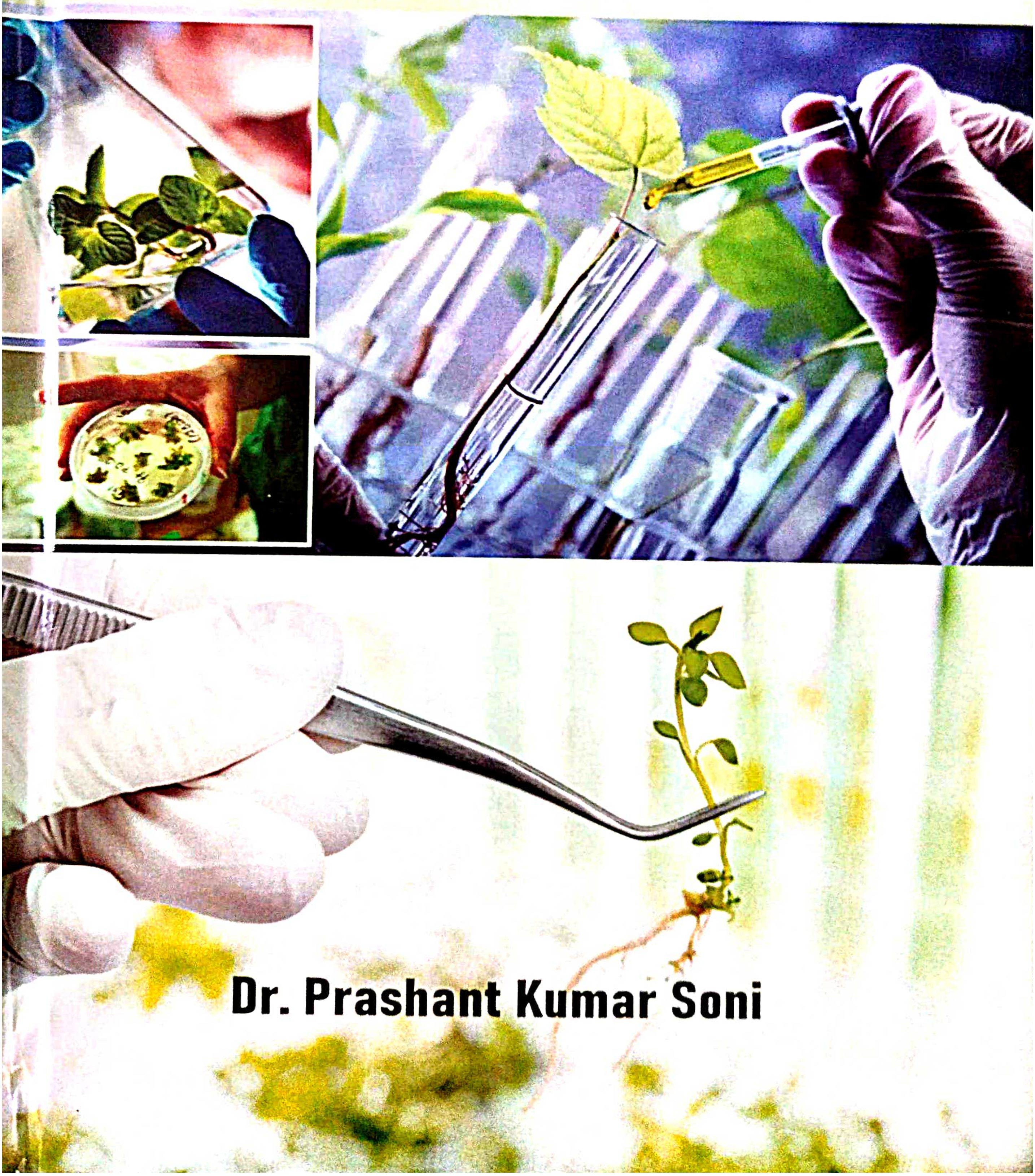
Medicinal Plants Research

Trends and Dimensions



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CHAPTER–2

Studies on AM Fungal Association in Selected Medicinal Plants of Kukru Hills in Betul District, MP, India

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& Prashant Kumar Soni[3]

ABSTRACT

In the present study we have determined occurrence and distribution of arbuscular mycorrhizal fungi (AMF) associated with different medicinal plants growing in Kukru hills of Satpura ranges of Betul district. In the present investigation seventeen medicinal plants belonging to nine families were surveyed for their mycorrhizal association.The result revealed that *Catharanthus roseus* of Apocyanaceae family showed greater percent root colonization as well as average AMF spore population. Thirty-one AMF species belonging to five genera were isolated during investigation. *Glomus* species were found dominating followed by *Acaulospora* species. *Acaulospora gerdemanii, Acaulospora scrobiculata, Glomus clarum, Glomus hoi,* and *Glomus mosseae* were found associated with all selected medicinal plants. Result also indicated that C. *roseus* could be used as natural source of mass cultivation of AM fungi.

Key Words: Arbuscule, Medicinal Plants, Mycorrhiza, Occurrence, Percent Root Colonization, Spore Count, Vesicle.

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INTRODUCTION

Mycorrhiza from the Greek terms myco (= fungus) and rhiza (= root), is the predominant root symbiosis. Majority of land plants species form symbiosis with soil borne fungi. In nature arbuscular mycorrhiza (AM) are the oldest and most widespread symbiosis. Recent fossil studies and molecular data have tracked the presence of this symbiosis all the way to the Ordovician era *i.e.*,to be at least 60 million years old (Redecker *et al.*, 2000). Arbuscular mycorrhizal (AM) fungal symbiosis are of great ecological importance. The ubiquitous AM fungi belonging to the phylum Glomeromycota (Terdosoo *et al*. 2018) colonize almost 72% of the plant species growing in the natural ecosystem (Brundrett & Tedersoo 2018).

The AM symbiosis aids in plant growth by increasing the availability as well as translocation of various nutrients especially phosphorous (P) (Rouphael *et al*. 2015).The main advantage of mycorrhiza is its greater soil exploration and increasing uptake of mineral element like P, N, K, Zn, Cu, S, Fe, Mg, Ca and Mn and the supply of these nutrients to the host roots. The pivotal involvement of arbuscular mycorrhizal fungi in plant mineral nutrition positions these fungi at the abiotic/biotic interface in ecosystems. AM fungi have been described as "keystone mutulists" in terrestrial ecosystem (O'Neill *et al.*, 1991).

Medicinal plants are widely recognized with high healing activity worldwide. The knowledge of medicinal plant is as old as man himself. From the dawn of civilization man has tried to find remedies against different ailments and revealed to use flowering plants for as medicine. Medicinal plants represent a rich source of antimicrobial agents. Many medicinal herbs possess some dietary supplements that are good sources of antioxidants and anti-inflammatory compounds.The world health organization (WHO) estimated 80 % of worldwide population used medicinal and aromatic plants for their medicinal therapy (Bharathy *et al.*, 2021).

In India, the use of different parts of several medicinal plants to cure specific ailments has been in vogue from ancient times. Medicinal plants play a dynamic role in traditional medicines. AM fungi are present in practically all soils and associated with a great variety of plants of different taxonomic groups.

Originally medicinal plants in India were reported to be non-mycorrhizal due to the presence of various secondary metabolites. Though arbuscular mycorrhizal fungal status of many medicinal plants has been investigated (Mohan *et al.*, 2005; Sundar *et al.*, 2011). AM fungal symbiotic association can be efficiently utilized for greater production of medicinal plants and, in turn, the plant metabolites. AMF symbiosis attributed to favourable characteristics of medicinal plants, by improving the production and accumulation of important active ingredients such as terpenes, phenols, and alkaloids. The association also optimized the composition of different active ingredients in medicinal plants and ultimately improved the quality of herbal materials and total yields.

A large number of medicinal plants have been reported by different researchers to harbour AMF. On the contrary such study has not been conducted in this study area. Furthermore, there have been reports that AMF symbiosis plays a positive role in the accumulation of alkaloids in some important medicinal plants, such as reserpine in *Rauwolfia serpentina* and vinca alkaloids in *Catharanthus roseus* which have important anticancer functions (Mishra 2008; Rosa-Mera *et al.*, 2011). Considering the benefits accrued from AMF symbiosis with medicinal plants it is paramount to utilize these fungi as bio-fertilizer for their cultivation.

Therefore, the present study was undertaken to examine the occurrence and distribution of AM fungal association with the rhizospheres of some commonly grown medicinal plants species belonging to different families in Kukru hills of Satpura region.

MATERIALS AND METHODS

Study Site

The study was conducted at Kukru hills (Kukru as the 'Crow flies') area in Bhainsdehi block of Betul district of Madhya Pradesh, India between the latitude of 21°29'42.3" N and longitude 77°27'40.7" E. Kukru is a scenic place known for coffee farms, windmills and jungle camping experiences. Kukru is marginally located at southern part of the state which is adjacent to Gugamal National Park (Chikhaldara) and Melghat Tigher Reserve of Amravati district of Maharashtra State.The site is located at an altitude of 1137 m above sea level with a relative humidity of 60-65%. The climate of the region is sub-tropical monsoon and the year is divisible into a mild winter (Oct.-Feb.), a mild hot summer (March-June), and a cold rainy season (July-Sept.).

The area is filled with undulated strips under Satpura plateau, and the entire hill is covered with various types of plants *viz.* herbs, shrubs, trees, climber and lianas. The area is very rich in medicinal plants owing to the occurrence of different forest types viz. Southern tropical moist deciduous teak forests, southern tropical dry deciduous forests, dry teak forests, Boswellia forests, bamboo forests and tropical thorn forests (Kumar A & Khanna KK 1998).

Collection of Root and Soil Samples

Seasonal field trips were performed from 2020 to 2021, in order to collect soil and fine root samples for assessment of AM diversity associated with some medicinal plants (Table 1) found in the Kukru region. Rhizospheric soil samples and fine terminal roots of different plants were collected by digging out small amount of the soil close to the plant roots up to the depth of 15-30 cm, and stored in sterilized polythene bags at 4-10°C for further processing of mycorrhizal assessment in the laboratory. Five sets of roots and rhizospheric soil samples were thoroughly mixed and a composite sample was taken for analysis.

Soil samples were collected in the adjoining regions of the roots as most spores were reported to be concentrated in this zone.

Isolation, Quantification and Identification of AM Spores

Isolation of AM spores were done by using 'Wet sieving and decanting technique' of Gerdemann and Nicolson (1963). Sieves of different sizes *i.e.*,150μm, 120μm, 90μm, 60μm and 45μm are used. 100 gm of composite soil sample was dissolved in water. After stirring, soil solution was allowed to settle down over night. Decanting water on a series of sieves in following order 150μm, 120μm, 90μm, 60μm and 45μm from top to bottom on which spores were trapped. The trapped spores were transferred to Whatman filter paper No.1 by repeated washing with water. Then spores were picked up by hypodermic needle under stereo binocular microscope and mounted in polyvinyl lactic acid alcohol (PVLA).

AM spores were counted by Gridline intersect method' proposed by Gaur and Adholeya (1994) under stereo-binocular microscope at 60x magnification. Mycorrhizal spores were identified according to their spore morphology by conventional taxonomic identification manuals of Walker (1983); Schenck and Perez (1990); and Mukerji (1996).

Assessment of AM Fungal Colonization

Mycorrhizal root colonization was done by 'Rapid Clearing and Staining Method' of Phillips and Hayman (1970). The collected roots were cut into 1cm segments and then 15 - 30 segments are selected randomly. These roots segments were cleaned in 10% KOH (24 hours), acidified with 1% HCl (20 minutes) and stained with trypan blue stain for 24 hours. After this root segments were destained with lactophenol for a day to remove excess of stain. Now roots were mounted in lactic acid: Glycerol (1:1) solution and examined for AM colonization. Evaluation of root colonization was done by root slide technique of Giovannetti and Mosse (1980).

The percentage of AM root colonization was determined as under :-

$$\text{Percent root colonization} = \frac{\textit{number of root segment colonized}}{\textit{Number of root segments}} \times 100$$

Statistical Analysis

All statistical analysis was followed by Banerjee (2006).

RESULT

Structural Features of AM Fungi

Data presented in (Table 1) shows general information and important properties of selected medicinal plant species. AM fungi are worldwide in distribution and forms symbiotic association. Most of the ecosystems harbour AM fungi as one of the main constituents of soil microbiota and form symbiotic association with roots of most terrestrial plants. Overall, 17 medicinally important herbaceous plant species belonging to different families were surveyed for the mycorrhizal association. In the present investigation, AM fungi showed wide range of variability in terms of root colonization and spore density. There are many reports of mycorrhizal occurrence, status and diversity in medicinally important plants of India (Sundar *et al.* 2011; Chouhan *et al.* 2013; Kumar *et al.* 2019).

The degree of arbuscular mycorrhizal fungal association is depicted in (Table 2). Significant occurrence was observed in arbuscule, vesicle and hyphal structure of AM fungi from rhizospheric zone of all selected plants. The AM fungi form various structures like mycelium, arbuscules, hyphal and arbusculate coils, and vesicles within and outside of plant roots.

Different types of mycelium like Y-shaped, H-shaped, coiled, beaded and parallel mycelia were reported in the roots of different plants. In some cases, extensive mycelial growth was also observed. Different shapes of vesicles also occurred. Vesicles forms varies from elliptical, round, globose, oval, beaked and elongated.

Moreover, there is a great difference in the distribution pattern of AM fungal structures in plant roots. Mycelium form is absent in three plants *Mentha viridis, Ocimum basilicum,* and *Rauwolfia serpentina*. Vesicles were found in *Adhatoda vasica, Ageratum conyzoides, Mentha viridis, Ocimum basilicum, Solanum nigrum, Tridax procumbans,* and *Withania somnifera*. Out of 17 medicinal plants, Arbuscular type of infections were observed in few plants like *Catharanthus roseus, Eclipta alba, Euphorbia geniculate, Euphorbia hirta, Mentha viridis, Ocimum basilicum, Physalis minima, Rauwolfia serpentina, Tridax procumbans,* and *Withania somnifera*.

Adhatoda vasica, Ageratum conyzoides and *Solanum nigrum* were infected with mycelium and vesicles structures. *Aloe barbadensis, Asparagus racemosus* and *Phyllanthus niruri* were found infected only with mycelium whereas *Achyranthus aspera* was infected only with vesicles. Such variation in colonization pattern were also seen in *Catharanthus roseus, Eclipta alba, Euphorbia*

Table – 1: General Information of Medicinal Plants Selected for Studying Mycorrhizal Association

S. No.	Plant Species	Vernacular/ Common Name	Family	Nature	Ailments against used
1.	*Achyranthus aspera* L.	Apamarga, Adha-jara, Latjira,	Amaranthaceae	Annual Erect Herb	Asthma, abscess, eye disease, fever, hysteria, piles, diarrhea, dysentery and uterine disease
2.	*Adhatoa vasica* Nees.	Malabar nut, Adusa, vasaka	Acanthaceae	Evergreen Herbaceous Shrub	Cough, Asthma, Bronchitis. It is also used to cure whooping cough and tuberculosis
3.	*Ageratum conyzoides* L.	Billygoat weed, Goat weed	Asteraceae	Annual Erect Herbaceous	Its leaves paste is very useful in wound healing, leprosy treatment, diarrhea, dysentery, intestinal colic, rheumatism and fever
4	*Aloe barbadensis* Mill.	Ghritkumari	Liliaceae	Succulent with Basal Rosette and Creeping Rhizome	The gel or mucilage is a wound- healing agent and shows antibacterial, antifungal and anti-inflammatory properties. *Aloe* gel are also used for burns, scalps, sunburns and wounds
5	*Asparagus racemosus* L.	Sparrow grass, Satavari	Asparagaceae	Perennial Climber	Root powder administered orally as aphrodisiac, also used in pain of stomach, urinary disorders, Nervous disorders, inflammation, liver diseases, infectious diseases, ulcers, cancer
6.	*Catharanthus roseus* (L.) G. Don.	Madagascar periwinkle, Sadabahar	Apocyanaceae	Herbaceous Annual	Used as 'Folk medicine' in diabetes, Children's leukemia, Hodgkin's disease. Catharanthus have anti-cancer and antitumor properties
7.	*Eclipta alba* Roxb.	Bhrangraj, Ghamara	Asteraceae	Erect & Slender	Used as hair tonic, eye disease, spleen enlargement, night blindness.
8.	*Euphorbia geniculata* Orteg.	Chagul, Put putti	Euphorbiaceae	Herbaceous	Used as 'Folk medicine' in Snake bites and Scorpion sting.

(Contd...)

S. No.	Plant Species	Vernacular/ Common Name	Family	Nature	Ailments against used
9.	*Euphorbia hirta* L.	Dudhi,	Euphorbiaceae	Prostrate Herb	Piles, diarrhea, asthma, worms, bowel complaints.
10.	*Mentha viridis* L.	Mint, Pudina	Lamiaceae	Perennial Herbaceous	Leaves and roots given in fever & bronchitis, oil is used for rheumatism and a decoction is used as lotion in aphthae
11.	*Ocimum basilicum* L.	Sweet Basil, Kali tulsi	Lamiaceae	Erect & Aromatic herb	The leaf juice is applied to cure scabies & other cutaneous diseases. Infusion of leaves is given as a remedy for gastric trouble, piles, habitual constipation, flu, sores, colds & bronchial infection. Leaves are mosquito repelent, expel worms and treat ring worms.
12.	*Phyllanthus niruri* L.	Jar-amla, Bhui-amla	Euphorbiaceae	Erect Herb	Used in Jaundice and liver complaint; cuts and wounds, chronic dysentery, tubercular ulcers.
13.	*Physalis minima* L.	Little Gooseberry, Chirpati, Rasbhari	Solanaceae	Erect Herbaceous Annual	All parts of the plant are used as a diuretic and antipyretic. The fruit is said to be alterative, appetizer, bitter, diuretic, laxative and tonic. Extracts from the plant have shown anticancer activity
14.	*Rauwolfia serpentina* Benth. ex. Kurz	Chandrika, Sarpgandha	Apocyanaceae	Small Erect Herb	High blood pressure, insomnia, insanity, tranquilizers, hyper tension, depression, and Snake bite
15.	*Solanum nigrum* L.	Makoi	Solanaceae	Erect & Perennial	It has expectorant, analgesic, sedative, diaphoretic properties. Its external application cures skin diseases and gives relief in burns, itching, pain etc. Leaves juice used in earache
16.	*Tridax procumbans* L.	Baramasi, Lugharia	Asteraceae	Prostrate Herb	*Tridax* has antimicrobial and antiseptic properties. Leaf juice directly used skin diseases, cuts and wounds, boils, blisters, eczema, leprosy and diarrhea
17.	*Withania somnifera* Dunal.	Asvagandha, Asgandha	Solanaceae	Erect Perennial Herbaceous	Asthma, bronchitis, ulcers, debility, inflammation, rheumatism and chest complaints.

Table - 2: Mycorrhizal Status of Studied 17 Medicinal Plants of Satpura Region

S. No.	Medicinal Plants	ASP (Mean ± SD)	Root Mycorrhization			
			Percent RC	A	V	H
1.	*Achyranthus aspera* L.	441.0 ± 24.72	56.0 ± 5.10	-	-	+
2.	*Adhatoda vasica* Nees.	375.0 ± 15.50	38.0 ± 6.29	-	+	+
3.	*Ageratum conyzoides* L.	590.0 ± 23.14	44.0 ± 4.40	-	+	+
4.	*Aloe barbadensis* Mill.	165.0 ± 24.25	52.0 ± 7.42	-	-	+
5.	*Asparagus racemosus* L.	140.0 ± 18.00	30.0 ± 9.25	-	-	+
6.	*Catharanthus roseus* (L.) G. Don.	783.0 ± 30.34	75.6 ± 1.27	+	-	+
7.	*Eclipta alba* Roxb.	740.0 ± 26.00	70.2 ± 2.41	+	-	+
8.	*Euphorbia geniculata* Orteg.	212.0 ± 17.54	42.8 ± 3.97	+	-	+
9.	*Euphorbia hirta* L.	401.0 ± 31.74	36.0 ± 3.08	+	-	+
10.	*Mentha viridis* L.	440.0 ± 24.70	50.0 ± 5.64	+	+	-
11.	*Ocimum basilicum* L.	378.0 ± 23.88	56.4 ± 3.21	+	+	-
12.	*Phyllanthus niruri* L.	64.0 ± 19.18	10.0 ± 5.88	-	-	+
13.	*Physalis minima* L.	534.0 ± 24.09	64.6 ± 3.85	+	-	+
14.	*Rauwolfia serpentina* Benth. ex. Kurz	544.8 ± 10.62	57.6 ± 5.59	+	-	-
15.	*Solanum nigrum* L.	662.4 ± 35.21	54.8 ± 3.97	-	+	+
16.	*Tridax procumbans* L.	468.0 ± 32.72	61.8 ± 2.87	+	+	+
17.	*Withania somnifera* Dunal.	561.0 ± 41.30	58.0 ± 11.52	+	+	+

(Percent RC) Percent Root Colonization; (A) Arbuscules; (V) Vesicles; (H) Hyphae alone; (+) Present; (-) Absent.
(ASP) Average Spore Population of 100g soil from the corresponding plant rhizosphere.
(Mean±SD) Each value is the mean of five observations.

Table -3 : Arbuscular Mycorrhizal Fungal Spore Diversity Associated with Rhizospheric Soil of Medicinal Plants of Satpuda Hills.

S. No.	AM Fungal Species	Medicinal Plant Species*																
		1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
1.	*Acaulospora bireticulata*	-	-	+	-	-	+	-	-	+	-	-	-	+	+	-	-	-
2.	*Acaulospora elegans*	-	-	-	+	+	+	+	-	-	+	-	-	+	-	-	+	+
3.	*Acaulospora foveata*	+	-	-	-	-	+	+	-	-	-	-	-	-	-	-	+	+
4.	*Acaulospora gerdemanii*	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
5.	*Acaulospora lacunosa*	-	-	+	-	-	-	+	-	-	-	-	-	-	-	+	-	-
6.	*Acaulospora laevis*	+	-	-	-	-	-	-	+	-	-	+	-	-	-	+	-	-
7.	*Acaulospora margarita.*	-	-	-	-	-	-	+	-	+	-	-	-	+	-	-	+	-
8.	*Acaulospora mellea*	-	-	+	-	-	+	+	-	-	+	+	-	-	-	-	-	+
9.	*Acaulospora scrobiculata*	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
10.	*Acaulospora sp.*	-	-	-	-	-	-	-	-	+	-	-	-	-	-	+	-	-
11.	*Acaulospora sp.*	-	-	-	+	-	+	+	+	-	-	-	-	-	+	+	+	-
12.	*Gigaspora albida*	-	-	+	-	-	+	+	-	-	+	-	+	-	-	-	-	-
13.	*Gigaspora rosea*	+	+	-	-	-	-	+	-	+	-	-	-	-	-	+	+	-
14.	*Glomus ambisporum*	-	-	+	-	-	-	-	+	-	-	+	-	-	-	+	-	-
15.	*Glomus clarum*	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
16.	*Glomus constrictum*	+	-	-	-	-	+	+	+	-	+	-	-	+	-	-	-	+
17.	*Glomus fasciculatum*	-	+	-	-	-	+	+	+	-	-	-	-	-	+	-	+	+
18.	*Glomus falvum*	-	+	-	-	-	+	-	-	+	-	+	-	-	+	+	-	-
19.	*Glomus geosporum*	-	-	+	-	-	-	+	-	-	+	-	-	-	-	-	-	+
20.	*Glomus hoi*	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
21.	*Glomus heterosporum*	-	-	-	-	-	+	+	-	-	-	+	-	-	-	+	+	-
22	*Glomus intraradices*	+	+	+	-	-	+	-	-	+	+	-	-	+	-	-	-	-
23.	*Glomus macrocarpum*	-	-	+	-	-	+	+	-	-	-	-	-	-	-	-	-	+
24.	*Glomus mosseae*	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
25.	*Glomus pallidum*	-	-	-	-	-	-	-	-	-	+	-	-	-	-	+	+	-
26.	*Glomus pustolatum*	-	-	-	-	-	+	+	-	-	-	-	-	-	+	-	-	-
27.	*Glomus reticulatum*	+	+	+	-	-	+	+	+	-	-	+	-	+	+	+	+	+
28.	*Glomus segmentatum*	-	-	-	-	+	-	-	-	+	-	-		-	-	-	-	+
29.	*Glomus sp.*	+	-	-	+	-	+	-	-	-	-	-	-	-	-	+	-	+
30.	*Sclerocystis sp.*	-	-	+	-	+	+	+	-	-	-	-	-	+	-	-	-	-
31.	*Scutellospora minuta*	-	-	+	-	-	-	+	-	-	-	-	-	-	-	+	-	-

Medicinal plant species name listed in table 1 and only their serial number shows here.

geniculata, E. hirta, and *Physalis minima* plants have mycelial and arbuscular infection, while *Tridax procumbans* and *Withania somnifera* both credited with all kind of infections *i.e.,* mycelia, vesicular and arbuscular structures.

Mycorrhizal Colonization

The result obtained from this study suggests that percent root colonization of arbuscular mycorrhizal fungi varies from plant to plant. The data of percent root colonization shows variation and minimum was recorded in *Phyllanthus niruri* (10.0 ± 9.88 %) and maximum was recorded in *Catharanthus roseus* (75.6 ± 1.27 %). *Eclipta alba* was observed as second most colonized host plant with 70.2 ± 2.41 % of AMF infection. Occurrence of high level of percent root colonization is a sign of better fungal- root interaction.

AMF Spore Population

Similar variation was also observed in average population of AMF spores extracted from the soil of corresponding plant rhizosphere.The result also showed that highest AMF spore count was observed with *Catharanthus roseus* (783.0 ± 30.34) and lowest was observed with *Phyllanthus niruri* (64.0 ± 19.18). Other medicinal plants showed intermediate range of average AM fungal spore count between higher and lower range.

Distribution of AM Fungi

It is clearly evident from the result that number of AM fungi belonging to order Glomales were found associated with different medicinal plants. A total of 31 AM fungal species belonging to five known genera of order Glomales were identified. *Glomus* was the dominant genus and have 16 species followed by *Acaulospora* with 11 species, 02 species of *Gigaspora* and one species of each *Sclerocystis* and *Scutellospora* was observed.

Among the 31 different AM fungi, the most dominant mycorrhizal species were *Acaulospora gerdemanii, Acaulospora scrobiculata, Glomus clarum, Glomus hoi,* and *Glomus mosseae* found to associated with rhizosphere of all selected medicinal plants.

The AM spores diversity was observed maximum in *Catharanthus roseus* (21 species) followed by *Eclipta alba* (19 species) and *Solanum nigrum* (18 species), while minimum spore diversity was recorded in *Phyllanthus niruri*.

DISCUSSION

AM fungi are worldwide in distribution and have been found on numerous plant species including many of medicinally importance. The result obtained from the study suggests that the colonization, average number of vesicles, spore population and AM fungal species differ with different medicinal plants. There are many reports of mycorrhizal occurrence, status and diversity in medicinally important plants of India (Sundar *et al.* 2011; Chouhan *et al.* 2013; Kumar *et al.* 2019).

The presence of arbuscules is normally used to designate AM association, but the presence of hyphae or vesicle has also been used as evidence for these associations. Generally, arbuscules are ephemeral in structures that may be absent. It is due to effect of when samples were collected, roots were inactive whereas vesicles are considered as storage organ produced in the older region of infection.The morphology of AM fungi chiefly falls into major types based on the colonization patterns: Arum-, Paris- and intermediate types. The Arum-type is represented by intercellular hyphae and arbuscules, on contrary, Paris-type is characterized by intracellular linear hyphae, hyphal coils, and arbusculate coils. The intermediate type of AM morphology shares both the characters of Arum- and Paris- types (Dickson 2004).

Colonization by native AM fungi in some medicinal plants has been reported earlier (Mishra 2008; Soni 2008; Vyas *et al.*, 2008). The high level of percent AM root colonization is a sign of better fungal- root contact and that increased benefits from AM fungal symbiosis (Vogeti *et al.* 2008). The AM root colonization is a dynamic process, which is influenced by several edaphic factors such as nutrients status of soil, seasons, AMF strains, soil temperature, soil pH, host susceptibility to AM colonization and feeder root condition at the time of sampling. The quality and type of AM propagules also affected the dynamics of root colonization, which were also increased by increasing the age of the plant (Chandra and Jamaluddin 1999). However, Saif and Khan (1975) found increased root colonization during the period of maximum vegetative growth of the plants. This being directly influenced by the number of spores on the soil.

During the present investigation except root exudation, other conditions are almost same because all the selected medicinal plant species were collected from the region of Kukru hills of Betul. So, any influence of aforementioned factor would have negligible effect regarding mycorrhizal association. Therefore, root exudation and other soil microbiota might influence the root colonization among these plants. It is reported that root exudation by plants influenced occurrence of mycorrhizal species (Walker *et al.*, 2003). The extent of root colonization may vary with host plant, growing season, edaphic factors and environmental factors (Husband *et al.* 2002; Muthukumar & Udaiyan 2002). The mycorrhizal root colonization has been reported to be affected by seasonal spore production, seasonal alterations and nutrient accessibility in the soil (Borde *et al.* 2010). The soil temperature and pH have positive influence on AM association, brings changes in physiology of association. The present studies revealed that the percent root colonization of surveyed plants could not be related to spores numbers and its diversity. Similar observation was also made earlier while studying AM fungal diversity associated with some plant species of Vindhyan region of Madhya Pradesh (Vyas *et al.* 2008; Mishra *et al.* 2008).

In our studies, a total of seven different medicinal plant species were lacks arbuscules in their root regime. Arbuscules are usually observed in vegetative growth stage of host plant due to availability of new cortical cell for infection and to cope up with high nutrient requirement (Mosse 1981). So, hyphal coil performs the potential role of arbuscules as suggested. Variations in AMF development are in accordance (Thaper *et al.*, 1992) attributed by differential preference of AM fungi to host plant, difference in quality and quantity of root exudates of the plant in the soil (Eom *et al.*, 2000). The differential nutrient requirements of host plants may have direct effect on spore density and frequency of mycorrhizal colonization (Senapati *et al.*, 2000). Phosphorous deficiency and spore degradation by other soil organism are also responsible for variation in AM infection among members of same family. Moreover, availability of root with poor architecture to AM fungus for colonization might be a reason for inadequate fungal mass development.

AM fungal taxa *Glomus* was highly abundant in the rhizosphere of all medicinal plants and are in agreement with the results obtained earlier (Mishra 2008).Vyas *et al.*, 2006 and Mishra *et al.*, 2008 reported the dominance of *Glomus* in the agricultural soil and forest land of Vindhyan plateau of Sagar region of Madhya Pradesh. Our results corroborate well with the findings of other investigators, who reported dominance of *Glomus* sp. while, studying the biodiversity of AM fungi (Vyas *et al.* 2006; Chouhan *et al.* 2013; Kumar *et al.* 2019).The dominancy of *Glomus* species has also been reported by AM fungi occurring throughout the world.

Acaulospora species was second dominant genus and found to be associated with medicinal plant commonly growing in acidic soil. Occurrence of high AM spore density might be favoured by the conducive edaphic conditions for sporulation like low nutrient status (Boerner 1990), optimal moisture, high aeration and the uninterrupted conditions of the soils.AMF species can colonize all potential hosts and some mycorrhizal species are more desirable to contest for one host than another, even then they may be able to infect the host only under ideal conditions (Laxman *et al.*, 2001). High species richness in the rhizosphere of host plant might be associated with organic matter that may assist root colonization of specific host plant.

The greater colonization and spore count of AM fungi in roots of *Catharanthus roseus* might be playing an important role for its wide spread on the plateau of the Kukru hills. *Catharanthus roseus* better known for its dimeric indole alkaloids *viz.*vinblastine and vincristine have high anti-tumour potentials. Medicinal value in the plant depends upon the availability of nutrients on soil where they growing and mycorrhization may influence the synthesis of alkaloids (Mishra *et al.*, 2006). Overall, it can be used as a source of mass cultivation of AM fungi under natural condition and its roots pieces can be also used as a source of inoculum.

CONCLUSIONS

It can be concluded from the present study that all medicinal plants harbour mycorrhizal association. However, diversity of arbuscular mycorrhizal fungi species differ in different medicinal plant and the extent of AMF infection is controlled by the host plant as well as environmental factors. AM spore density was found to be maximum in wildly grown medicinal plant as compared to cultivated plant species. These observations could be attributed by seasonality, edaphic factors, age of host plants, the sporulation abilities of AMF and host dependence. The abundance of *Glomus* and *Acaulospora* sp in the soil makes it more favoured AM fungi for the mass multiplication and can be utilized for increasing growth and productivity of medicinal plant. Moreover, this type of investigations may also be important while studying the effect of different anthropogenic activities on the AMF. From practical point of view, the use of a species with widespread distribution implies that mycorrhizal inoculum produced with one or many species can potentially be used under different soil and climatic conditions.

Conflict of interest: All authors declare that they have no conflict of interest.

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