



## STUDIES ON ARBUSCULAR MYCORRHIZAL FUNGI FROM SOIL OF SITAKHANDI FOREST OF MAHARASHTRA

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### ABSTRACT

Forest ecosystems are the regions where there is least human interference and therefore we see luxury of plant growth. Sithkhandi forest is one of the least human interfered forests present near Bhokar region of Nanded district in Maharashtra. *Arbuscular mycorrhizal* fungi (AM fungi) develops symbiotic relation with most of the land plants. This association is very common in the terrestrial ecosystem throughout the world. The objective of the current study is to identify and determine frequency of various AM fungal species. Five GPS marked sampling sites were selected for isolation of mycorrhizal spores. A total 34 species belonging to genera *Glomus*, *Acaulospora*, *Scutellospora*, *Gigaspora*, *Diversispora*, *Enterophora* and *Pascispora* were reported. Little variation is seen in their frequency. In this study, more species diversity among *Glomus* followed by *Acaulospora* were found. High frequency percentage of *Glomus mosseae* has been reported at all study sites.

**Keywords:** Mycorrhiza, *Glomus*, *Acaulospora*, Sitakhandi forest.

### 1. INTRODUCTION

Symbiotic *Arbuscular Mycorrhizal* Fungi (AM fungi) forms an extensive hyphal network for providing water and nutrients to living plants [1]. These symbionts are most commonly found in large majority of terrestrial plants [2]. The AM fungi belong to phylum Glomeromycota which forms main component of soil mycoflora. From the past three decades, this group of soil mycoflora have drawn the attention of researchers because of their ability to form intimate association with 70 to 90% of plant species [3]. In addition to nutrient uptake, this association is involved in protection against soil borne pathogens and improvement of soil fertility and stability. Their detection and studying diversity in soil is very essential for any agro-ecosystem [4]. Approximately, 150 AM fungi have been described by means of morphological characteristics of spores [5]. Because of their wide presence in soil, it is believed to contribute significantly to global phosphate and carbon cycle and influences primary productivity in terrestrial ecosystem [6]. Establishment of such mutualistic relationship can stimulate, activation of antioxidant, phenylpropanoid, and carotenoid pathways [7]. Synthesis of plant secondary metabolite which are important for increased plant

tolerance to abiotic and biotic stress are beneficial to human health through their antioxidant activity [8]. These obligate symbionts are not host specific and one species may found to be associated with various plants in the same locality [9] and also one host plant can support mixed population of AM fungal species [10].

Sitakhandi forest is about 39 km away from Nanded and Bhokar respectively. It is divided in to minor south and major north region by a middle road. The forest cover is approximately 7.0 km<sup>2</sup>. It is dominated by plant like Teak (*Tectona grandis* L) and also consists of various types of herbs and grasses. The vegetation of forest comes under the category of dry deciduous type. Since this forest has thick plant cover therefore we observe least interference from human. Therefore, we have undertaken this study.

### 2. MATERIAL AND METHODS

#### 2.1. Collection of Soil Samples

For the present work, rhizosporic soil samples were collected from Sitakhandi forest. Samples were collected in the month of June-21 from five different sites. The geographic locations of sites are shown in the Fig 1. Sampling sites are named as S-I (19° 14' 18° N and

77°35' 29°E); S-II (19° 14' 39° N and 77°35' 33°E); S-III (19° 14' 39° N and 77°35' 56°E); S-IV (19° 14' 03° N and 77°36' 03°E); S-V (19° 14' 09° N and 77°35' 43°E). The rhizosporic soil samples were collected from above mentioned locations in clean polythene bags and brought to laboratory for the isolation of mycorrhizal spores.



**Fig. 1: Geographical locations of sampling sites from Sitakhandi forest**

## 2.2. Isolation and identification of AM fungi

Mycorrhizal spores from soil samples were isolated by wet sieving and decantation technique as described by Gerdemann and Nicolson [11]. About 100 gm of individual air dried rhizosporic soil sample was suspended in a beaker containing 500 ml water and the suspension was left behind for about 15-20 minutes. The suspension was then decanted through series of sieve stacks of 500µm to 25µm arranged in decreasing order from top to bottom. Similar procedure was repeated for 2 to 3 times. The residue from each sieve was collected in separate dishes with little distilled water. The intact fungal spore was examined and counted using high power stereo zoom microscope. The spores were mounted on glass slide using polyvinyl alcohol, Lactic acid and Glycerol (PVLG). Identification of spore was done by observing diagnostic features like spore wall, colour, size and hyphal attachment [12]. The additional spore identification were done as per the description given in INVAM worksheet (<http://invam.caf.wvu.edu>) [13].

## 2.3. Frequency Determination of AM fungal Species

Frequency is used to assess the establishment and survivability of AM fungi in the rhizosporic soil. It shows the number of sampling in which spore of particular AM fungus present during the study period expressed as percentage [14]. In the present study, dominant AM

fungal species according to their frequency (F) were determined using following formula given by Yang, et. al. [15, 16] and Elizabeth Temitope, et.al. [17].  

$$\text{Percentage frequency (\%F)} = \frac{\text{Number of times of certain AM species occurrence}}{\text{Total number of AM fungal species occurred in the study}} \times 100.$$

## 2.4. AMF Spore Quantification

Enumeration of AM fungal spore was done by modified grid line intersect method [18]. In this method, filter paper was divided in to many small compartments using ball point pen of approximately 1cm<sup>2</sup> and numbered separately. The total number of spores are counted in each segments by using high power Labomed make stereo zoom microscope [19].

## 3. RESULTS AND DISCUSSION

The identification of indigenous AM fungi is a fundamental requirement to understand biodiversity and essential for monitoring changes in natural, managed or disturbed ecosystems [9]. The significance of AM fungi in plant ecology is based on its widespread occurrence in natural ecosystem [20]. AM fungi have considerable impact on plant growth and therefore management of these fungi in terms of their isolation, mass cultivation for production of effective biofertilizer is essential for sustainable agriculture. In this regard, it is very much essential to study the AM fungal diversity with respect to dominance and their frequency of occurrence. This will be helpful to identify effective AM fungal species which occurs in diversified habitat. With this background, the attempt was made to study AM fungal diversity and to isolate frequently widespread occurring species from Sitakhandi forest area of Nanded. The results of study shows that AM fungal spores having 35 different fungal species belonging to seven different genera were reported. The diversified AM fungal genera reported are *Acaulospora*, *Diversispora*, *Entrophospora*, *Gigaspora*, *Glomus*, *Pascispora* and *Scutellospora*. Among all genera the maximum special diversity were recorded in *Glomus* followed by *Acaulospora*, *Diversispora*, *Scutellospora* and *Gigaspora*.

Species diversity and percentage frequency of *Glomus* species are shown in Fig.2. Fifteen different species of *Glomus* have been reported from all sample sites. The species of *Glomus* such as *Glomus aggregatum*, *G. ambisporum*, *G. clarum*, *G. constrictum*, *G. deserticola*, *G. fasciculatum*, *G. fecundisporum*, *G. flavisporum*, *G. halonatum*, *G. intradices*, *G. lacteum*, *G. macrocarpum*, *G. mosseae*, *G. pansihalos*, *G. versiformae* were recorded. The

maximum frequency of occurrence of *G. mosseae* followed by *G. intradices*, *G. deserticola*, *G. constrictum*, *G. clarum*, *G. halonatum*, and *G. flavisporum* were observed from all the sites of forests. Khade and Rodrigues [21] found that forest species shows maximum colonization of *Glomus* species. These species are known to be widely distributed and are commonly found in different geographical locations. Similarly, Hindumati and Reddy [22] also reported abundance of *Glomus* in the field of Nizamabad and Karimnagar districts of Andhra Pradesh State.

Moreover, these species are easily adaptable to adjustment of sporulation patterns in variety of environmental conditions which may results in its dominance in occurrence [23]. In the present study, high frequency occurrence of *G. mosseae* in all the sampling sites shows its diversified nature of adaptability to different locations. Therefore, it is necessary to make detailed study on its isolation, further characterization and mass cultivation for benefits of sustainable agriculture.

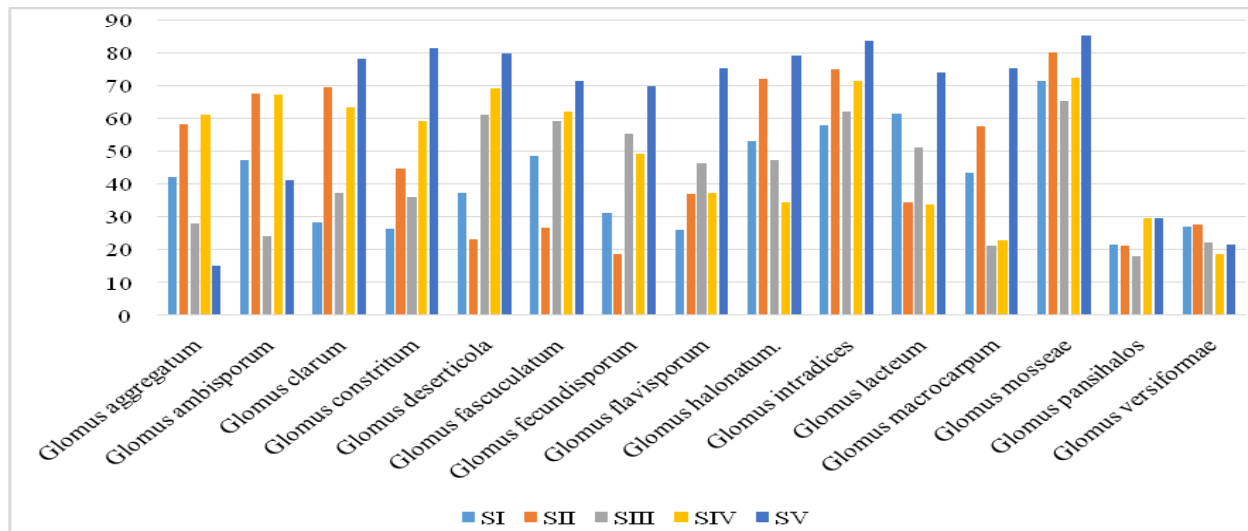


Fig. 2: Percentage frequency occurrence of *Glomus* species at different locations of Sitakhandi forest.

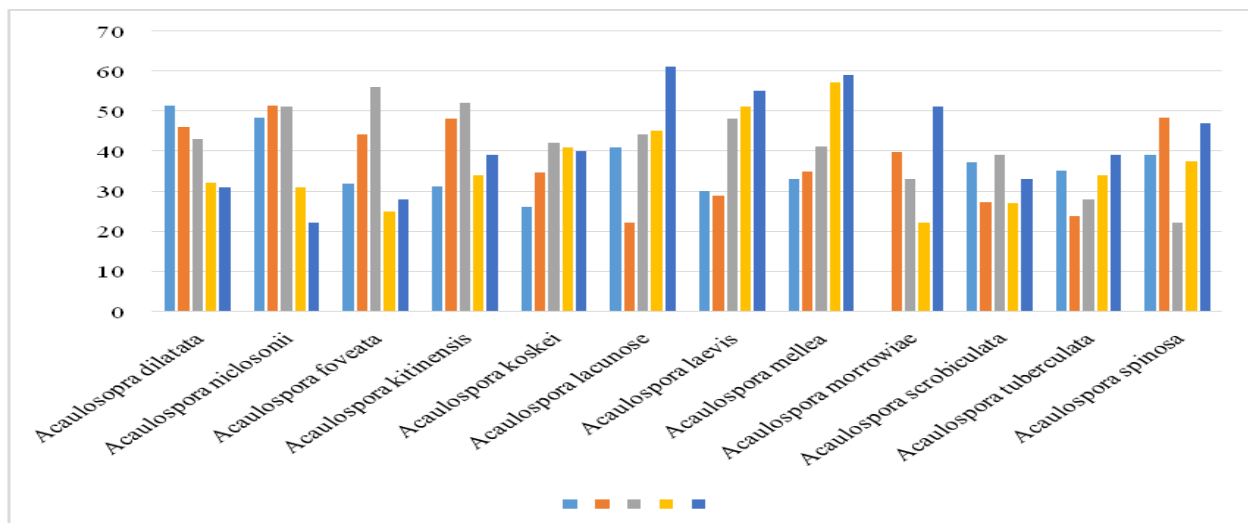


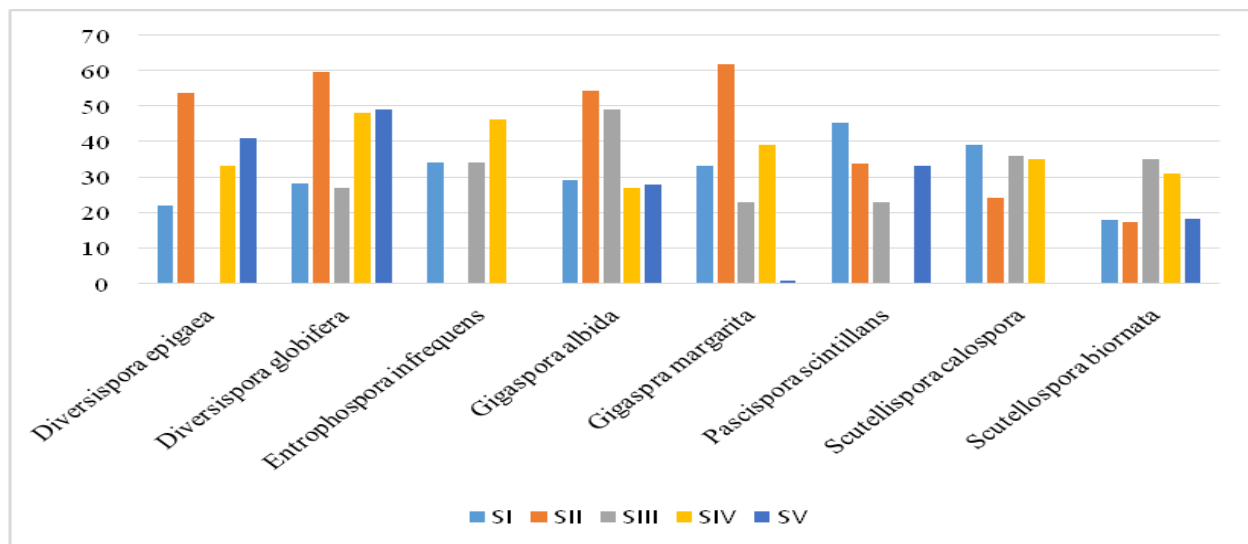
Fig. 3: Percentage frequency occurrence of *Acaulospora* species at different locations of Sitakhandi forest

Species diversity in *Acaulospora* is shown in Fig. 3. In the present study followed by *Glomus*, *Acaulospora* showed twelve species at all five studied sites except *Acaulospora*

*morrowiae* at S-I. The different species of *Acaulospora* reported in the current study are *Acaulospora dilatata*, *A. nicolsonii*, *A. foveata*, *A. kitinensis*, *A. koskei*, *A. lacunose*, *A.*

*laevis*, *A. mellea*, *A. morrowiae*, *A. scrobiculata* and *A. tuberculata*. From all the studied sites, the frequency of *A. lacunose* was found higher followed by *A. mellea*, *A. laevis*, *A. dilatata* and *A. niclosonii*. *Acaulospora* and *Glomus* species sporulate profusely in shorter span of time due to their small size of their spores than *Gigaspora* and *Scutellospora* and therefore these two genera shows high adaptive values in different habitats [24]. Sporulation pattern among *Glomus* and *Acaulospora* is little different.

*Glomus* species sporulate first, while *Acaulospora* species do so later in seasonal period [25]. Earlier studies on *Glomus* and *Acaulospora* showed that their pattern of sporulation and colonization of many forest and cultivated plants enables them suitable for high adaptability in different habitats. In the present study all sites i.e. S-I, S-II, S-III, S-IV and S-V sites showed high abundance of both these genera.



**Fig. 4: Percentage frequency occurrence of *Diversispora*, *Gigaspora*, *Scutellispora*, *Entrophospora* and *Pascispora* species at different locations of Sitakhandi forest.**

The frequency percentage of *Diversispora*, *Entrophospora*, *Gigaspora*, *Pascispora* and *Scutellospora* is shown in Fig 4. From the figure, it is clear that *Diversispora globifera* and *D. epigaea* are present at all sampled sites of forest except S-III where later was not reported. *Gigaspora albida* and *G. margarita* are reported from all studied sites. Only 0.78% frequency of *G. margarita* was reported from S-V. Similarly, *Scutellospora biornata* was reported from all sites whereas *S. calospora* was absent from S-V. *Entrophospora infrequens* was found at all sites except S-II. Similarly, *Pascispora scintillans* was not found at S-IV. In this study, the frequency of occurrence of *Gigaspora* and *Scutellospora* were found little less as compared to *Glomus* and *Acaulospora*, this might be because the spores of these genera took longer time for formation and maturation [19]. The members of Gigasporiaceae typically establishes an extensive mycelium in the soil but produces less spores as compared to other members of Glomaceae and Acaulosporaceae [26, 27]. This might be one of the reasons in getting less frequency of these genera in the soil.

In this study AM fungi belonging to genus *Glomus* was found in more number with higher frequency. Therefore, it is one of the dominant genus reported in the forest soil of Sitakhandi region. Earlier studies made by Sharma et al [28], Byra Reddy et al.[29], Uniyal and Uniyal [30], Tamuli and Boruah [31], Mohan Kumar and Sivaswamy [32] in this regard supports the similar findings. The dominance of *Glomus* species in particular *G. mosseae*, has wider adaptability to different habitats and therefore, we have reported high frequency percentage in our study. Through scientific sets of experiments, this species of *Glomus* can be further explored for biofertilizer production.

#### 4. CONCLUSION

The current study carried out on study of mycorrhizal fungi at Sitakhandi forest showed domination of *Glomus* followed by *Acaulospora* species. Maximum occurrence of both these species supports its wide adaptability to diversified habitats.

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