	128.02	138.17	147.15	55.26	99.36	127.64	5.96	8.02	10.22
	SE	1.30	1.43	1.46	1.64	1.10	1.05	1.53	1.18	0.91	0.08	0.17	0.06
	CD (p = 0.05)	4.00	4.07	4.12	4.71	3.15	2.97	4.34	3.37	2.54	0.21	0.48	0.15

98071 was 31.80 per cent. Based on this, sugarcane was considered moderately dependent on arbuscular mycorrhiza.

The effect of inoculations of five different AM fungi was *G. mosseae, G. fasciculatum, G. versiforme, A. laevis* and *Gi. margarita* was compared with uninoculated unsterilized soil on shoot dry weight of sugarcane var. CoSi 98071 was studied in a pot culture experiment. Based on shoot dry weight at 150 DAP, the mycorrhizal inoculation effect (MIE) of the five different AM fungal cultures were recorded. The data are presented in Table 5. All the five AM fungal cultures inoculated in sugarcane var. CoSi 98071 significantly increased the shoot dry weight over uninoculated in unsterile soil. The mean mycorrhizal inoculation effects (MIE) of *G. mosseae, G. fasciculatum, G. versiforme, A. laevis* and *Gi. margarita* were 18.8, 20.5, 14.8, 17.7 and 16.4 respectively. The inoculation of *G. fasciculatum* recorded the highest MIE among the five AM cultures tested and selected for further studies. Based on the root colonization percentage, spore number, acid and alkaline phosphatase activities, RMD and MIE *G. fasciculatum* was found to be efficient and selected for further studies.

Effect of different organic amendments on the growth parameters and mycorrhizal colonization (*G. fasciculatum*) of sugarcane (CoSi 98071) in pot culture

In general, all organic amendments showed positive influence on the proliferation of AM fungi (Table 6). Among different organic amendments farm yard manure (FYM) significantly stimulated the highest per cent root colonization and spore number (56.94 and 157.81) followed by press mud (52.21 and 147.15), coconut cake (48.20 and 144.82) and neem cake (48.1 and 143.13) on 180 DAP. The corresponding trend was observed in plant parameters *viz.,* plant height, girth of the cane (Table 6). The significant increase in plant height (137.32 cm) and cane girth (10.87) were recorded in FYM amended treatment followed by press mud, coconut cake and neem cake on 180 DAP. The results obtained in press mud and neem cake were on par.

DISCUSSION

AM fungi is the most abundant kind of mycorrhizae found in association with every taxonomic group of plants and the list

of species not infected is probably far shorter than the infected ones and in the current status these fungal and bacterial associations are beneficial to crop plants in many ways, including enhancing the nutrient availability especially nitrogen and phosphorus, enhance water uptake and induces resistant against diseases and increased the yield (Lekberg and Koids, 2005; Hu *et al.,* 2009).

Survey for the occurrence of AM fungi

The results of the present survey in thirteen different locations in Cuddalore District of Tamil Nadu State of India, where sugarcane is grown as a commercial crop revealed the ubiquitous nature of AM fungi in sugarcane rhizosphere soil and the occurrence of AM fungi in soils has been reported in various kinds of environments (Koide and Mosse, 2004; Rilling, 2004, 2006; Bouwmeester *et al.,* 2007; Cho *et al.,* 2009). The rhizosphere soil and root samples collected from wheat and lentil at 11 sites across four zones of Saskatchewan in Canada were analysed for spore number, level of AM fungal colonization and AM fungal species. The number of spores ranged from 78 to 272 100 g^{-1} soil. The level of colonization varied from site to site and the difference were more pronounced in wheat than lentil (Talukdar and Germida, 1993; Begum and Priya, 2004). The root colonization percentage in sweet potato ranged from 30 to 70 per cent (Sathiyavathi, 1996) and in cotton ranged from 40-80 (Jayanthi, 2008). Interestingly in the present study, the root colonization percentage and spore number of AM fungi in the rhizosphere samples and sugarcane in the range of 34.8 to 62.8 and 67.0 to 128.0 100 g^{-1} of soil respectively. The eight locations comprised of four different soil types i.e., sandy clay, sandy loam, clay loam and clay. Although there was no apparent relationship between soil characteristics (pH, EC and organic carbon) and AM fungal root colonization and spore number was noticed. The available phosphorus inversely correlated with AM fungal root colonization and spore number. These results are in accordance with the findings of Miranda and Harris (1994a) in leek and Chandrasekara *et al.* (1995) in sunflower.

Soil texture and AM spore population

In the present study, the four different AM fungi were isolated from sugarcane rhizosphere soil and characterized as *Glomus mosseae, Glomus fasciculatum, Glomus versiforme, Acaulospora laevis and Gigaspora margarita*. In the present investigation in all the soil type were qualitatively and quantitatively enumerated. *G. fasciculatum* was predominant in all the soil types and the highest spore population was observed in clay loam soil (Hu *et al.,* 2009).

Screening AM fungal cultures

AM fungal colonization in the roots of sugarcane may be due to fungal preference by the host and due to the factors influencing the mycotrophy of sugarcane. Mallosha *et al.* (1994) worked on the selection of efficient AM fungi for tomato. Tomato seedlings inoculated with *Glomus leptotichum* recorded higher root colonization percentage, growth and yield parameters than *G. intraradices*. The results of the present investigation under pot culture revealed that among the five AM fungal species tested, *G. fasciculatum* was found to the most effective AM fungus for colonizing the roots of sugarcane (CoSi 98071) (Speir *et al.,* 2004). The root colonization percentage was 76.28 the number of spores were 181.48 100 g^{-1} of soil and the acid and alkaline phosphatase activities were 28.78 and 27.38 µg of phenol released 24 h^{-1} 10 g^{-1} of fresh root. Though a particular AM fungi can colonize many host plants, it has a preferred host which exhibits maximum symbiotic response when colonized by that particular AM fungal species (Bagyaraj, 1989; Jayanthi, 2008).

Interaction of sugarcane and AM fungi

Species and strains of AM fungi have been shown to differ in enhancing nutrient uptake and plant growth (Simpson and Daft, 1990; Hu *et al.,* 2009). Variations in the extent and effect of AM fungal colonization have also been linked to the genotype of the host plant (Lackie *et al.,* 1988; Gai *et al.,* 2006). The recent isolation of mycoplant mutants and the discovery that AM fungal colonization was a heritable trait (Mercy *et al.,* 1990) suggest the possibilities of tailoring plant-fungus combinations for maximum efficiency. Plant differs greatly in their need for a response to mycorrhizal infection. Gerdemann (1975) defined relative mycorrhizal dependency (RMD) as the degree to which a plant is dependent on the mycorrhizal condition to produce its maximum growth or yield at a given level of soil fertility. Plenchette *et al.* (1982) proposed another formula to calculate relative mycorrhizal dependency (RMD) of crop plants under field conditions. They compared plants in fumigated and non-fumigated soil. In the present study, the relative mycorrhizal dependency of the crop was 31.80 per cent. The sugarcane is a moderately dependent crop to AM fungi. In the present pot culture study, sugarcane var. CoSi 98071 was highly responsive to *G. fasciculatum* inoculation with 20.15 per cent mycorrhizal inoculation effect (MIE) compared to *G. mosseae* (18.8%), *A. laevis* (17.7%), *Gi.margarita* (16.4%) and *G. versiforme* (14.9%). This clearly established the sugarcane CoSi 98071 and *G. fasciculatum* interaction increased the growth (MIE). Among the different AM fungi, the genus *Glomus* was more effective AM fungal symbiosis to sweet potato than *Acaulospora* or *Scutellospora* (Gai *et al.,* 2006).

Effect of different organic amendments on the growth parameters and mycorrhizal colonization (*G. fasciculatum*) of sugarcane

Organic amendments influence proliferation of AM fungi (Geethakumari *et al.,* 1990; Hu *et al.,* 2009). VA-mycorrhizal inoculation and addition of organic manure increased plant growth and increased shoot dry weight, N and P uptake of wheat cv cyiza 157 plants (Mikhaeel *et al.,* 1997). Organic amendments with narrow C:N ratio and moderate N, P and C like Farmyard manure (FYM) have shown a more pronounced effect on AM fungal proliferation. The highest root colonization percentage (100, 98) and spore number (307, 274.50 g^{-1} of soil) were observed in inoculation of *G. fasciculatum* in conjunction with FYM for wheat genotypes DWR-39 and DWR-187, than biogas spent slurry, groundnut cake, glyricidia, jowar straw and rice bran. Interestingly in the present study, the highest root colonization percentage (58.94) spore number (157.84), cane height (137.2 cm) and cane girth (10.87 cm) were observed in inoculation of *G. fasciculatum* with FYM followed by press mud, groundnut cake and neem cake.

Summary and Conclusion

Eight locations were selected in the Cuddalore district of Tamil Nadu, to study the occurrence of AM fungi in sugarcane soils. The survey revealed the presence of AM fungi in the sugarcane soils. The root colonization and spore number 100 g^{-1} of soil ranged from 34.8 to 62.8 and 67.0 to 128.0 respectively. The sample collected from Vallampadugai recorded the highest root colonization percentage and spore number (62.8 and 128.0) and Pichavaram recorded the least (34.8 and 67.0). The physico-chemical properties have no apparent correlation with AM fungal root colonization percentage and spore number, whereas the soil available P, negatively correlated with AM colonization and spore number. Four different soil types were observed in the thirteen different locations viz., sandy clay, sandy loam, clay and clay loam. Five different AM fungi were isolated from sugarcane rhizosphere soils collected from each of the above mentioned thirteen locations and they are identified as *Glomus mosseae, G. fasciculatum, G. versiforme, Acaulospora laevis* and *Gigaspora margarita* according to Gerdemann and Trappe (1974). Among the five different soil types, clay loam recorded highest number of AM fungal spores 100 g^{-1} of soil and *G. fasciculatum* was predominant in all the soil types. Among the five different AM fungal isolates, *G. fasciculatum* recorded highest root colonization percentage (76.28), spore number (181.48/100g^{-1}), acid and alkaline phosphatase enzyme activities respectively (28.78, 27.38 µg 24 h^{-1} 10 g^{-1} of root) in pot culture experiment. The growth of sugarcane in sterilized and unsterilized soils was compared in pot culture conditions so as to determine relative mycorrhizal dependency (RMD) of this crop. The sugarcane grew better in unsterilized soil compared to sterilized soil. The RMD was found to be 31.80 per cent and sugarcane considered as moderately dependent plant for mycorrhiza. Effect of inoculation of five different AM fungi was estimated as MIE. Among the five AM fungi tested, *G. fasciculatum* recorded higher MIE (20.58) than other AM cultures. The effect of different organic amendments, farm yard manure (FYM), groundnut cake (GNC), pressmud (PM) and neem cake (NC) on the growth parameters and mycorrhizal colonization of sugarcane var. CoSi 98071 was studied in pot culture. Among the different organic amendments, FYM recorded highest per cent root colonization (58.94), spore number (157.81), plant height (137.32cm) and can girth (10.87 cm) on 180 DAP. The investigation conducted in the pot culture house of Department of Microbiology, clearly indicated that the AM fungi *G. fasciculatum* was found to be a best species in all the beneficial characters and also *G. fasciculatum* along with different organic amendments was found to influence the growth and development of sugarcane. Hence, these types of studies need to exploit under field condition.

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