RESEARCH ARTICLE

Diversity of AM Fungi in *Sesamum indicum* L. from Sanjay Gandhi National park, Borivali, Mumbai, Maharashtra, India

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ABSTRACT

The present study was carried out to evaluate the status of Arbuscular Mycorrhizal Fungi (AMF) colonizing the rhizosphere of *Sesamum indicum* L. by setting up soil trap culture and to compare the species diversity in trap cultures with natural conditions. *Sesamum indicum* L. is a common wild medicinal herb, belonging to family Pedaliaceae. The study revealed that spore number was amazingly high in natural conditions. It was found to be 135spores/10g soil. The trap cultures after 3 months showed only 90 spores as against expected. The type of spores found in the natural conditions belonged to ten different types of species .The spores isolated from trap cultures belonged to only seven species. The spores isolated from the natural rhizospheric soil belonged to *Glomus geosporum, Glomus callosum, Glomus clarum, Glomus tortuosum , Acaulospora myriocarpa, Ambispora appendicula, Scutellospora sp. Gigaspora margarita, Glomus clavisporum, Kuklospora sp* . The spores isolated from trap cultures belonged to *Glomus halonatum, G. constrictum , G.aureum, Gigaspora decipiens , Ambisporum, Pacispora boliviana and Acaulospora sp*. The study of trap culture demonstrated that all AM Fungi do not sporulate in three months period , but had good amount of root colonization percentage. The sections of roots from trap cultures showed plenty of fungal hyphae, vesicles and arbuscules. Other endophytes with swollen hyphae and bulbous structures were observed in natural and trapped cultures.

Key words: AM Fungi, *Sesamum indicum* L., *Kuklospora, Ambispora, Glomus clavisporum*

INTRODUCTION

Arbuscular mycorrhizal Fungi are widely distributed in terrestrial ecosystems and can be found in both natural and agricultural areas. Arbuscular Mycorrhizal Fungi (AMF) belonging to the phylum Glomeromycota are important soil organisms that form mutualistic associations with plants, and which are involved in the uptake and transport of mineral nutrients to plant roots (Barea *et al*. 2002). Up to 90 % of analysed plant species are able to form this symbiosis (Smith & Read

1997). AM Fungal ubiquitous presence and their taxonomic, genetic and functional diversity are directly related to plant and soil processes and therefore there is an increasing interest in the assessment of the biodiversity and functions of AM Fungal communities. Although biodiversity has been a major research topic in terrestrial ecology, it has been largely ignored in terms of soil biota mainly in the tropical regions (Patrícia Lopes Leal *et al*.,2009).

The documentation of new patterns of species distribution is necessary for the accurate estimation of the diversity and distribution of this important group of symbiotic fungi. Diversity of AM Fungal species is measured mainly by extracting, counting and identifying their field collected asexual spores, the fungal propagule that possess morphological characters to define species in this group of organisms although molecular techniques have been revealed as useful tool for characterization and identification of AM Fungi.

Field-collected spores, however, are found in some circumstances in low numbers, parasitized, lacking informative taxonomic characteristics impairing a more accurate identification as components of spore walls are susceptible to alteration and deterioration by a wide sort of agents in the soil. Establishment of trap cultures using bulk soil or by mixing rhizosphere soil and root pieces with sterilized diluents and growing with suitable hosts, represents a strategy to yield a large number of healthy spores which can be readily identifiable and supplement the assessment of local species diversity in different ecosystems. This methodology not necessarily allows the identification of all species, because sporulation of the fungal community may be affected by the plant host chosen for trapping whereas in some cases it can promote the sporulation of cryptic AM Fungal species that were not sporulating at the sampling time or natural field conditions. Despite this, trap cultures have been widely used to access AMFungal diversity and isolate indigenous AMFungi (Patrícia Lopes Leal *et al*., 2009).The purpose of this work was to record the number and type of AM Fungal species that colonized *Sesamum* plant in Sanjay Gandhi National Park.

MATERIALS AND METHODS

Soil sampling : Root samples and rhizosphere soil of *Sesamum* was collected from Sanjay Gandhi National Park, Borivali, Mumbai and preserved in sterile polythene bags and stored in refrigerator at 4^0 C until use. Soil sample up to 20 cm depth was collected . Root samples were cut into 1cm bits and preserved in FAA until use.

Trap Culture was done by Rodrigues & Muthukumar method, 2009. It is frequently observed that isolation of spores directly from field soil has drawbacks viz. they appear intact but may not be viable as they may persist in the soil as spore cases for years, they change appearance in their structural characters in response to root pigments, soil chemistry, temperature, moisture and microbial activity. Therefore trap cultures have to be prepared to recover intact , fresh and healthy spores.

Spore extraction was done by Gerdeman and Nicolson method ,1963 and t he spore number was counted by Gaur and Adholeya method ,1994, Taxonomic identification of spores up to species level was made using the identification manual of Schenck and Perez and description provided by the International collection of VAM (INVAM).

Root Colonization of AM Fungi was done by Philips and Hayman method, 1970 and percentage of root colonization was calculated by Read *et al*.,1976.

RESULTS AND DISCUSSION

Ten species of AM Fungi were identified from natural field soil (fig 1) and seven species sporulated in 3 months period in trap cultures (fig 2). The species identified are listed in the table. Spore density was also less in trap culture compared to natural conditions. Percent root colonization was less in trap cultured roots of *Coleus* compared to the *Sesamum* from field conditions. Cuenca et al. (2003) reported that AM Fungal propagation in trap cultures exhibits difficulties because exact natural conditions cannot be reproduced; this causes a bias towards proliferation of species that are able to tolerate greenhouse conditions, and that associate better with the plant host under the specific trap culture conditions.

Table 1: AM Fungal status of *Sesamum indicum* L.

Sr. No.	Habitat	AMF Structure in Roots				% of Root Colonization	Spore Density	No. of Spore Sps.	Type of Spore Species
		H	A	V	Endophytes				
1	Natural Rhizosperic Soil	+++	--	++	+	100%	135±14.5	10	*Glomus geoporum, G.callosum,G. clarum, G. tortuosum, Acaulospora sp, Ambispora appendicula, Scutellospora sp. Gigaspora margarita, Glomus clavisporum, Kuklospora sp.*
2	Trap Culture	+++	+++	++	+	90%	90±5.57	7	*Glomus halonatum, G. constrictum, G. aureum, Gigaspora decipiens,, G.ambisporum Pacispora boliviana, Acaulospora sp.*

+Poor, ++Moderate, +++Good, ++++ Excellent, - Absent

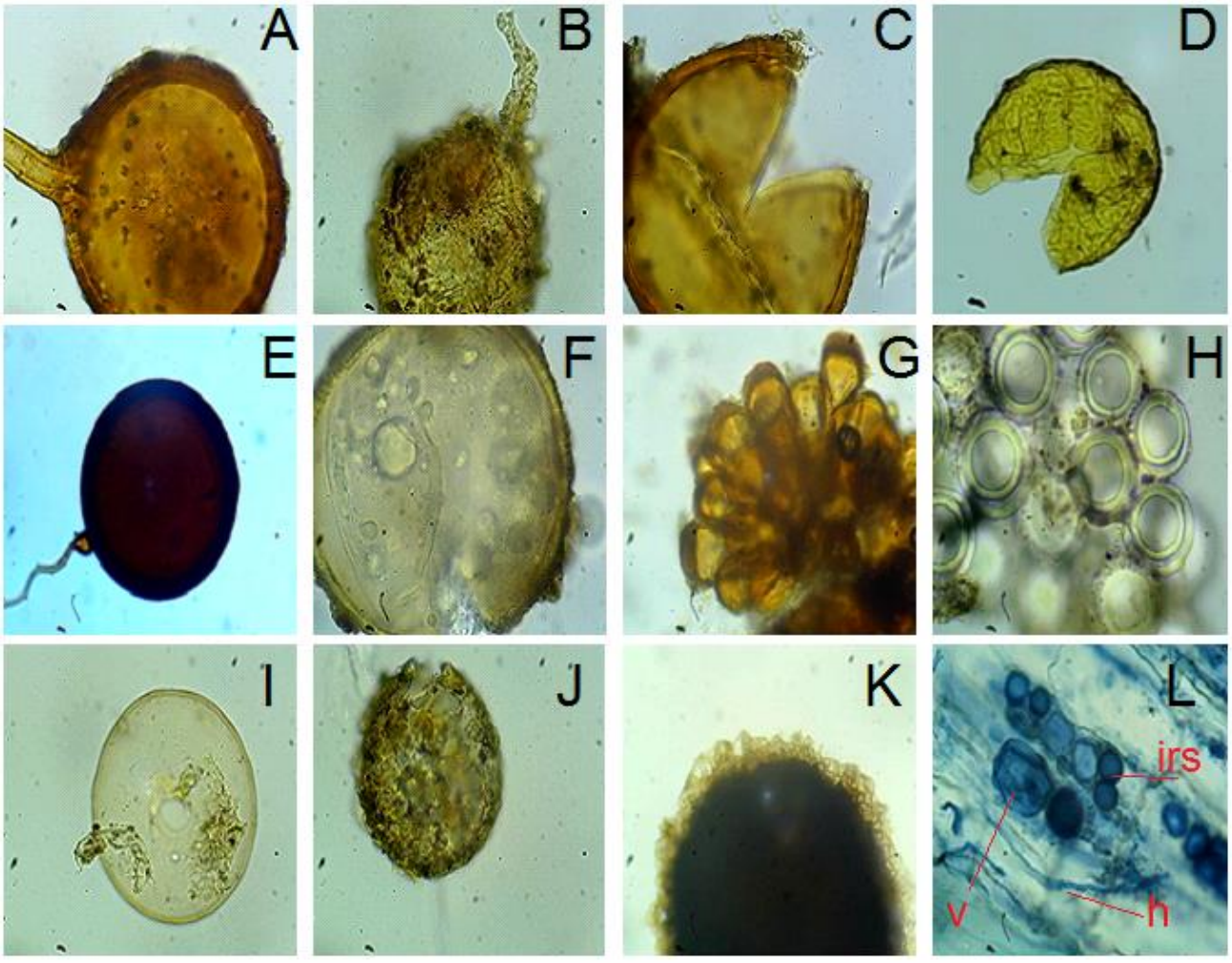

Fig. 1. AM fungal status of *Sesamum indicum* L. in Natural habitat. **A.** *Glomus geosporum* **B.** *Glomus callosum* **C.** *Glomus clarum* **D.** *Ambispora appendicula* **E.** *Gigaspora margarita* **F.** *Acaulospora sp* **G.** *Glomus clavisporum* **H.** *Kuklospora sp* I. *Scutellospora sp* **J.** *Acaulospora sp* **K.** *Glomus* tortuosum **L.** AMF-colonized roots showing different internal structures indicated by arrows: hyphae **(h)**, and vesicles **(v)** ,intraradical spores **(irs).**

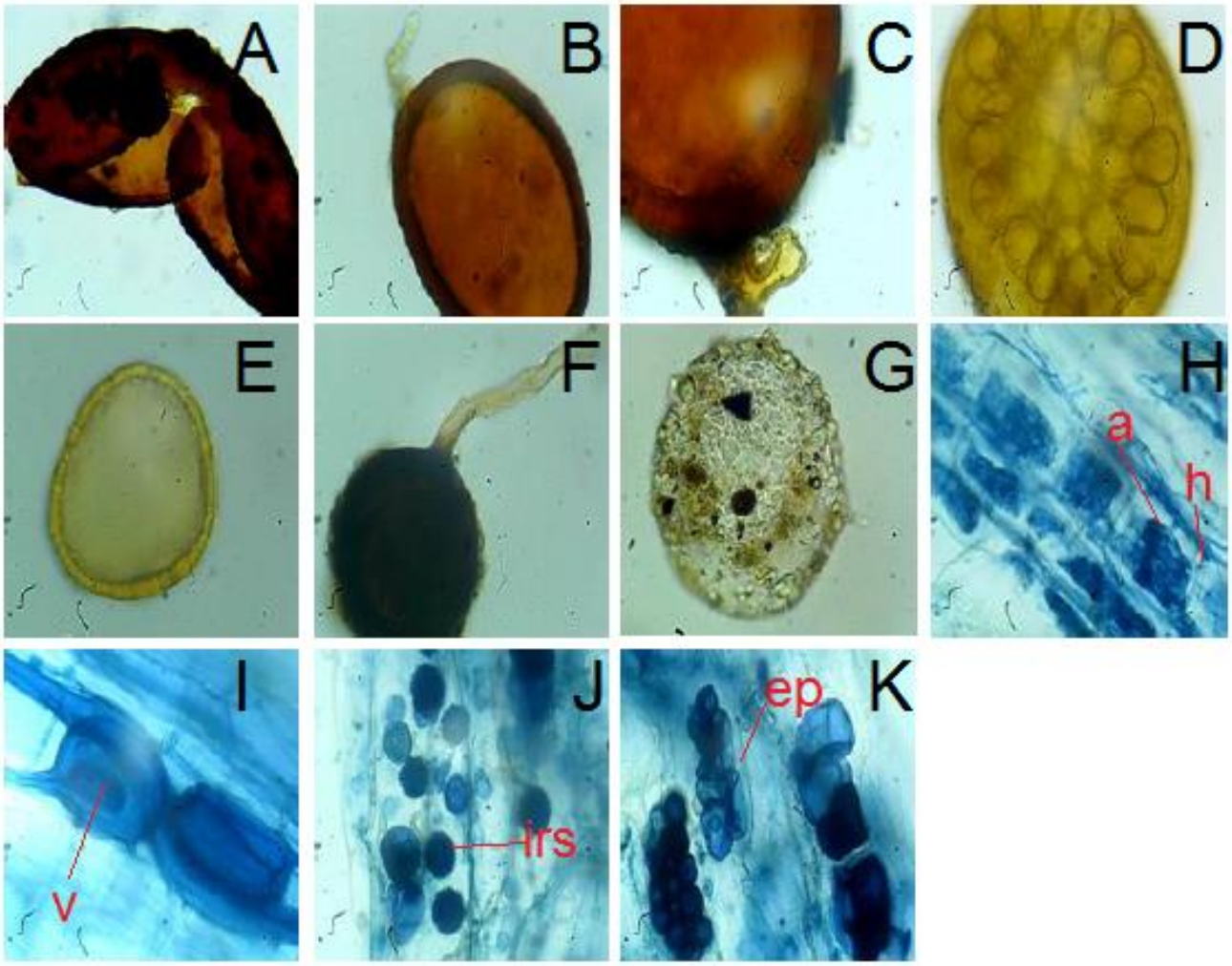

Fig. 2. AM fungal status of *Sesamum indicum* L. in Trap Culture. **A.** *Gigaspora decipiens* **B.** *Glomus halonatum* **C.** *Glomus constrictum* **D.** Sporocarp of Glomus aureum **E.** *Pacispora boliviana* **F.** *Glomus ambisporum* **G.** *Acaulospora sp* AMF-colonized roots showing different internal structures **H.** hyphae **(h)**, arbuscule **(a)** **I**. vesicles **(v)** J. intraradical spores **(irs)** **K.** endophytes **(ep).**

In the present study, the mycorrhizal structures observed were arbuscules, vesicles and hyphae in trap cultures. The arbuscule percentage was very poor whereas vesicle percentage was excellent. The vesicles were elliptical type with oil droplets very clearly seen. The arbuscules were of coiling (Paris) type. Some endophytes were also observed in roots. Very few studies describe the selective pressure on AM Fungal species when they are taken from their natural environments to trap cultures (Antunes,2012, Oliveira 2010). *Acaulospora and Glomus* genera were common in both Natural habitat and Trap culture. In trap culture *Glomus halonatum, G. constrictum, G. aureum, Gigaspora decipiens, Pacispora boliviana* were found to be sporulating which were not observed in natural habitat. According to Adholeya 1994, the non-sporulating species can often be coaxed to sporulate in 'trap cultures'. The genus *Glomus* is the most dominant AMFungal genera isolated and identified in *Sesamum* followed by *Acaulospora.*

CONCLUSION

Sesamum indicum L. is a medicinal herb. The soil trap culture using *Coleus* as the host plant showed presence of mycorrhizal structures - coiling (Paris) type of arbuscules, hyphae and elliptical intra and

intercellular vesicles along with Intraradical spores. Arbuscules were not observed in the roots from field.

The spores isolated from trap cultures belonged to *Glomus halonatum, G. constrictum , G.aureum, Gigaspora decipiens , Ambisporum, Pacispora boliviana and Acaulospora sp*.

The spores isolated from the natural rhizospheric soil belonged to *Glomus geosporum, Glomus callosum, Glomus clarum, Glomus tortuosum , Acaulospora myriocarpa , Ambispora appendicula, Scutellospora sp. Gigaspora margarita, Glomus clavisporum, Kuklospora sp*

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