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Diversity of AM (Arbuscular mycorrhizal) Fungi in Wheat Agro-climatic Regions of India

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Abstract

AM (Arbuscular mycorrhizal) fungi are important yet unknown components of biodiversity in the agricultural fields in India. To study their diversity and habitat relationships, we characterized and enumerated spores in 55 fields of wheat from 11 agro-climatic regions of India, varying in climatic and edaphic characteristics. The AM fungal spore count, species richness, most frequent species, and intra-radical colonization were studied in various samples drawn from these regions. A total of 165 samples were collected at the time of the wheat harvest. These samples were used as trap cultures and multiplied in a green house for a period of one year, which yielded 34 species scattered over 6 genera. The genera *Glomus* Tulasne & Tulasne occurred most frequently, constituting 89.1% of the total species. The number of species in a given region ranged from 1–9. *Glomus albidum* Walker & Rhodes and *G. macrocarpum* Tulasne & Tulasne were found to be the most commonly occurring species.

Keywords: AM fungi; Diversity; Wheat; Agro-climatic regions

Introduction

AM (Arbuscular mycorrhizal) fungi are ubiquitous in soils around the globe and have been associated with improved plant growth for over 100 years. These symbiotic fungi are the main components of the soil micro biota in most of the agro-ecosystems and account for 25% of the biomass of the soil micro flora and micro fauna combined [1]. They are generally known to increase the absorption and translocation of mineral nutrients from the soil to the host plant [2], to improve the tolerance of the host plant towards biotic [3] and abiotic stresses [4], and to build up the macro-porous structure of the soil that allows penetration of water as well as air and prevents erosion [5].

The beneficial effects of AM fungi on plants and soil health prompted great interest in their research. The diversity of AM fungi has been extensively studied in natural ecosystems and its importance for plant diversity, productivity and ecosystem processes has also been recognized. However, little is known about AM fungal diversity in agricultural lands. Agricultural lands are artificial ecosystems that are subjected to constant human intervention. Whereas, in natural ecosystems the internal regulation of functions is a product of plant biodiversity through the flow of energy and nutrients, and this form of control is progressively lost under agricultural intensification [6]. Modern agriculture implies simplifying the structure of the environment over vast areas, and replacing nature's diversity with a small number of cultivated plants and domesticated animals. Several workers noted that the diversity of AM fungal communities tends to decrease when natural ecosystems are converted into agro-ecosystems and their diversity decreases as the intensity of agricultural inputs increases [7,8].

With a geographic area of 328.048 million hectares, stretching between 80N and 360N latitude and between 680E and 980E longitude, its altitude varying from the mean sea-level to the highest mountain ranges of the world, India presents a range and diversity of climate, flora and fauna, with a few parallels in the world. Thus, with many climates and varieties of soils, India affords scope for much diversity in agriculture. The country may be broadly divided into five agricultural regions (Rice Region, Wheat Region, Millet-Sorghum Region, Temperate Himalayan Region, and Plantation Crops Region). To work out the suitable cropping plan for field crops, the country is broadly divided into 15 agro-climatic zones based on physiognomy and climate. It is evident from their effects upon soil health and host plant growth that AM fungi are an important part of sustainable agricultural systems. Modern, intensive agricultural practices, such as chemical fertilization and pest control, continuous monoculture, and tillage impact AM fungal and plant interactions. These practices affect mycorrhizae both quantitatively and qualitatively. Thus, describing the diversity of the community of AM fungi at a site becomes, therefore, an important step in determining the effects of agricultural treatments upon AM fungi and the eventual development of management regimes for these fungi.

However, only 14 AM fungal species were recorded from the agricultural fields of India [9,10] with only 4-5 species being from the wheat fields [10]. These studies were either conducted at single or relatively few study sites. Although such findings are invaluable while invaluable in increasing knowledge about the ecology of AM fungi, they are limited in application as they are largely based on site-specific research. Studies on a larger scale are needed to allow robust prediction of the relative importance of different agro-climatic regions for these fungi.

In India, wheat, the second important food crop, is grown on 27 million hectares out of the total 114 million hectares of land under cultivation [11]. The wheat region occupies most of the northern, western and central India and fields from these regions were selected for the study. To the best of our knowledge, there is no detailed study on the diversity of AM fungi in these regions. Our study primarily aims at improving the understanding of the broad-scale distribution of AM fungi in different wheat-growing agro-climatic regions of India. To achieve this, we sampled the spores of AM fungi in 55 study fields,

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and analyzed the distribution of some of the more commonly recorded species and the overall number of species in relation to different agro-climatic regions. This study will help in exploiting the potential of these fungi in sustainable agriculture, particularly in wheat.

Materials and Methods

General study area

The study was established within the northern, western, and central regions of India (Figure 1), which form the major wheat-growing belt and contribute two-thirds of the 45 million tonnes of the total production of wheat [12]. The area included a range in soil types and climate. More specifically, the study area formed a rectangle bound by the longitudes 72035'E and 82009'E, and the latitudes 21014'N and 28054'N, encompassing agro-climatic regions of the states in India of Uttar-Pradesh, Madhya Pradesh, Haryana, Rajasthan, and Gujarat.

Strategy for selecting field sites

Out of 18 states, which are divided into agro-climatic regions (Planning Commission, working group report on agricultural research

Figure 1: Sample collection sites in northern, western, and central India.

and education for the formulation of 8th five year plan, government of India 1989), we identified 5 states, which are major wheat- growing zones, and a total of 11 agro-climatic regions were selected. The basis of the division of agro-climatic regions is essentially based on climate, soils, and existing cropping patterns of each state as a unit. These parameters have been shown to influence patterns in the occurrence of plants and animals [13,14].

From each region, one or two replicates were selected and for each replicate, five collection fields were chosen (Table 1), thus totaling 55 collection fields. Wherever possible, it was possible that the fields were at least 500 m apart to allow for the collection of independent data. The location and elevation of each site was determined from fine scale (1:25000) topographic maps. To minimize heterogeneity with respect to agricultural practices and inputs, only the fields having similar agricultural practices (wheat under cultivation for more than five years and tillage with tractor), fertilizer inputs (120-150 Kg/ha [kilogram per hectare] N [Nitrogen], 40-60 Kg/ha P [Phosphorus], 40-60 Kg/ha K [Potassium], 25 Kg/ha Zn [Zinc]) and irrigation inputs (four to five irrigations) were selected.

Sampling AM fungi

For sampling purposes, each collection field was divided into four blocks. From each block, five undisturbed core samples (500 g each) were collected (soil and roots) from the rhizosphere of wheat plants from a depth of 0-30 cm using a core sampler at the time of wheat harvest. Thus, a total of 20 soil cores (54) were collected from each collection field. The samples were air-dried in the shade to the point where there was no free moisture and were placed into zip bags, and stored at 40˚C in a cold room until processed. The samples were used for three different purposes: (1) propagation of AM fungal isolate of each collection field for their characterization, (2) analysis of AM fungal parameters, and (3) analysis of soil chemical parameters.

Agro-climatic regions	Replicates*	Place	Region code	Soil type	No. of sites
Bhabar and Tarai Zone of Uttar Pradesh (28^{0}22'N latitude 79^{0}25'E longitude)	1	Pantnagar	Bht	Clayey loam	5
Western Plain Zone of Uttar Pradesh (28^{0}40'N latitude 77^{0}43'E longitude)	3	Dadri Gajroula Ghaziabad	Wep 1 Wep 2 Wep 3	Loamy sand Sandy loamy Silty loamy	5 5 5
Mid-Western Plain Zone of Uttar Pradesh (28^{0}02'N latitude 79^{0}07'E longitude)	1	Budaun	Miw	Clayey loam	5
Gird Zone of Madhya Pradesh (21^{0}14'N latitude 81^{0}38'E longitude)	1	Chambal	Giz	Light alluvium	5
Malwa Plateau Zone of Madhya Pradesh (22^{0}05'N latitude 82^{0}09'E longitude)	1	Pachmari	Mal	Medium alluvium	5
Semi-Arid Eastern Main Zone of Rajasthan (26^{0}55'N latitude 75^{0}48'E longitude)	1	Jaipur	Samz	Loamy sand	5
South Gujarat Zone of Gujarat (23^{0}02'N latitude 72^{0}35'E longitude)	1	Ahmedabad	Sgz	Clayey	5
Western Zone of Haryana (28^{0}54'N latitude 76^{0}35'E longitude)	2	Rohtak Gual-Pahiri	Whz 1 Whz 2	Fine loam Alluvium	5 5

*Within each combination of the agro-climatic region we selected five sites. Hence, for each of these regions sampled only once, there were five sites. For two of the agro-climatic regions, we replicated twice or thrice in each of the agro-climatic categories.

Table 1: Distribution of the 55 study sites in relation to agro-climatic regions of India.

Propagation of AM fungi in trap cultures

Previous studies of studying the distribution and abundance of AM fungi have largely been based on recording AM fungal species at the time of sample collection. Several issues must be considered when traditional taxonomic identification of spores is used to describe the AM fungal community diversity. First, the number of spores in the soil may not reflect the relative amount of colonization of roots by this fungus or the amount and distribution of hyphae in the soil. Second, non-sporulating species may be present [15]. A fungus may be a significant member of the 'vegetative' community, but because of the date of sampling, local environment, or host plant regulation of carbon expenditure, be unable to produce spores yet able to persist to the following year as infective hyphae in the roots or the soil. Third, spores collected from the field may be difficult to identify as a result of degradation of the spore walls [16]. The non-sporulating species can often be coaxed to sporulate in 'trap cultures'. As these fungi are obligatory mycotrophs, propagation of AM fungal cultures require their growth in association with a living plant.

Plastic trays (460290240 mm^3) were used to establish AM fungal cultures in a greenhouse using soil samples from all collection fields. For each collection field, four plastic trays (comprising four blocks) having 10mm hole at the bottom were prepared. A 20 mm thick drainage mat (Enkadrain ST, Schoellkopf AG, CH-8057 Zurich, Switzerland) was placed at the bottom of each tray and the tray was filled with 25 kg of substrate (50% Terragreen [American aluminium oxide, oil dry US special, Type IIIR] and 50% soil sediment having Olsen P=1.56 ppm; Organic C=0.28%; Total N=0.052% and K=52.66 ppm). The substrate was autoclaved at 1200˚C for one hour at 15 psi (pounds per square inch) before filling. Substrate cores (500 g) were taken out from five different places (four corners and centre) in each tray and were replaced by five undisturbed soil cores (500 g, containing collection field's AM fungi) to inoculate the hosts. Seeds of *Allium cepa* (onion), *Tagetus* spp. (marigold), *Daucus carotus* (carrot), *Medicago sativa* (alfalfa), and *Trifolium alexandrianum* (berseem) were pre-germinated. Five pre-germinated seeds of each species were placed on top of the five soil cores. The plants were watered to a moisture level of approximately 60% of the water-holding capacity and were grown in a greenhouse at 20 ± 5˚C with 60% relative humidity. The pots were arranged on a greenhouse bench in a completely randomized design with three replications. Half-strength Hoagland's nutrient solution [17] was provided to the plants at fortnightly intervals. After four months of the growth cycle, the pots were left to dry undisturbed at a fairly stable temperature so that the drying period would not be too rapid. After completion of the growth cycle the dried shoots were cut at the ground level without disturbing the substrate and pre-germinated seeds of different hosts; *Gossypium* spp. (Cotton), *Vetiveria zizanioides* (Vetiver), *Vigna radiata* (mungbean), *Sorghum* spp., and *Tagetus* spp. (marigold) were sown again. After completion of each growth cycle, rhizosphere soil cores were taken from the vicinity of plants growing in trap cultures at a depth of 0–15 cm and species characterization was done.

AM fungal spore isolation, quantification, and characterization

The spores were quantified and characterized at three different time intervals at the time of sample collection (collection field soil), after the first and second trap culture cycle. The spores were isolated following the modified technique of wet sieving and decanting [18] and were quantified as given by Smith and Skipper [19]. The diagnostic slides of different species of AM fungal spores were prepared. The initial observation of spores (colour, shape, etc. in water) was recorded under stereomicroscope (Leica) occlusions.

The measurement of AM fungal spores (spore diameter, wall thickness, hyphal thickness and thickness at the attachment point) was done using an image analyzer system (Image Pro-Plus, 4.0 version) attached to a compound microscope (Olympus BH 2). Once the data on the spores was generated they were characterized up to the level of species using the manual for the identification of VA mycorrhizal fungi [20]. Some species were also characterized by the monograph [21], the INVAM (http://invam.caf.wvu.edu/fungi/taxonomy/speciesID) and BEG (http://www.kent.ac.uk/bio/beg/) websites, and the existing published taxonomic literature. Collections, which could not be thus determined, were characterized up to the genus level and labelled sp. 1, sp. 2, and so on. The collections, which were characterized up to the species level, were deposited at CMCC (Centre for Mycorrhizal Culture Collection), TERI, India, which has been designated as the mycorrhizal germplasm culture collection by the Department of Biotechnology, Government of India.

Intraradical colonization by AM fungi

The roots were cleared and stained by the technique by Phillips and Hayman [22] and percentage colonization was calculated according to Biermann and Lindermann [23] at three intervals of time: at the time of sample collection (collection field soil) and after the first and second trap-culture cycle.

Soil chemical parameters: The collection field soils were analyzed at the time of sample collection for their chemical parameters. A soil suspension of 1:2.5 (soil-to-water mixture) was made. The pH of the soil suspension was measured by a digital pH meter (Expandable Ion Analyser EA 940, Orion Research) and the electrical conductivity was measured by a digital electrical conductivity meter (controlled dynamics). A protocol by Datta et al. [24] was followed for measuring the percent organic carbon. The percent total nitrogen was calculated using Kjendahl's method by Bremner [25]. The available phosphorus was determined using Olsen's method [26] and the estimation of available potassium was done using a flame photometer with filters [27].

Data analysis: We analyzed AM fungi community structure using the following ecological parameters: population abundance at each site, mean population abundance, total population abundance at each site, species richness at each site, species diversity at each site, species evenness at each site, and species dominance at each site. Population abundance at each site was defined as the sum of individuals of a particular species, counted at each site during all the observations. Mean population abundance was obtained by averaging the population abundance of sites only where a specific species was found. Total population abundance was calculated as the population abundance of all component species at each site. Species richness was expressed by the number of species found in each study site during the observation period. Species diversity at each site was expressed by Shannon-Wiener function. H'=Si=1 pi, Where S is the number of species in each site. Species evenness was expressed by the Shannon equitability index, J'=H'/In S, where H' is the Shannon-Wiener function and S is the number of species in each site. Species dominance was expressed by McNaughton's dominance index, D1=(n1+n2)/N, where n1 and n2 is the population abundance of the first and second dominant species, respectively.

Inter-regional differences in richness, diversity, dominance and the effects of soil chemical properties on their scores, were analysed by means of ANOVA (Analysis of Variance) with the software costat and means were separated using DMRT (Duncan's Multiple Range Test) at 5% level and significance (LSD, P=0.05). It should be noted that data are spatially autocorrected {that is, two points close to each other will

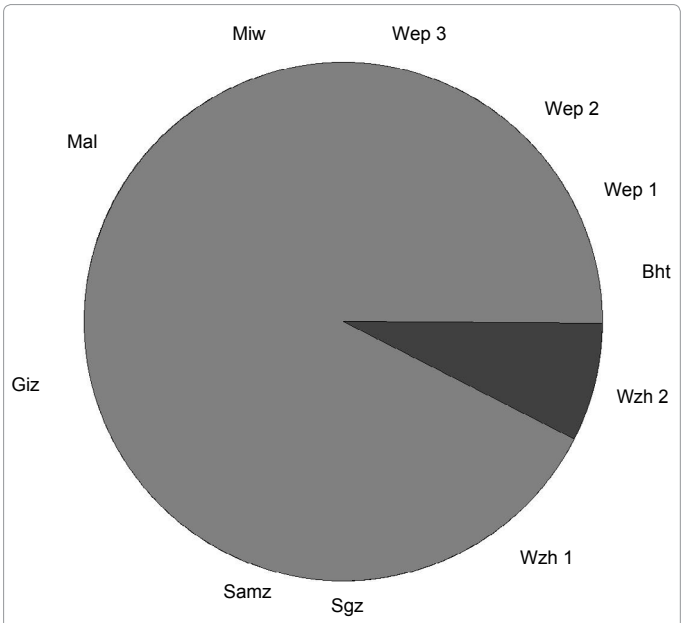

Figure 2: AM fungal species richness observed in different agro-climatic regions of India.

Region code	pH	Electrical conductivity (dSm^{-1})	% Organic carbon	% Total nitrogen	Olsen's phosphorus (ppm)	Available potassium (ppm)
Bht	7.45 b	0.30 b	0.68 cd	0.07 a	5.73 cd	57.80 d
Wep1	8.62 a	0.75 a	0.30 d	0.02 c	11.11 b	75.80 cd
Wep 2	7.39 b	0.54 b	1.78 a	0.02 c	19.23 a	216.60 a
Wep 3	7.55 b	0.37 cd	0.66 cd	0.02 c	7.56c	52.40 d
Miw	6.65 c	0.22 d	0.39 d	0.02 c	6.16 cd	92.40 cd
Mal	7.14 b	0.19 d	1.14 a	0.02 c	4.85 d	61.40 d
Giz	7.41 b	0.16 d	1.72 a	0.02 c	2.66 d	133.20 b
Samz	8.22 a	0.20 d	0.43 d	0.03 c	5.91 cd	104.20 bc
Sgz	7.26 b	0.82 a	0.87 bc	0.04 b	5.36 cd	71.20 cd
Whz 1	7.41 b	0.21 d	0.55 cd	0.03 b	4.89cd	72.80 cd
Whz 2	7.45 b	0.42 bc	0.58 cd	0.02 c	5.49 cd	70.60 cd
LSD (P=0.05)	0.43	0.18	0.36	0.01	3.33	35.41
F value	11.97	14.21	16.33	17.34	14.89	14.62
Level of significance	***	***	***	***	***	***

Bht, Pantnagar; Wep 1, Dadri; Wep 2, Gajroula, Wep 3, Ghaziabad; Miw, Budaun; Mal, Pachmari; Giz, Chambal; Samz, Jaipur; Sgz, Ahmedabad; Whz 1, Rohtak; Whz 2, Gual Pahiri.

Table 2: Comparison of soil chemical properties in different agro-climatic regions.

be less independent of each other than two points located at a larger distance from each other [28,29]. This would lead to a pseudoreplication problem [30], if each point were to consider as an independent unit. Multiple regression procedures allow for the generation of models for conditioning on the contribution of macronutrients of sampling units to the variation in the studied variables [29]. Coefficient of correlation (r^2) was calculated between different soil chemical features and mycorrhizal parameters.

Results and Discussion

This is the first study that has attempted to describe the diversity and distribution of AM fungi in the wheat agro-climatic regions of India. Because the sampling effort was standardized, conducted within a relatively short time frame during the wheat harvesting period and the trap cultures established were studied for successive trap culture cycles, the occurrence and the number of fungal taxa across the agro-climatic regions can be compared meaningfully. Although isolation from the spores has the advantage that these may belong to an identified fungus and result in a single-species isolate. However, not all AM fungi produce sufficient quantities of spores in field soils to allow for isolation or identification, so trap cultures can reveal species not observed to sporulate in soils [31]. Trap culturing methods often produce more healthy spores than the soils from which they originated, but they usually result in a mixture of species which changes with subsequent cultures generations [16,32].

Many host plants were chosen as trap plants. Grasses and legumes are known hosts for the successful propagation of AM fungi and hence were included as trap plants. *Sorghum* was used mainly because of its compatibility with many fungal species in all genera (except *Sclerocystis*) originating from a wide range of habitats [16]. Marigold was chosen as a trap plant as it gives a good root biomass and is a good host for the propagation of AM fungi. These host plants have been used historically in the experimentation of the mycorrhizal phenomena. Onion and subterranean clover (*Trifolium subterraneum*) are also used widely as a culture host [33]. Sometimes, the number of species isolated into trap cultures exceeded those identified from field-collected spores, suggesting the inaccuracy of fungal surveys based solely on spore observations.

Overall, 34 species and 165 isolates of AM fungi were recorded in the 55 study fields scattered in 11 different wheat-growing agro-climatic regions of India. These regions exhibited variable trends. Of all the study regions, maximum AM fungal diversity i.e., 15 species were recorded in the Gird Zone of Madhya Pradesh, which is characterized by light alluvial soil and only one species of AM fungi was recorded from the Western-Plain Zone (Wep 2, Gajroula) and the Mid-Western Plain Zone of Uttar Pradesh, associated with sandy loam soil (Figure 2). Table 2 shows the results of soil chemical analysis. The role of soil nutrient concentration on the diversity and colonizing ability of AM fungi was also investigated during the course of the study. In the present study, the spore density was found to be related to the phosphorus concentrations (r^2=-0.51) and organic carbon. Thus, regions with low phosphorus concentrations and high organic carbon exhibited higher spore count and diversity. There was also a positive correlation between spore density and the number of AM fungal species. However, whether the increased spore population or increased diversity confers more benefits to plants is a matter of further investigation.

A major highlight of our study is the low diversity of AM fungi in the agricultural fields. The genus *Glomus* Tulasne & Tulasne was recorded as the most frequently occurring (89.1%), followed by *Gigaspora* Gerdemann & Trappe emend. Walker & Sanders (10.9%), *Scutellospora* Walker & Sanders (9.09%), and *Entrophospora* Ames & Schneider (3.6%). Spores of *Glomus albidum* Walker & Rhodes (30%), *G. macrocarpum* Tulasne & Tulasne (25%) and *G. mosseae* (Nicolson & Gerdemann) Gerdemann & Trappe (20%) were amongst the frequently occurring species (Figure 3). Table 3 depicts the AM fungal species recorded in different agro-climatic regions of India. Each region contained some characteristic species and displayed a characteristic pattern of population The spores of *G. fulvum* (Berk. & Broome) Trappe & Gerdemann, *G. clarum* Nicolson & Schenck and *G. dimorphicum* Boyetchko & Tewari were very rare which were observed

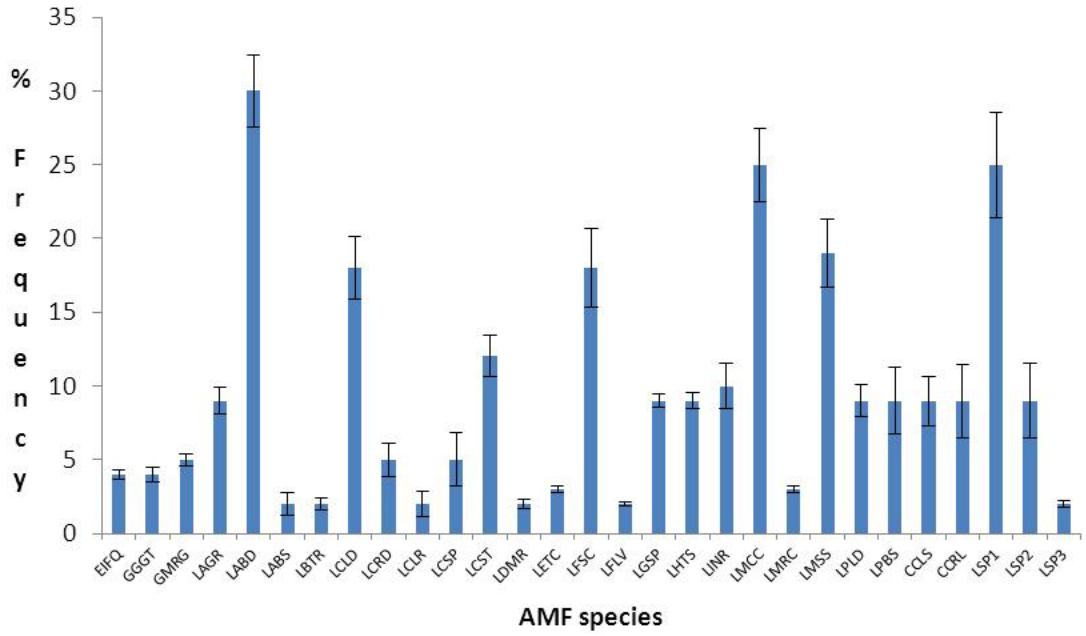

Figure 3: Percentage frequency of AM fungal species observed in different agro-climatic regions of India.

AM fungal species diversity	*CMCC accession number*	*Collection fields where AM fungal species were reported*
Entrophospora infrequens (Hall) Ames & Schneider	AM WE01	Wep 3a*, Wep 3e*
Gigasporagigantea (Nicolson & Gerdemann) Gerdemann & Trappe	AM WGI02	Sgz b*♦, Sgz c*♦, Sgz d*, Sgz e*♦
Gigaspora margarita Becker & Hall	AM WGI03	Wep 2d*, Samz a♦, Samz b*, Samz c*♦, Samz d*, Samz e♦
Glomus aggregatum Schenck & Smith emend. Koske	AM WG04	Mal a*♦, Mal b*♦, Mal d*♦, Mal e*♦, Giz b*♦, Giz c♦, Giz d*♦, Giz e♦, Wzh 2b*, Wzh 2c*♦
G. albidum Walker & Rhodes	AM WG05	Bht d♦ , Bht e*, Wep1a*, Wep1b*, Wep1d♦, Wep 1e*, Wep 2a*, Wep 2e*, Miw b*, Miw d♦, Wzh 1a*♦, Wzh 1c*, Wzh d*♦, Wzh 2a*, Wzh 2c*, Wzh 2d*♦, Wzh 2e*♦
G. ambisporum Smith & Schenck	AM WG06	Giz a*♦, Giz b *♦, Giz c♦, Giz d*♦, Giz e*♦
G. botryoides Rothwell & Victor	AM WG07	Giz a*♦, Giz b*♦, Giz c♦, Giz d*, Giz e*♦
G. caledonium (Nicolson & Gerdemann) Trappe & Gerdemann	AM WG08	Wep 3a♦, Wep 3b*♦, Wep 3c*♦, Wep 3d*, Wep 3e*, Mal a*♦, Mal b*♦, Mal d♦, Mal e*♦, Giz a*, Giz b*♦, Giz c*♦, Giz d♦, Wzh 1a*♦, Wzh 1e*♦
G. claroideum Schenck & Smith	AM WG09	Wep 1a*, Wep 1b*, Wep 1c*, Wep 1d*, Wep 1e*
G. clarum Nicolson & Schenck	AM WG10	Mal a*
G. clavisporum (Trappe) Almeida & Schenck	AM WG11	Giz a*♦, Giz d*, Wzh 1a*, Wzh 1b*, Wzh 1c*♦
G. constrictum Trappe	AM WG12	Wzh 1c*
G. dimorphicum Boyetchko & Tewari	AM WG13	Giz a♦, Giz b*
G. etunicatum Becker & Gerdemann	AM WG14	Sgz a*♦, Sgz b*♦, Sgz c*♦, Sgz d*♦, Sgz e♦, Wzh 1a*, Wzh 1c*♦, Wzh 1d*
G. fasciculatum (Thaxter) Gerdemann & Trappe emend. Walker & Koske	AM WG15	Bht a*♦, Bht b*♦, Bht c*♦, Bht d*♦, Bht e*♦, Wep 2b*♦, Wep 2c♦, Wep 2e♦, Wep 3c*♦, Wep 3d*♦, Wep 3e*♦, Miw a*, Miw b♦, Miw c♦, Miw d♦, Wzh 1e*♦
G. fulvum (Berk. & Broome) Trappe & Gerdemann	AM WG16	Mal a♦, Mal d*, Giz a♦, Giz c*, Giz d*,
G. geosporum (Nicolson & Gerdemann) Walker	AM WG17	Mal b♦, Mal d♦, Mal e♦, Giz a*♦, Giz c*♦, Giz d, Giz e*♦, Samz a*♦, Samz b*♦, Samz c*, Samz d*♦, Samz e*
G. heterosporum Smith & Schenck	AM WG18	Giz a*, Giz c*♦
G. intraradices Schenck & Smith	AM WG19	Bht a*♦, Bht b*♦, Bht c*♦, Bht d*♦,Bht e*♦, Wep 1a*♦, Wep 1b*♦, Wep 1c*♦, Wep 1d*♦, Wep 1e*♦, Wep 3a*♦, Wep 3b*♦, Wep 3c*♦, Wep 3d*, Wep 3e*♦, Miw a♦, Miw b♦, Miw c♦, Miw d♦, Miw e♦, Mal a*♦, Mal b♦, Mal c♦, Mal d*♦, Mal e*♦, Giz a*, Giz b*♦, Giz c*♦, Giz d*♦, Giz e*♦, Samz a*♦, Samz b*, Sam c*♦, Samz d*♦, Samz e*, Wzh 1a♦, Wzh 1b*♦, Wzh 1c*, Wzh 1d*, Wzh 1e*♦, Wzh 2a*♦, Wzh 2b*♦, Wzh 2c*, Wzh 2d*, Wzh 2e*♦
G. macrocarpum Tulasne & Tulasne	AM WG20	Giz a♦, Giz b♦, Giz c*♦Giz c*♦, Giz d*, Wzh 1a*♦, Wzh 1b*♦, Wzh 1c*♦
G. microcarpum Tulasne &Tulasne	AM WG21	Giz a*
G. monosporum Gerdemann & Trappe	AM WG22	Mal a*, Mal e*
G. mosseae (Nicolson & Gerdemann) Gerdemann & Trappe	AM WG23	Bhtc♦, Bhtd♦, Bhte*, Wep 2c*♦, Wep 2d♦, Wep 2e♦, Wep 3a♦, Wep 3b♦, Wep 3c♦, Wep 3e♦, Wzh 1a♦, Wzh 1b*♦, Wzh 1c*♦, Wzh 1d*♦
Glomuspallidum Hall	AM WG24	Wep 3d*
G. pubescens (Sacc. & Ellis) Trappe & Gerdemann	AM WG25	Giz c*
Scutellosporacalospora (Nicol. & Gerdemann) Walker & Sanders	AM WS26	Wep 3c*, Wep 3d*, Wep 3e*, Miw c*, Miw d*, Miw e*, Mal e♦, Giz c*♦, Giz d♦, Giz e*♦, Wzh 1b*♦, Wzh 1d*
S. coralloidea (Trappe, Gerd. & Ho) Walker & Sanders	AM WS27	Giz d*, Wzh 2a*♦, Wzh 2b*♦, Wzh 2c*, Wzh 2d*, Wzh 2e*♦

Table 3: AM fungal species diversity in samples collected from different regions of India at the time of sample collection (*), after the first trap culture cycle (♦) and after the second trap culture cycle (♣).

in only one sample out of the 55 samples studied. *G. ambisporum* Smith & Schenck and *G. botryoides* Rothwell & Victor were observed only in Giz region. Likewise, each region contained its characteristic most frequent species and varied with respect to the agro-climatic region. *Glomus intraradices* Schenck & Smith was found to sporulate in most of the trap cultures even if it is absent at the time of sample collection. The photographs of the recorded AM fungal species are documented in Figures 4-6.

Although a total of 34 AM fungal species were recorded from 11 agro-climatic regions of India, an average of only 3-4 species per field was found. Another striking observation was the ubiquity and adaptability of a few genera and species in the field.

These findings indicate that the species present in the agricultural fields, which are constantly subjected to human intervention, are a restricted subset of those which would occur naturally in the region. Two possible explanations could account for this pattern. First, the diversity of AM fungal communities has been related to the diversity of plant communities [34,35]. Although most natural ecosystems contain a great variety of plant species, intensively managed agro-ecosystems contain very few plant species, generally a single crop per field with occasional weeds. Second, cultural practices probably exert strong selective pressures on AM fungal communities [36]. All the fields chosen for study are conventionally managed fields, which integrate the use of commercial seed-bed preparation, mechanized planting and inorganic inputs i.e., chemical fertilizers, pesticides, etc. All these cause changes in the habitat and substrate availability that may discourage the growth of selected microorganisms, so fungal species and strains most tolerant to these stresses proliferate. During the present study, the genera *Glomus* was found to be ubiquitous. The comparatively higher frequency of some species, *G. albidum* (33%) and *G. macrocarpum* (25%) among the spore communities indicates their adaptability to varied soil conditions, whereas other species showed a narrow range of their host/environment adaptation.

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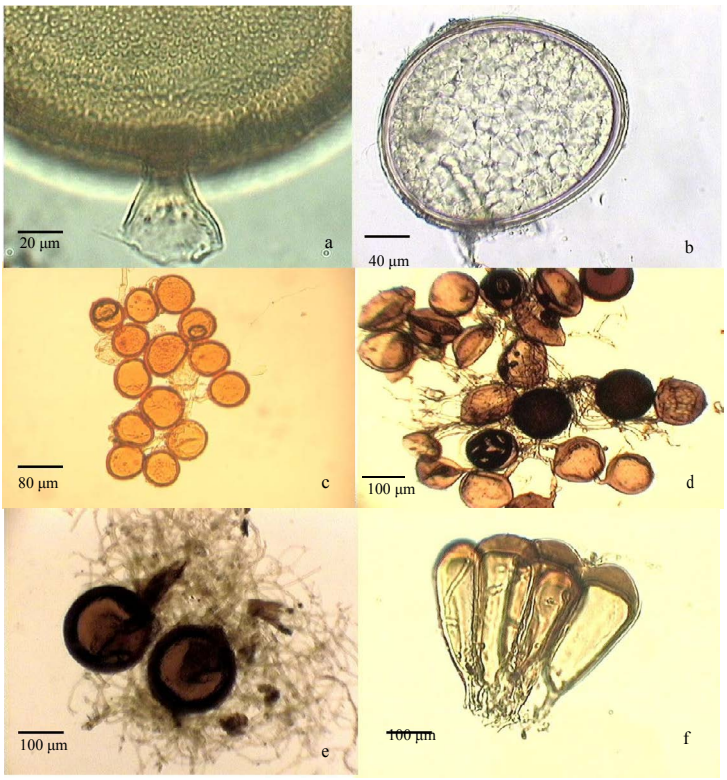

Figure 4: Representative AM fungal species observed in different agro-climatic regions of India. a) Azygospore of *Entrophospora infrequens* (40×), b) Chlamydospore of *Glomus albidum* (20×), c) Loose sporocarp of *G. aggregatum* (4×), d) Sporocarp of *G. ambisporum* (10×), e) Chlamydospores of *G. botryoides* (10×), f) Sporocarp of *G. clavisporum* (10×).

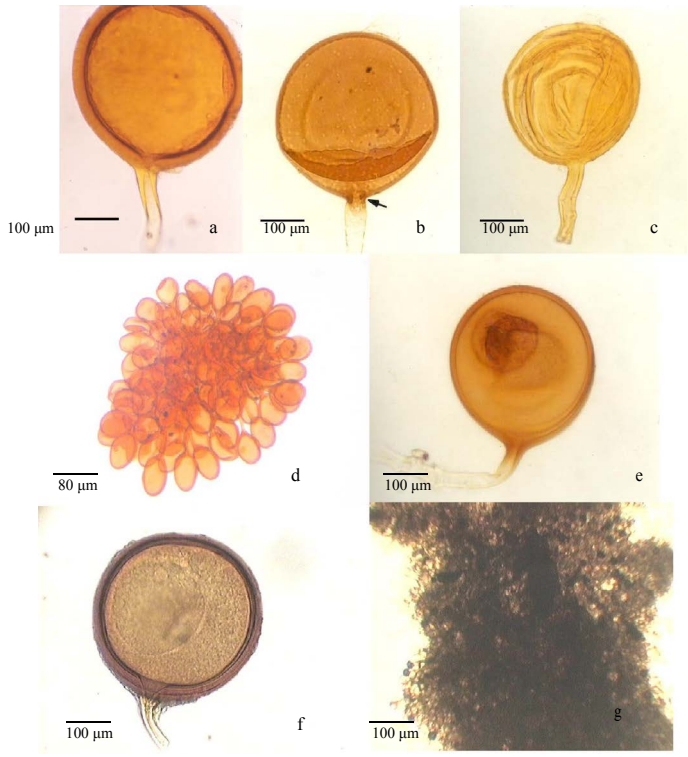

Figure 5: Representative AM fungal species observed in different agro-climatic regions of India. a) Chlamydospore of *G. caledonium* (10×), b) Chlamydospore of *G. constrictum* (10×), c) Chlamydospore of *G. fasciculatum* (10×), d) Sporocarp of *G. fulvum* (4×), e) Chlamydospore of *G. macrocarpum* (10×), f) Chlamydospore of *G. mosseae* (10×), g) Sporocarp of *G. pubescens* (10×).

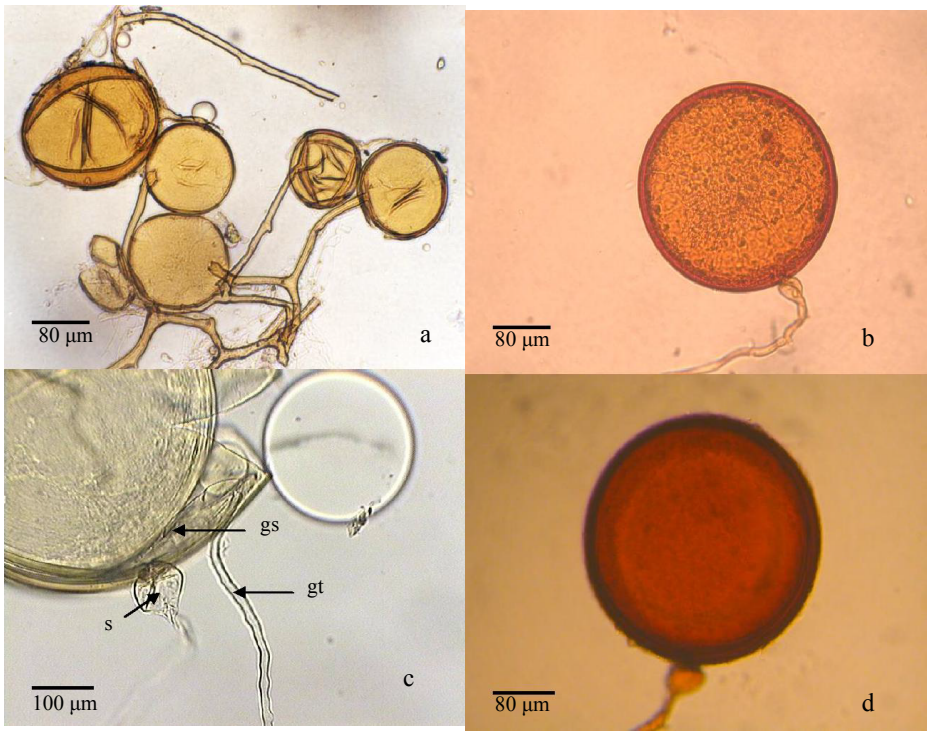

Figure 6: Representative AM fungal species observed in different agro-climatic regions of India. a) Broken sporocarp of *G. intraradices* (4×), b) Azygospore of *Gigaspora margarita* (4×), c) Azygospore of *Scutellospora calospora* (10×), d) Azygospore of *Scutellospora coralloidea* (4×).

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