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Biodiversity of the arbuscular mycorrhizal fungi isolated from *Cynodon dactylon*

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ABSTRACT

Arbuscular mycorrhizal fungi are essential helpers in the soil environment. The symbiotic association helps them in various ways to improve stem, growth, tolerance to certain insects or prey, and sustainability under unfavorable conditions such as salinity, temperature, anaerobic conditions, and low nutrient requirements. In a study, the diversity of arbuscular mycorrhizal fungi of *Cynodon dactylon* was isolated and identified. *Cynodon dactylon* was preferred because it is readily available. The soil to be analyzed for the arbuscular mycorrhizal spores was obtained from the roots of *Cynodon dactylon*. Mycorrhizal colonization and the number of mycorrhizal spores were positively correlated with host plant growth. *Glomus* species were abundant, unlike the other species found in the soil. *Glomus fasciculatum* and *Glomus macrocarpum* were identified by comparison with previous research. The study also helps in developing biofertilizers for plant growth by providing them with proper mycorrhizal nutrients in the form of colonized roots.

Key words: *Arbuscular mycorrhiza, Root colonization, Percent Mycorrhization*

Introduction

Arbuscular mycorrhizal fungi form a symbiotic relation with the plants which benefits it in exchange of nutrients, improves pathogen or drought resistance. The symbiotic relationship proves that AMF has broad application in the agriculture field. Spores are highly beneficial to the increase growth of the plants (Ropars *et al.*, 2016). Myocorrhizae not only increased plants survival, but also promoted faster development and, ultimately, higher yield without compromising soil sustainability for production (Phirke *et al*., 2002). AMF are obligate symbionts which belongs to phylum Glomeromycota which forms 80% mutualism with plants, host plants nutrients requirement are fulfilled by providing nutrients and water in exchange of photosynthetic products. Carbohydrates and minerals are exchanged through the external surface of the roots.Arbucular mycorrhizae forms differentiable structure such as vesicles,arbuscles by glomeromycota. AM fungi supports plants to absorb various nutrients mainly phosphorus, sulfur, nitrogen and some nutrients from soil. The arbuscular mycorrhhizal symbiosis plays pivotal role to plant colonisation and vascular plant growth. (Brundrett *et al*., 2002). Arbuscles are formed in the thin layer of parenchyma tissue. AM spores have tendancy to germinate whenever the faverable condition are their such as temperature, carbon dioxide concentration, phosphorous concentration and pH (Wright *et al.*, 2005). Hyphal growth and branching depends upon the phosphorous concentratio in the soil. Concentration of the phosphorous and the hyphal growth are inversely proportional to each other. Each roots have the special sites for exchanging the nutrients. Arbuscles are the main site of the roots from where the nutrients are exchanged. There are mainly two forms, paris and

the arum, paris type and arum type are distinuished on the basis of hyphal growth. Paris type is characterised by the emerging point of the hyphae which rises from one cell to the next cell. In arum type, the hyphae are grown in between the space of the plant cells (Lara *et al.,* 2002). The plant family will decide under which type they will fall either paris or arum (Yamato and Masahide, 2005). There are primary two types of mycorrhiza; ectomycorrhiza and endomycorrhiza depending upon the site of the fungal hyphae inrelation with the root tissue of the plant. Ecto indicates outside the root and endo indicates inside the root. Endomycorrhiza are then subdivided into 3 major groups; Arbuscular endomycorrhiza, ericoid endomycorrhiza and orchidaceous endomycorrhiza.

Arbuscular endomycorrhia: These are the most common mycorrhizas which are firstly evolved belonging to glomeromycota. They have 80% association with the roots (Selosse and Le Tacom, 1998).

Ericoid endomycorhiza: These mycorrhiza are present in the challenging environment.It mainly provides nitrogen to the plant. It rarely provides the carbon source.

Orchidaceous endomycorrhiza: There main role is to provide the carbon source to the growing plants. It is somewhat similar to ericoid endomycorrhiza.

Ectomycorrhiza: It indicates the assocciation outside the roots. It is advanced symbiotic association between fungi and the higher plant. They have 3% of association with the roots. Mostly basidiomycota are involved in the ectomycorrhiza.

Materials and Methods

Stacking sieves with nylon or stainless steel mesh and large range of pore size for isolating spore, stereo microscope, trypan blue stain, root trainer, peat soil, micropipettes.

Collection of the Soil Sample

The soil samples were collected from the area where *Cynodon dactylon* are in more amount. The soil was taken from the roots of the experimental plant.

Collection of the Root Sample

The roots were collected from the mycorrhizal soil which was exposed to the mycorrhizal endospore.

Isolation of the Endospore

The extraction of spore was carried out by wet sieving and decanting tehnique by Gerdermann and Nicolson (1963). The spores were identified by comparing it with finding of other research paper.

Staining of the Roots

The roots were taken from the plant which was exposed to the mycorrhizal endospore. Mycorrhizal root colonization was measured in fresh roots cleared in 10% KOH for 10 min at 90 °C, after that it is placed in the solution of H_2O_2 for 20 min and washed afterwards. Then roots are treated with the HCl for 5 min and stained in 0.05% lactic acid-glycerol Trypan Blue (Phillips and Hayman, 1970).

Determination of Root Colonization

Per cent of AMF colonization was estimated by microscopically examination at 10 X Magnification, after clearing of roots in 10% KOH and staining with 0.05% trypan blue in Lactophenol according to method described in Phillips and Hyman (1970).

The mycorrhizal colonization was determined by using following formula:

Per cent of mycorrhizal colonization =Number of root segments colonized/Total number of root segments examined × 100.

Results and Discussion

From the above methods, the main purpose of this study was to isolate the endospore from the soil near the plant *Cynodon dactylon.* The isolated endospore was used as a source of nutrition to the plants. To check the effect of the spores on the plants activity was the aim of the study. The mycorrhizal association helps in the insoluble phosphorus intake. The insoluble phosphorus is taken as a nutrient to the plant growth with the help of mycorrhizae.

These are the spores which have been isolated from the soil which was taken from the roots of *Cynodon dactylon* (Photo 1). Different types of spores were isolated from the spores. The glomus species are present in abundance.

The isolated spores were compared to the other research paper and the identification was done on the basis of the appearance, cell wall layer and colour. Following table shows different spores of the glomus and *Scutellospora* sp.

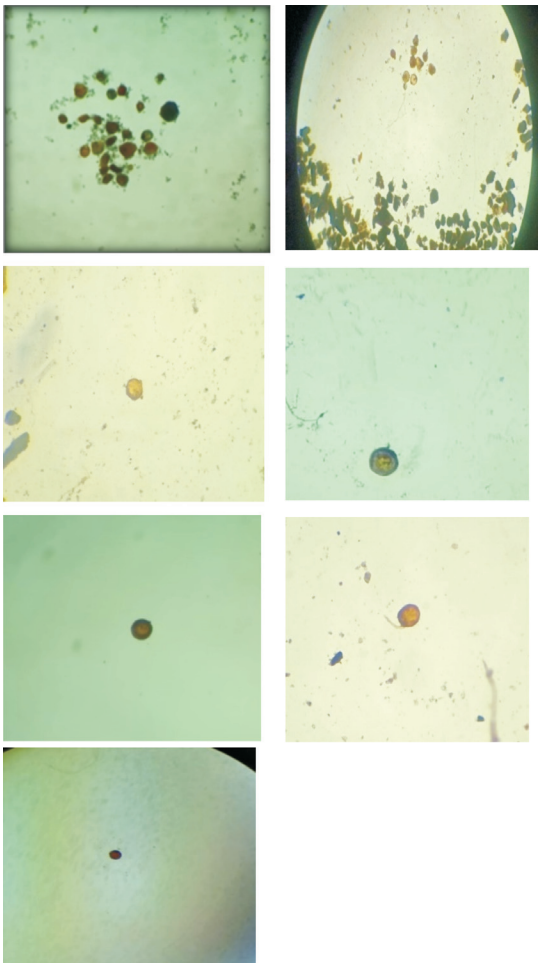

Photo 1. Microscopic observation of spores from *Cynodon dactylon*

Root Colonization

Total roots-30
Infected roots-13

Root colonization = Number of root segments colonized / Total number of root segments examined X 100

= 13/30 x 100
= 43.33%

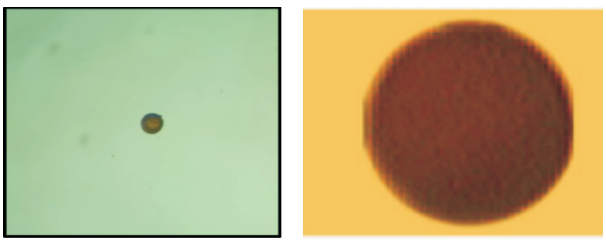

Photo 2. *Scutellospora* sp (Suchitra *et al.*, 2021)

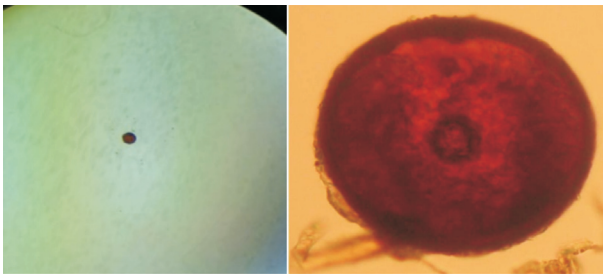

Photo 3. *Glomus maculosum* (Miller and Walker,1985)

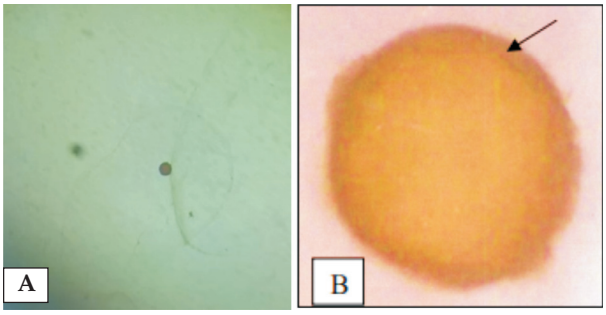

Photo 4. *Glomus macrocarpum* (Martina and Vosatka, 2005)

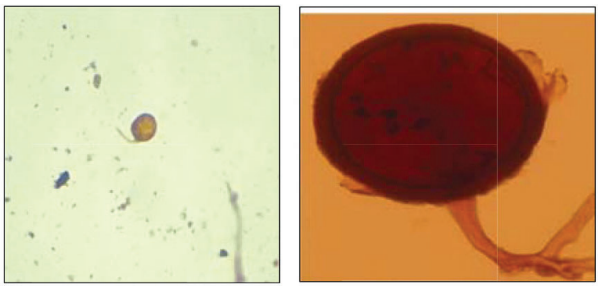

Photo 5. *Glomus dimorphicum* (Boyetchko and Tewari, 1987)

Root colonization study was found to be 43.33%.

Staining of the mycorrhizal root was done to identify the infected roots.

These photos show vesicles along with hyphae, Cortex along with vesicles

Arbuscules (A) along with vesicles and Root cortex (Photo 7).

H and V are hyphae and vesicles of the roots.

Description of Photo 10 to 13.
The comparative study of the plants which differs in sterile mycorrhizal, non-sterile mycorrhizal soil with respect to the control soil

The comparative study was done by taking sterile mycorrhizal soil, non-sterile mycorrhizal soil in comparison with the control soil to identify the dif-

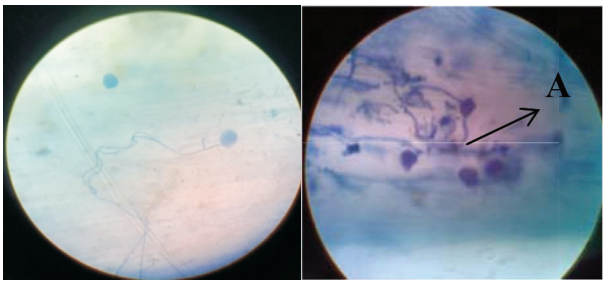

Photo 6 **Photo 7**

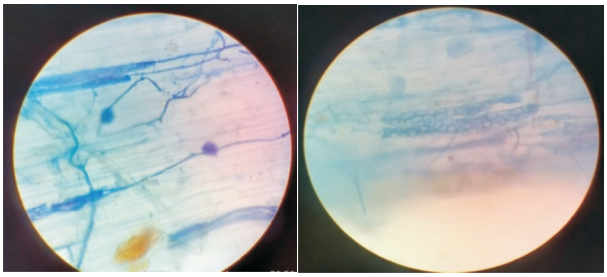

Photo 8 **Photo 9**

ferent growth parameters of the plants in response to the addition of the mycorrhizal endospore.

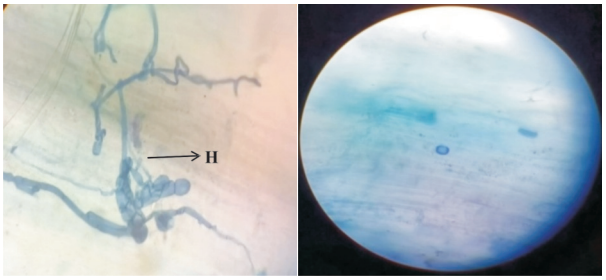

Photo 10 **Photo 11**

Conclusion

The preceding investigation comprises the isolation and application of the endospore on various plants

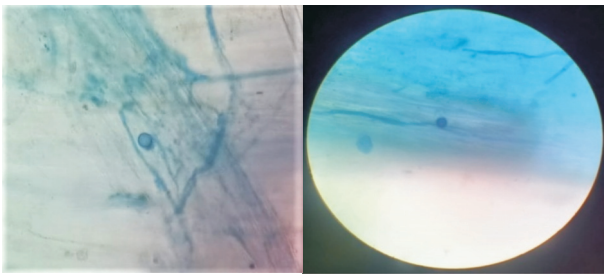

Photo 12 **Photo 13**

Photo 14. Day 3 **Photo 15.** Day 4 **Photo 16.** Day 5

to define the various parameters.

The major goal of this study was to separate spores from soil; the isolated spores were used for various purposes and belonged to the genus *Scutellos pora*, aculospora, and glomus species, which were recognized by comparing it to conventional spore characteristics. The spore's activity included monitoring the plants, which included moong, maize and mataki. The spores had a beneficial effect on the plants. Plant growth was enhanced as compared to plants that did not get mycorrhizal inoculation. Arbuscular mycorrhizal fungi infested the plant roots, which is good to plant growth.

The fundamental job of the Arbuscular mycorrhiza was to promote strong host plant development under stressful conditions by building a series of intricate communication channels between the plant and the fungi, resulting in increased photosynthetic

Table 1. Effect of mycorrhizal treatments on different growth parameters in pot trials

	Seed Wt. (g)	% of seed germination	Shoot			Root			Number of leaves	Total Mass
			Height (cm)	Fresh Wt.(g)	Dry Wt. (gm)	Max. Length (cm)	Fresh Wt.(g)	Dry Wt. (g)		
Control	1	100	20.1	1.11	0.54	3.16	1.1	0.4	12	1.52
Moong with Sterile mycorrhizal soil	1	100	22	1.18	0.62	4.15	1.5	0.6	14	1.6
Moong with Non-Sterile mycorrhizal soil	1	100	17	1.02	0.42	3.01	1.2	0.3	10	1.42

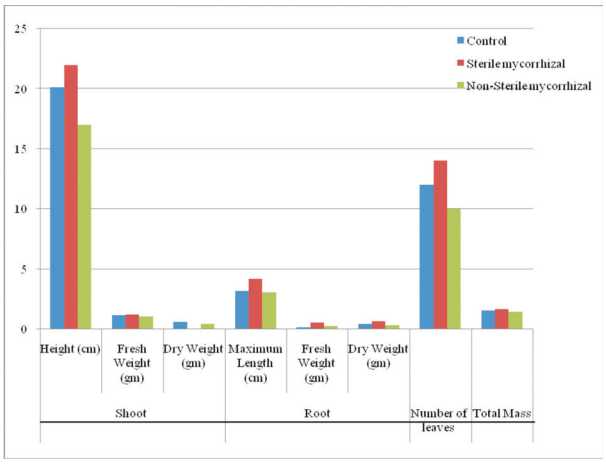

Fig. 1. Graph showing the comparative study of the plants which differs in sterile mycorrhizal non-sterile mycorrhizal soil with respect to the control soil

rate and other gaseous exchange. AMF boost plant nutrition by enhancing nutrient availability and translocation. The AMF-infected roots can be utilized to make biofertilizer. The biofertilizer produced in this manner is advantageous in many ways, including availability, cost, and an effect that is not damaging to both plants and the environment. It is both human and ecologically beneficial. It is also affordable.

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