Research Article

Diversity of Vesicular Arbuscular mycorrhizal fungi in *Parthenium hysterophorus* from different sites of Jabalpur M.P. India

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Abstract

Background: Arbuscular mycorrhizal fungi are found in a wide range of habitats usually in the roots of angiosperms, gymnosperms and pterydophytes, whose diversity can be estimated by studying rhizopheric soil and also by colonization patterns in roots of the host plants. **Objective:** To find diversity of Vesicular Arbuscular mycorrhizal (VAM) fungi associated with *Parthenium hystophorus*. **Materials and methods**: Thus, an investigation was carried out on a weed plant '*Parthenium hysterophorus*' to determine the mycorrhizal association potential of different fungi in Jabalpur district of Madhya Pradesh, India. Ten species of arbuscular mycorhiza fungi of the genus *Glomus aggregatum, Glomus geosporum, Glomus fasciculatum, Glomusmosseae, Sclerocystiscorymeoides, Scetullospora jabalpurensis, Acaulospora appendiculata, Acaulospora rehmii, Gigaspora margarita* and *Archeospora* were studied. **Results:** The highest VAM spore density was measured from RDVV Campus of approximately 119 density/50 gram soils and least was noticed from site Gauri ghat area. Species richness was maximum from RDVV Campus and from site Dumna park while site Sadar, Gorabazar, Adhartal contains species richness of 3 each. Site Gorabazar area has species richness of 4 each. The result suggested that AM fungi are well distributed in *Parthenium* plant species in central Jabalpur area.

Keywords: *Parthenium hysterophorus*, Vesicular Arbuscular mycorrhizal fungi, Jabalpur district and frequently occurring *Glomus* species

Introduction

Increased efficiency of mycorrhizal roots and non efficiency in non-mycorrhizal roots is due to active uptake of following nutrients like, Phosphorous, Zinc and Copper (Phiri et al., 2003, Jamel et al., 2002). Khubato et al. (2005) described that the morphology of A.M. type association depend upon the interaction between plant and fungal species. Frank (1885) first give this term 'Mycorrhiza' to define it as the essential structure and functioning of the peculiar connection between the roots and ectomycorrhizal fungi. They are known to enhance plant tolerance to biotic and abiotic stresses like nutrients, drought, metal toxicity, salinity and pathogens, which may affect successful establishment. *Glomus* was found to be most dominant as trace metal tolerant, as Arbuscular mycorhiza fungi isolated from contaminated part (Joner, 2003; Malcova, 2003).

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The AM fungi can increase the survival rate of plants, reduce susceptibility to environmental stresses, and increase nutrient aquision in plants, and increase carbon sequestration and nitrogen fixation in the soil (Almas et al., 2004). Spain et al., (2006) classified AM fungi into different groups based on the structure of their soil-borne spores and DNA sequences. The AM fungi, belonging to Glomeromycota phylum, was classified into four orders, namely Archaeosporales with two families (Ambisporaceae and Archaeosporaceae), Diversisporales with four families (Gigasporaceae, Entrophosporaceae, Diversisporaceae and Acaulosporaceae), Paraglomerales with only one family of Paraglomeraceae and Glomerales with only one family of Glomeraceae. *Glomus* is the largest genus of AM fungi, belonging to Glomeraceae family, currently defined as non-monophyletic. Mycorrhizal associations may be initiated by spore germination. Most of the plant species can be colonized by AM fungi under natural stressed rangeland conditions (Neeraj et al., 1991). Miller (1979) showed that when soil from rhizosphere is removed, it resulted in

decrease of mycorrhizal propagules. Local variant of AM fungi are more beneficent compared to foreign AM fungi used in that region (Requenta et al., 1997). A number of factors for successful colonization of AM fungi are pH, soil nutrients, organic matter, moisture, temperature, and the age of disturbed sites, which have shown correlation with AM root colonization and diversity (Mukhopadhyay and Mati, 2009). AM fungi colonise the plant roots to derive carbon for their survival. AM fungi gives both intra and extra radical structures. It is fact that rate of translocation of nutrients becomes high during plant-microbe interaction. Microbes present in plant root rhizosphere are treated as plant growth promoting rhizobacteria (Kumar et al., 2014-2015). Mycorrhizal diversity is mostly in forests compared to other area (Chaturvedi et al., 2009).

Materials and methods

Site and location

The location of Jabalpur is 23°10' North latitude and 79° 59' East longitude. Jabalpur is situated on Varanasi-Nagpur NH-7. Nestled in the 'Mahakaushal' region in the central part of India, it is has a peaceful ambiance and a tranquil environment, Jabalpur enjoys a prime location. It is located at the centre of Madhya Pradesh.

Collection of soils and root samples

Roots and soil samples were collected from the rhizosphere of plants growing in that area. From each site, 3-4 healthy plants of Parthenium were selected. The roots of plant and rhizosphere soil was dug out with a trowel to a depth of 0-15 cm after scrapping away the top 1 cm layer of soil. Samples were collected randomly from different zone in each site, pooled and homogenized. Before processing, all the samples were sieved (< 2 mm mesh size) to remove stones, coarse roots and other litter, and fine roots were collected from each sample. Soil samples were air dried and stored at 4°C for further experiments.

Isolation of Arbuscular mycorrhizal Species from plant roots

After sample collection, next step was isolation of *Arbuscular mycorrhizal* species collected from root samples. The following steps were made:

Fine root samples were collected and then washed with running tap water and fixed in FAA (Formalin Acetic acid). Roots were segmented into 1cm bits. Three replicates of 100 root bits each, selected at random were processed separately for determining the mycorrhizal intensity in the roots. Root bits were treated with 10% KOH solution for 30 min. at 40 ºC temperatures. The concentration of KOH and time of incubation of roots depend upon the age and softness of the roots. Pour off the KOH solution and rinse the roots well in a beaker using at least three complete changes of tap-water or until no brown colour appears in the rinse water. After thorough washing, root bits were stained with trypan blue (0.01% trypan blue) for 24 hours at room temperature. Stained root pieces were mounted in lactoglycerol and examined under microscope for the mycorrhizal colonization and its spore's structures study.

Isolation of VAM spores from rhizosphere Soil mixtures

Spores were isolated from field-collected root-rhizosphere soil mixtures. Spores of AM fungi were isolated by using the 'wet sieving and decanting method' described by (Gerdemann and Nicolson, 1963). In soil remove the coarse materials like straw, debris and rocks should be removed with a 2-mm sieve.100 g of air-dried root-rhizosphere soil

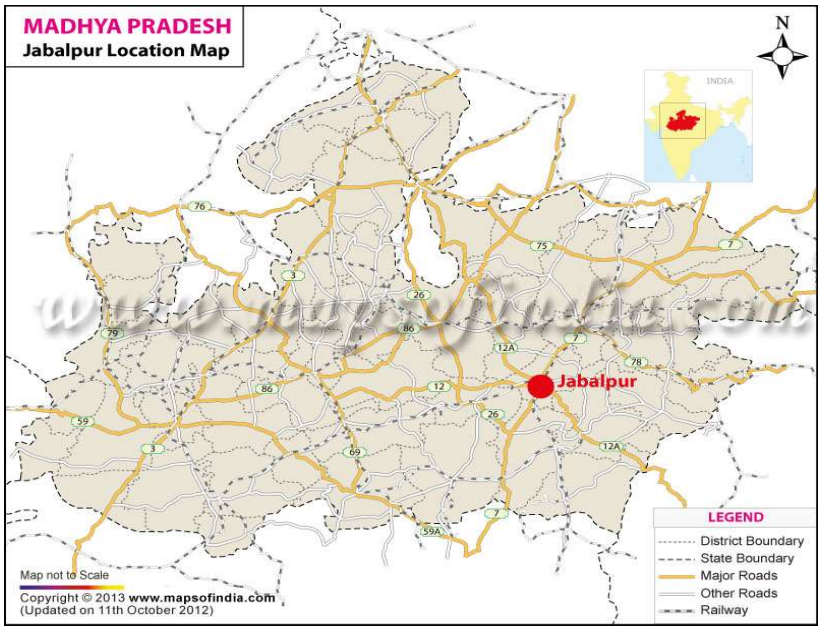

Figure 1. Location of Jabalpur

mixture were put into a glass container with 1000 ml of tap water. The root-soil mixture was vigorously mixed with a glass rod for 30 seconds. A 10-second pause enabled to settle heavier particles and organic material, the remaining soil-water suspension were slowly poured through a set of two sieves. The sieves used are those with pores of diameters of 0.5mm (the top one) and 0.045 mm (lowest one). Most spores retain on the 0.045 mm sieve. The extracts were washed away and spores collected from the sieves in to Petri dishes. Using a microscope, spores and aggregates were picked by means of dropper and needle. Selected spores were separated with a needle. A drop or two of (poly vinyl lacto glycerol) was spread on the centre of a clean and dry slide so as to hold cover slip. Spores were placed on the mount and the cover slip was placed gently by avoiding air bubbles. Such prepared slides were allowed to dry in a dust free chamber for 3-5 days. The edge of the cover slip was sealed with clear nail polish to prevent the desiccation and entry of air bubbles. Spores were examined under microscope and photographs were taken.

Analysis of Soil Samples

Soils constitute the weathered surface of the earth's crusts, which is mixed with organic material and microorganisms live and plants grow. Soil testing is one of the most important tools to determine the status of plant nutrients in a field. The air dried and sieved soil samples were analyzed for pH, Organic carbon, macro nutrients and micro nutrients.

Results and discussion

Table 1. Analysis of soil samples by different parameters

Soil sites	Water holding capacity	pH	Organic matter %	Arbscular Mycorhizal spore density/50gm soils	Species richness
Site 1	30.73	6.67	2.07	119	5
Site 2	32.81	6.72	0.98	73	3
Site 3	38.76	6.55	2.90	31	3
Site 4	36.60	6.62	1.97	101	3
Site 5	35.76	6.78	1.39	33	5
Site 6	34.40	6.56	3.56	94	2
Site 7	37.70	6.71	1.34	50	4
Site 8	38.20	6.81	0.41	53	3
Site 9	33.31	6.69	3.31	84	3
Site 10	30.36	6.66	3.15	76	4

Site1 RDVV campus, Site 2nd Sadar area, Site 3rd Gorigath, Site 4th Adhartal area, Site 5th Dumna park, Site 6th Hathital, Site 7th Gorabazar area, Site 8th Ranital area, site 9th SFRI Jabalpur, Site 10th TFRI Jabalpur.

The highest VAM spore density was measured from RDVV Campus of approximately 119 density/50 gram soils and least was noticed from site 3rd. Species richness was maximum from RDVV Campus and from site 5thDumna park, while site 2nd , 3rd, 4th and site 9th contains species richness of 3 each. Site 7th and 10th has species richness of 4 each. Lowest species richness was to be seen in site 6th. Organic matter was highest located from site 6th while lowest located from site 8th.

Table 2. VAM fungi associated with *Parthenium hysterophorus*

Sites	VAM fungi identified from *Parthenium hysterophorus*
RDVV Campus	*Glomus badium, Glomus geosporum, Sclerocystis corymeoides, Acaulospora laevis.*
Sadar area	*Glomus geosporum, glomus mosseae, Scetullospora jabalpurensis.*
Gaurighat area	*Glomus badium, Gigaspora margarita, Glomus fasciculatum, Acaulospora appendiculata, Glomus geosporum.*
Adhartal area	*Sclerocystis corymeoides, Glomus mosseae, Acaulospora rehmii, Gigaspora margarita.*
Dumna park	*Gigaspora margarita, Glomus fasciculatum, Glomus badium, Acaulospora laevis.*

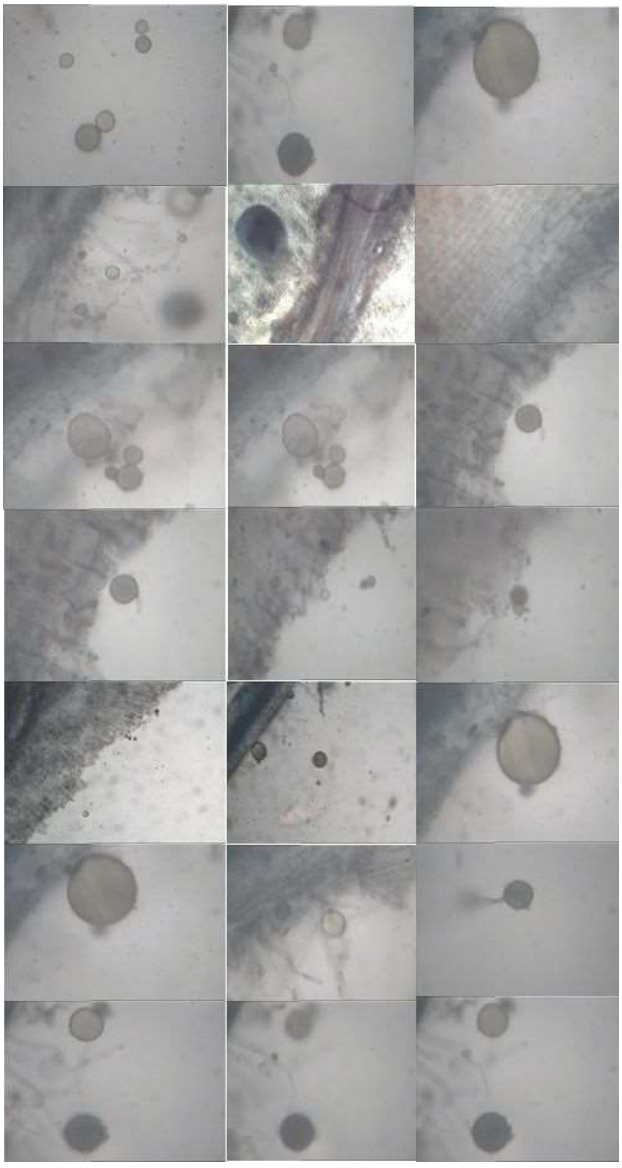

Figure 2. Pictures showing different species of VAM fungi isolated from *Parthenium hystophorus*

Glomus badium were isolated from RDVV campus area, Gauri ghat area and Dumna park *Glomus geosporum* were

isolated from RDVV campus, Sadar area and Gauri ghat area. *Sclerocystis corymeoides* were isolated from RDVV campus and Adhartal area. *Acaulospora laevis* from RDVV campus and Dumna park, while *Glomus mosseae* were isolated from Sadar area and Adhartal area. *Scetullospora* was recorded only from site Sadar area. *Gigaspora margarita* was found from site Gauri ghat, Adhartal and Dumna park. *Glomus fasciculatum* were found from Gauri ghat and Dumna park. *Acaulospora apendiculata* were to be seen only in Gauri ghat area. *Acaulospora* from Adhartal area only. Only genus *Glomus* seems to show high occurrences with this *Parthenium* species. It can be surveyed that the majority of species of genus *Glomus* may adapted a stronger mechanism of symbiosis with different plant hosts as some sort of a co-evolving mechanism. From the result *Glomus species* was the predominant genus, followed by *Acaulospora* Species. The predominant occurrence of *Glomus* is due to their potential to survive in any kind of area, which shows adaptation in any area of our Jabalpur region. Our findings were similar to the observations of Opik et al (2006). Our results also indicated that soil pH decreased with increasing soil depth, due to percolation of water in this region, leading to formation of acids. Spore population were having correlation with the PH of soil. Soil moisture also increased with increased depth. AMF fungi give negative relation with moisture content.

Conclusion

The highest VAM spore density was measured from RDVV Campus of approximately 119 density/50 gram soils and least was noticed from site Gauri ghat area. Species richness was maximum from RDVV Campus and from site Dumna park, while site Sadar, Gorabazar, Adhartal contains species richness of 3 each. Site Gorabazar area has species richness of 4 each. The result suggested that AM fungi are well distributed in *Parthenium* plant species in central Jabalpur area. Population of AM fungi, frequency of occurrences and their distribution varied with the soil depth, pH, and moisture content in that region.

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