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RESEARCH ARTICLE

DIVERSITY OF AM FUNGI OF *Centella asiatica* L IN KANYAKUMARI DT., OF TAMIL NADU

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Abstract

A study on diversity of vesicular-arbuscular mycorrhizal fungi associated with *Centella asiatica* L (Kondangal or Vallari) plant root colonization and rhizosphere soils were carried out at rainy season consecutively. They impact of physical and chemical factors of rhizosphere soils in relation to the quantitative and qualitative assessment of AM fungi in Kanyakumari district of Tamil nadu. To found twelve species of AM fungi belonging to five genera viz: *Glomus*, *Aculospora*, *Sclerocystis*, *Gigaspora*, and *Scutellospora* were recorded and identified from rhizosphere soils. The most frequent most genera were found *Glomus mossea* and *Gigaspora margarita* noticed in the *Centella asiatica* rhizosphere soil.

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INTRODUCTION

The genus *Centella asiatica* (Indian pennywort) is one of the important aromatic group of herbaceous plant belonging to the family Apiaceae. Many species of *Centella* contain various economically important essential oils used in perfumery and cosmetic industries. *Centella asiatica* has wide application in Indian and Chinese traditional medicines with documented evidence for wound healing and neuroprotective and anti-aging potential. Asiaticoside, a trisaccharide triterpene, is the most medicinally active compound in the plant. β -Amyrin synthase and squalene synthase have been identified as the two key genes in the triterpenoid pathway which regulate the production of asiaticoside in *C.asiatica* (Jisha *et al.*, 2011).The genus of *Centella* native to India, Northern Australia, and Sri Lanka. The certain species of *Centella* has the antiviral, antibacterial, anti-inflammatory, anti-ulcerogenic and anxiolytic properties too.

Besides from the industrial point of view, the isolated steriods from the plant have been used to treat leprosy (Hausen 1993), and there reports of potential application of the plant in re-vitalization of the brain and nervous system (Brinkhouse and Linder 2000), protection by directly or indirectly modulating the act of ATPase (Visweswari *et al*, 2010), and in companging (Bradwejn and Zhou 2000). Asiaticoside, a trisaccharide triterpene, has been identified as the most compound in the plant, which also possesses other bioactive triterpenoid glycosides (asiatic acid, madeecassic acid), saponin glycosides (Brahmoside, Brahminioside), and flavonoids (Maquart *et al.*, 1999).

VAM is the most abundant kind of mycorrhiza described as 'a universal plant symbiosis' The mycorrhizae are the feeder root of plant growing in natural world and are beneficial to their host plant. The plant's root with zone of powerful microbial metabolic activity occurring where, there is a high concentration of carbon is called the rhizosphere (Barea, *et al.*, 2005). For plant, seedling growth of non-mycorrhizal plants requires extremely high levels of readily-soluble P fertilizers. In non-mycorrhizal plants, the critical deficiency level of available soil P is 190 mg, compared with only 15 mg in mycorrhizal plants. The cost of production of chemical fertilizers, the agricultural planners are compelled to reorient their approach towards cost effective, cheap, and environmentally friendly source to supplement chemical fertilizers (Charles, *et al.*, 2008).

Materials and methods

Selection of study sites.

AM fungal spores were isolated from rhizosphere soil of *Centella asiatica* L. plant collected from Kanyakumari district, Tamilnadu state, India. Site selection was based on soil characters. They were Agasteeswaram, Killiyoor, Vilavancode.

Collection of soil and root samples.

Plants were surveyed for colonization by AM fungi at three different study sites of Kanyakumari district. At each study site all area of 3m² was chosen for sampling. Both the study site plant root sample and rhizosphere soil were selected for the study. The soil samples were examined for AM spore numbers after wet sieving and decantation (Gerdemann and Nicolson, 1963). The root samples were stained with trypan blue in lactophenol (Phillips and Hayman, 1970). Percentage of root colonization was calculated (Krishna and Dart, 1984). The isolated AM fungal species were brought in to pot culture studies with plants of *Allium cepa* L. After 90 days, the spore and sporocarps were reisolated for identification (Schenck and Perez, 1988). Study site soils were examined by standard physico-chemical method (Jackson, 1973).

Results and Discussion

Soil physico-chemical characteristics of the three study localities were found to be red soil, sandy clay and laterite soil. Three different localities namely, Agasteeswaram, Killiyoor and Vilavancode were chosen for AM studies (Table 1). Some researchers worked on the characteristics of soil in different places are Alexander (1974), Griffin (1972), Charles *et al.*, (2008) and Selvaraj, *et al.*, (2011). All these study sites were acidic soil and generally deficient in phosphorus. The other soil elements showed only minor variations.

The plant species were positive for AM colonization in the roots of all the study sites. The root zone soils of Vallarai plants harboured AM fungal structures. The AM structures were including sporocarps and spores belonging to different AM fungal species. The AM fungal species isolated from the study sites belonging to five genera viz., *Glomus*, *Sclerocystis*, *Acaulospora*, *Gigaspora*, *Scutellospora*. (Table 2). Similar results were observed in different medicinal plants (Selvaraj, 1989) and *Hedychium Coronarium* (Thomas, *et al.*, 2014).

Totally, twelve AM fungal species were isolated from the rhizosphere soils of *Centella asiatica* collected from three different localities (Table 2). They were mass multiplied using suitable compatible host *Allium cepa* L. by pot culture method for identification. The significance of AM in plant ecology is based on its widespread occurrence in natural ecosystems (Bergelson, *et al.*, 1988; Kavitha and Nelson, 2013; Hussain and Srinivas, 2013). Two species of dominant AM fungal species such as *Gigaspora margarita* and *G. mosseae* root zone soils of Vallarai. In the present study indicates a predominance of *Glomus* over other genera isolated. Similar observation has been made by Regupathy and Mahadeven (1993), Muthukumar and Udayan (2000). The possible reason for the predominance of *Glomus* is known to be more common in natural and slightly alkaline soils (Mukerji *et al.*, 2002). The number of spores in root zone soil ranged from 142 to 482. There was an impact of soil physico-chemical characters on the distribution of AM spores in rhizosphere soil sites. The numbers of AM spores were more in site Agasteeswaram, whereas the lowest occur in Vilavancode (Table 2). There was a certain specificity among the different sites. Variation in spore density and colonization of AM associated with different host plant species may be generated by a variety of mechanisms, including variation in host species and their phenology, mycorrhizal dependency, host mediated alterations of the soil microenvironment or other host plant traits (Eon *et al.*, 2000; Lorgio *et al.*, 1999).

At the three study sites, all the plants were mycorrhizal. The present root colonization among the plants ranged from 58.2 % in Vilavancode to 98.5 % in Agasteeswaram.

The results suggested that the variation in soil pH, temperature and other factors seems to be the decisive factors in tropical soils influencing distribution of AM fungi. It can be concluded that physico-chemical factors significantly alter the distribution of native AM fungi both quantitatively and qualitatively.

Table 1
Physico - Chemical Characteristic of different rhizosphere soil sample of *Centella asiatica*.

S.No	Study Type	Soil type	pH	EC (dsm-1)	Organic matter (%)	Macro nutrients g/kg			Micronutrients in ppm			
						N	P	K	Zn	Cu	Fe	Mn
1.	Agasteeawaram	Red Soil	5.9	1.92	8.0	0.340	0.004	0.196	0.82	0.28	3.8	0.93
2.	Killiyoor	Sandy clay	6.8	1.41	8.5	0.380	0.015	0.210	0.66	0.21	4.1	1.51
3.	Vilavancode	Laterite	6.4	1.31	9.0	0.102	0.023	0.095	0.50	0.14	3.62	2.64

ppm- parts per million ** General nutrient status of the soils (Anonymous, 1988)

	N	p205	K20
Low	< 140	< 24.2	< 140.7
Medium	141 - 280	24.3 - 32.2	140.8 - 281.6
High	> 280	> 32.3	> 281.6

Table 2

Per cent root colonization, spore density and species richness of VAM fungi in *Centella asiatica*, rhizosphere soils

Study Sites	Root Colonization (%)	Total number of VAM fungal Spores/100g soil	Associated VAM fungal species	Positive for AM fungi in the roots
Agasteeswaram	98.5	482 + 10.8	ABRT, LAGR, LABS, LAGR, LDST, LFSE, CHTH, LMSS	<i>Glomus aggregatum</i>
Killiyoor	75.6	384 + 8.2	LABS, LAGR, LDST LFSC, GMRG, CHTG, SSNS, LMSS	<i>Glomus mosseae</i>
Vilavancode	58.2	142 + 4.2	ABRT, ASCB, LFSC, LAGR, LABS, LMSS	<i>Glomus mosseae</i>

** Unique code for VAM fungal species (Schenck and Perez, 1990)

ABRT- *Acaulospora bireticulata*

GMRG- *Gigaspora margarita*

LMRG- *Glomus macrocarpum*

SSNS- *Sclerocystis sinuosa*

LDST- *Glomus deserticola*

MCM- *Glomus macrocarpum*

ASCB- *Acaulospora scrobiculata*

LABS - *Glomus ambisporum*

CHTG - *Scutellospora heterogama*

LAGR - *Glomus aggregatum*

LFSC - *Glomus fasciculatum*

LMSS - *Glomus mosseae*

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