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Variation of Native VA-Mycorrhizal Association on Cultivated Species of Mint

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Abstract

Variation in the responses of six cultivated species of mint (*Mentha arvensis* subsp. *haplocalyx*, *Mentha citrata*, *Mentha piperita*, *Mentha spicata*, *Mentha cardiaca* and *Mentha viridis*) to the colonization by VA-mycorrhizal (VAM) fungi are reported. All species of mint had abundant VAM associations. Four species of *Glomus*, one species of *Entrophospora* and one species of *Sclerocystis* were isolated from the rhizosphere soil of these plants. Root colonization varied from 37.2% to 56.0%. The highest level of VAM colonization (56.0%) was observed on the roots of *M. spicata* and *M. citrata*. Rhizosphere soil of these plants had a VAM spore population ranging from 416 to 707, 100 g^{-1} soil. The highest VAM spore population was observed in the rhizosphere of *M. spicata*. The results showed that cultivated species of mint have significant variation in their responses to native VAM fungi. This is the first report of the occurrence of *Glomus aggregatum*, *Glomus fasciculatum*, *Glomus mosseae*, *Glomus geosporum*, *Entrophospora* sp. and *Sclerocystis* sp. on mints.

Keywords: VA-mycorrhizae, mint, *Glomus aggregatum*, *Glomus fasciculatum*, *Glomus mosseae*, *Glomus geosporum*, *Entrophospora*, *Sclerocystis*

1. Introduction

Mints (*Mentha* species) are important essential oil bearing plants and are cultivated on a large scale in different parts of the world. Major commercially

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produced species are *Mentha arvensis* subsp. *haplocalyx* Briquet (Japanese mint), *Mentha piperita* L. (Peppermint), *Mentha spicata* L. (Spearmint), *Mentha citrata* Ehrh. (Bergamot mint), *Mentha cardiaca* (S.F. Gray) Baker (Scotch spearmint) and *Mentha viridis* L. (Garden mint). Essential oils of these plants contain menthol, menthone, linalyl acetate, linalool and carvone. These aromatic compounds are extensively used in food, medicine, perfumery and cosmetics. A number of *Mentha* species are cultivated on a large scale in Europe, Australia, China, Japan, United States, India, South America and parts of Africa. India is one of the major mint-growing countries of the world. Mints grow luxuriantly when the crop gets moderate and regular rainfall during its growing season (Husain et al., 1988).

Vesicular-arbuscular mycorrhizal (VAM) fungi are present nearly in all soils and these fungi colonize roots of the great majority of plants. The beneficial effect of this symbiotic association is of special importance for those plants having coarse or poorly developed root systems (Howeler et al., 1987). Although essential oil producing plants are cultivated throughout the world, no serious attempt had been made to determine the mycorrhizal associations of these plants and the applications of VAM fungi to improve their productivity. Recent investigations have shown the beneficial effect of the VAM association on the growth and biomass production of *Cymbopogon martinii* (Roxb.) Wats var. *motia* (palmarosa), an important essential oil bearing plant (Gupta et al., 1990; Gupta and Janardhanan, 1991). In this communication, we report the occurrence of VAM fungi on six species of mint growing in the experimental farm of Central Institute of Medicinal and Aromatic Plants (CIMAP) and the extent of variation of mycorrhiza development by native VAM fungi in these species.

2. Materials and Methods

Six species of *Mentha* (*M. arvensis*, *M. piperita*, *M. spicata*, *M. cardiaca*, *M. citrata* and *M. viridis*) growing in the experimental farm of CIMAP were screened for VAM association during the month of August when these plants were 7-months old and ready for a second harvest. Samples of rhizosphere soil along with the fine roots were collected for each plant species using the pattern of randomized complete block design (RCBD) with five replications. Roots were separated, washed and chopped into pieces approximately 1 cm in length. The roots were treated with 10% KOH and stained by the method of Phillips and Hayman (1970). Colonization by VAM fungi was estimated by the method of Biermann and Linderman (1981) using 40 to 50 stained root segments. The colonized portion of each root segment was estimated to the nearest 10% under a microscope. At least five determinations were done for each sample of each plant species.

Soil samples (100 g) were collected from the rhizosphere of each plant species. VAM spores were extracted by wet sieving and decanting method of Gerdemann and Nicolson (1963). The spores were suspended in 50 mL water and counted in a well shaken 1 mL sample. The process was repeated 10 times and the number of VAM spores present in each 100 g soil sample was determined. At least 5 samples of each plant species were used for each determination. The spores recovered from soil suspension were preserved in lactophenol (lactic acid : phenol : glycerol : water - 1:1:2:1). Spores (50 to 100) were drawn from the suspension and identified according to Schenck and Perez (1988). Differences between mean per cent root colonization and mean chlamydospore population in rhizosphere soil (100 g^{-1}) amongst the six species of mint were statistically analysed using critical difference (CD) test at 5% and 1% levels of significance.

Soil and root samples collected in August 1989 were examined during this study. Soil samples were analysed according to Jackson (1973). Soil characteristics were sandy loam, pH 7.5±0.21, mineralizable N 85±2.39 μg/g, available P 14.6±0.98 μg/g, available K 48±0.89 μg/g and organic carbon 1.73±0.08%. Available P was estimated according to Olsen et al. (1954).

3. Results and Discussion

All the *Mentha* species examined were found to be colonized by VAM fungi (Fig. 1A, B, C, D, E, F). The range of colonization varied from 37.2% (in *M. viridis*) to 56.0% (in *M. citrata* and *M. spicata*) (Table 1). Davis et al. (1984) found an average root colonization of 26.5% in peppermint (*M. piperita*). However, 100% VAM colonization in Japanese mint (*M. arvensis*) and 94% in peppermint (*M. piperita*) growing in red sandy loam soil was observed by Govind Rao et al. (1989). In most of the samples examined in the present study, a smaller number of arbuscules was observed than vesicles. Arbuscule formation is most prominent during the vegetative growth (Saif and Khan, 1975). Mints often have poorly developed root systems. The major part of the root system consists of older roots and abundant vesicles are normally found in older roots.

The largest number of chlamydospores of VA-mycorrhizal fungi was observed in the rhizosphere soil of *M. spicata* (700/100 g^{-1} soil). However, no relationship between the number of VAM spores present in the rhizosphere soil and per cent root colonization could be found for any of the mint species examined except *M. spicata* (Table 1). Although Govind Rao et al. (1989) observed 94 to 100% root colonization in *M. piperita* and *M. arvensis*, they found only a

Table 1. Colonization on *Mentha* species by vesicular-arbuscular mycorrhizal fungi under field conditions

	Per cent root coloni-zation	Chlamydospores/ 100 g dry rhizosphere soil	Vesicles/ arbuscules	ENT	GA	GF	GG	GM	SCL
Mentha arvensis	49.2	582	++/++	+	++	++	+	++	+
M. cardiaca	44.0	663	++/+	−	++	++	+	+	−
M. citrata	56.0	545	+++/+++	−	++	++	−	−	−
M. piperita	45.2	520	++/+	−	++	++	−	+	−
M. spicata	56.0	707	+++/++	−	++	++	+	++	+
M. viridis	37.2	416	++/+	−	++	++	−	−	+
C.D. 5%	2.368	30.086							
C.D. 1%	3.229	41.033							

Abbreviations: ENT: *Entrophospora* sp., GA: *Glomus aggregatum*, GF: *Glomus fasciculatum*, GG: *Glomus geosporum*, GM: *Glomus mosseae*, SCL: *Sclerocystis* sp., −: absent, +: scanty, ++: moderate, +++: high.

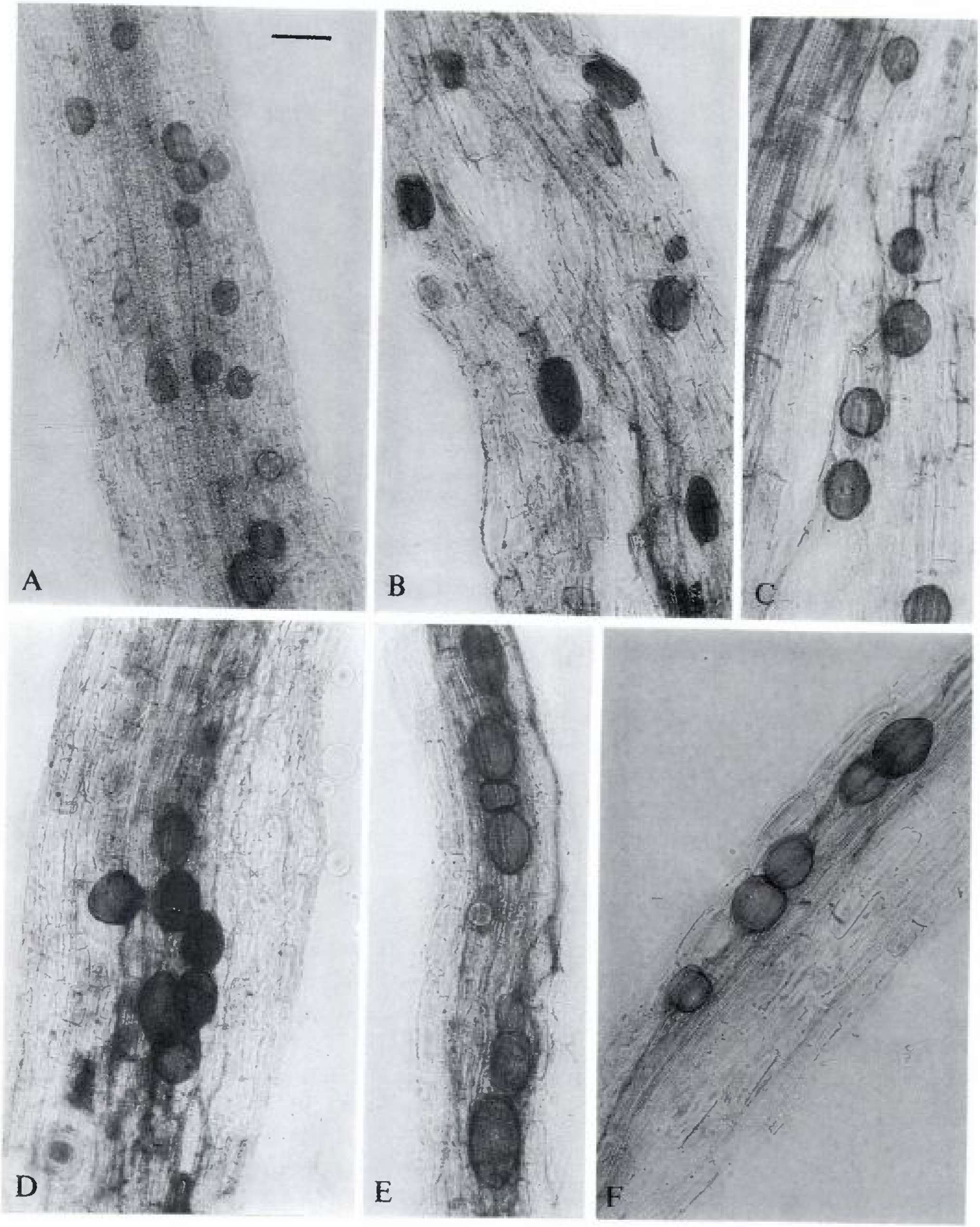

Figure 1. Colonization of *Mentha* roots with native VAM fungi: (a) *Mentha arvensis*, (b) *M. cardiaca*, (c) *M. citrata*, (d) *M. piperita*, (e) *M. spicata*, (f) *M. viridis*. Bar = 50 μm.

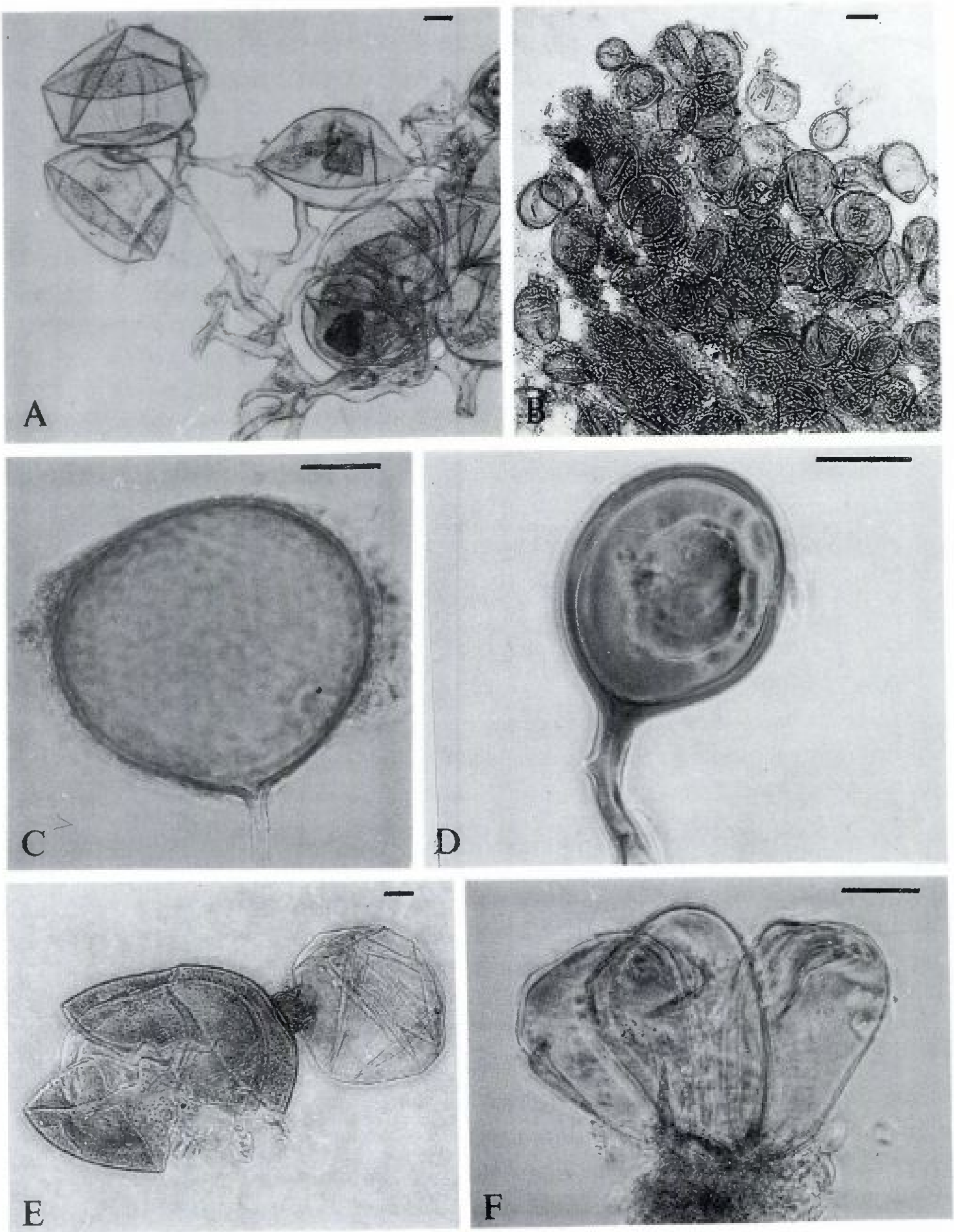

Figure 2. (a) *Glomus aggregatum*, (b) *Glomus fasciculatum*, (c) *Glomus mosseae*, (d) *Glomus geosporum*, (e) *Entrophospora* sp., (f) *Sclerocystis*. Bar = 20 μm.

small number of spores (16 to 34/22 g^{-1} soil) in the rhizosphere soil of these plants.

Microscopic examination revealed that the extramatrical spores present in the rhizosphere soil of mint species belonged to three genera of VAM fungi, namely, *Glomus*, *Entrophospora* and *Sclerocystis*. These VAM fungi were identified as *Glomus aggregatum* Schenck & Smith, *G. fasciculatum* (Thaxter) Gerdemann and Trappe emend. Walker & Koske, *G. geosporum* ((Nicolson & Gerdemann) Walker, *G. mosseae* (Nicol. & Gerd.) Gerdemann & Trappe, *Entrophospora* sp. Ames and Schneider and *Sclerocystis* sp. Berk. & Broome (Schenck and Perez, 1988) (Fig. 2). *Glomus aggregatum* and *G. fasciculatum* were found to be the most predominant species distributed among mints. *Glomus mosseae* was found to be associated with *M. spicata*, *M. arvensis* and to a lesser extent with *M. cardiaca* and *M. piperita*. *G. geosporum* showed only scanty presence in the rhizosphere. *Entrophospora* sp. was found associated only with *M. arvensis*. However, *Sclerocystis* sp. rarely occurred. Davis et al. (1984) reported four *Glomus* spp., one *Acaulospora* sp. and one *Gigaspora* sp. in the rhizosphere soil of peppermint plants growing in fields with P level of 44 to 244 ppm. However, this is the first report of occurrence of *G. aggregatum*, *G. fasciculatum*, *G. mosseae*, *G. geosporum*, *Entrophospora* sp. and *Sclerocystis* sp. on mints. These studies also constitute the first report of the quantitative variation of native VAM associations with field grown mint plants.

The results of the present investigation show that *Mentha* species are associated with several VAM fungi, indicating that colonization by more than one VAM fungus could be beneficial to the plants under field conditions. Daft and Hogarth (1983) observed more consistant benefits to plant growth from a combination of four *Glomus* species than from any single species. However, *Glomus aggregatum* and *G. fasciculatum* occurred abundantly on all mint species while occurrence of *G. mosseae* was found only with *M. arvensis* and *M. spicata*, suggesting some degree of host preference.

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