# **Fungal biodiversity: Distribution, conservation and prospecting of fungi from India**

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The variety and galaxy of fungi and their natural beauty occupy prime place in the biological world and India has been the cradle for such fungi. Only a fraction of total fungal wealth has been subjected to scientific scrutiny and mycologists have to unravel the unexplored and hidden wealth. One third of fungal diversity of the globe exists in India. Out of 1.5 million of fungi, only 50% are characterized until now. Unfortunately, only around 5–10% of fungi can be cultured artificially. Fungi are not only beautiful but play a significant role in the daily life of human beings besides their utilization in industry, agriculture, medicine, food industry, textiles, bioremediation, natural cycling, as biofertilizers and many other ways. Fungal biotechnology has become an integral part of the human welfare.

#### **Diversity spectrum**

THE number of fungi recorded in India exceeds 27,000 species, the largest biotic community after insects<sup>1</sup>. The true fungi belong to kingdom Eukaryota which has four phyla, 103 orders, 484 families and 4979 genera. The eighth edition of *Dictionary of the Fungi*<sup>2</sup> has recognized eleven phyla. The Deuteromycotina is not accepted as a formal taxonomic category. The number of fungal genera reported from the world and that from India between 1905 and 1995, are shown in Table 1.

About 205 new genera have been described from India, of which 32% were discovered by C. V. Subramanian of the University of Madras. Of these, approximately 27,000 species are reported to colonize diversified habitats<sup>1</sup>. This indicates a ten-fold increase in the last 70 years. Mano-harachary and his co-workers<sup>3</sup> have added 12 new genera, 60 new taxa and 500 new additions to fungi of India. The fossil record of fungi dates back to the early phanerozoic and into the proterozic geological era<sup>4</sup>. The existence of fossil fungi indicates their evolutionary significance besides solving certain phylogenetic complexities.

Major groups of fungi are discussed briefly to highlight the extent of diversity and this is followed by the examples of habitats that are unique and deserve greater attention.

#### Mastigomycotina

Fungi belonging to mastigomycotina form a prevalent group of fungi in water. They comprise of members Chytridiomycetes, Hyphochytridiomyctes and Oomycetes and colonize diverse habitats, such as water, humid soils, insects, keratin, chitin, angiospermic tissue, pollen grains and others, and live either as saprophytes or parasites. Such fungi have been arbitrarily grouped in this sub-division on the basis of zoospore and oospore and comprise 204 genera and 1160 species. Chytridiomycetous fungi occur as saprobes on plants and animal remains in water while other members occur as parasites on algae and aquatic animals. Considerable information on the morphotaxonomy, ecology, physiology, methodology and activities of flagellate fungi has been comprehensively compiled<sup>5-7</sup>. The Hyphochytridiomycetes are those aquatic fungi whose thallus is holocarpic or eucarpic, monocentric or polycentric and their vegetative system is rhizoidal or hypha-like with intercalary swellings. Sparrow<sup>7</sup> has discussed various aspects of this group of fungi. The Oomycetes contain 74 genera and 580 species, which are mostly aquatic and live as parasites or saprophytes. Das Gupta<sup>8</sup> and Manoharachary<sup>6</sup> have made detailed studies of the floristics, taxonomy and ecology of aquatic fungi from India.

Table 1.	• Fungal genera	
Phyla	World	India
Myxomycotina	450	380
Mastigomycotina	308	205
Zygomycotina	55	50
Ascomycotina	2000	745
Basidiomycotina	357	232
Deuteromycotina	4100	468
Total	7270	2080

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Chytridiales is the largest and least understood order of the Chytridiomycetes that contains over 80 genera, which are poorly described; the families and genera in this group are not phylogenetically delimited<sup>9</sup>. The Oomycetes are the largest group of heterotrophic Stramenopiles, most of which are inhabitants of freshwater, and humid soils; some are however indwellers of salt water habitats besides being parasitic (Figures 1 and 2). Barr<sup>10</sup> had recognized a new order Spizellomycetales under Chytridiomycetes. The fungi belonging to this order occur in soil and are seldom found in strictly aquatic habitats. Altogether 260 zoosporic fungi are reported from India which include, predominantly members of Oomycota with some chytrids. Manoharachary<sup>5</sup> has classified aquatic fungi into low-temperature species, moderate-temperature species, constant species and high-temperature species.

#### Marine and mangrove fungi of India

Fungi are cosmopolitan in oceans and estuaries and occur commonly on decomposing organic matter such as drift and intertidal wood. Initial studies of marine fungi in India were mostly confined to marine sediments and mangrove mud. An extensive survey of marine fungi from the west coast of India, particularly Maharashtra coast, was made by Borse<sup>11</sup> and Raghukumar<sup>12</sup>.

One-fourth of the world's coastline is dominated by mangroves, which are distributed in 112 countries and territories comprising about 181,000 sq km<sup>13</sup>. Mangroves constitute the second most important ecosystem among the marine ecosystems in productivity and sustain yield of coral reefs<sup>14</sup>. Mangrove forests generate considerable amount of detritus such as leaf litter, woody debris and inflorescence<sup>15</sup> and hence constitute an ideal habitat for many detritus-dependent fauna and microbes. Plant–fungus

ratio is one of the important yardsticks to estimate the richness and diversity of fungi of a region<sup>16</sup>. Many postulations have estimated global fungal population (May, 2000) between 0.5 and 9.9 million species<sup>17</sup>. Plant–fungus ratio in tropics has been predicted as 1 : 33 against 1 : 6 in temperate regions<sup>16,18</sup>. Mangrove fungi are the second largest group among the marine fungi<sup>19</sup>. However, the current pattern of assessment of higher fungi in mangrove habitats is mainly oriented towards assessment of typical marine fungi and the rest, e.g. freshwater, terrestrial, aero-aquatic fungi, are neglected.

Indian peninsula comprises about 7000 sq km of mangroves, out of which 70, 18 and 12% are distributed in the east coast, Andaman-Nicobar Islands and west coast, respectively<sup>20</sup>. Mangrove forests of India are dispersed in tropical as well as subtropical conditions (estuarine, deltaic and small-large near shore-off shore islands). Unique conditions prevail in these habitats that are responsible for detritus generation, accumulation, processing and turnover. For instance, heavy rainfall in Western Ghats (May-June) results in flushing freshwater and sediments to mangrove habitats and leads to decline of salinity to zero. During post-monsoon until January, salinity increases up to about 50%  $(17.5\%)^{21}$ . Such conditions are favourable for freshwater fungi to exploit mangrove substrata. Live and dead twigs of mangrove canopy harbour terrestrial fungi and addition of such substrata into mangrove waters in monsoon<sup>15</sup> results in domination of terrestrial fungi for several months<sup>22</sup>. Recent study of west coast mangroves revealed that nearly one-third of wood-inhabiting fungi belong to terrestrial group<sup>22</sup>. Endophytic fungi of mangrove leaves, stem and roots consist of more terrestrial than marine fungi<sup>23,24</sup>. In mangrove habitats, at least up to six months under low salinity, freshwater and terrestrial



Figure 1. Allomyces cystogenus R. Emers. 400 × (Chytridiomycota).CURRENT SCIENCE, VOL. 89, NO. 1, 10 JULY 2005



Figure 2. Oogonium with oospores of Achlya sp.  $1000 \times (Oomycota)$ .

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fungi are involved in litter conditioning. In summer (February–May), increased salinity supports the activity of mainly marine fungi on detritus. Core group fungi (frequency, 10%) on woody debris differ between west and east coast of India largely due to difference in diversity of mangrove plant species<sup>25,26</sup>.

Techniques of assessment of substrata have significant role in occurrence of fungi. Plating methods usually result in isolation of terrestrial fungi, damp-incubation helps marine and terrestrial fungi, and bubbling-chamber incubation facilitates recovery of freshwater hyphomycetes<sup>23</sup>. Incubation of leaf and woody detritus up to eight weeks resulted in dominance of terrestrial fungi, whereas sporulating marine fungi reached a peak at about 16 weeks; arenicolous (sand-inhabiting) fungi appeared after 16 weeks of incubation<sup>27</sup>. Thus, if interval of observation is too long, many sporulating anamorphic taxa may disappear.

If the host plant species is endemic, its fungal component seems to have restricted distribution. Hence, loss of such plants results in total elimination of host-specific fungi from the ecosystem. Kandelia candel (Rhizophoraceae) has been recorded only in two locations of the west coast, while Heritiera fomes (Sterculiaceae) and Nypa fruticans (Palmaceae) were found in one location of east coast of India<sup>28</sup>. Such endemic or endangered plants need special attention for mycological survey as mangroves are threatened by human interference. Recent checklist of mangrove fungi revealed that a total of 625 fungi exist at global scale (278 ascomycetes, 277 anamorphic taxa, 30 basidiomycetes and 14 oomycetes) $^{29}$ . A rough estimate reveals that about 150 species of mangrove fungi (onefourth of globally known) have been reported from the mangroves of the Indian subcontinent. The above global and Indian scenario on mangroves provides unique opportunities for mycologists to explore fungal diversity and exploit their ecological, medicinal and industrial potential.

# Zygomycotina

These fungi reproduce asexually by sporangiospores and are dispersed either violently or passively by wind, rain or animals. They are ubiquitous in soil and dung, occurring mostly as saprophytes; few are parasitic on plants and animals. Trichomycetous fungi live in the guts of arthropods. About 1000 fungal species belonging to Zygomycotina have been reported from India. Members of this fungal group are important in industry, food and in understanding the physiology, biochemistry and genetics. *Saksenea vasiformis*, a unique indigenous fungus, has found special attention in medical mycology.

#### Ascomycotina

Ascomyceteous fungi comprise a wide variety that differ in morphology, ontogeny, ascocarp details, ascus organization, nature of ascospores, ultrastructure and other characters besides occurrence in diversified habitats. Ascomycotina is the largest sub-division of the fungi encompassing 2700 genera and 28,500 species. Ascomycetous yeasts are common in moist, sugar-rich environments like plant surfaces and fruits but are also prevalent in soil, and fresh and marine water bodies. Importance of yeasts in industrial fermentation, like brewing and baking, is wellknown. The mycelial members, *Chaetomium, Xylaria*, *Neurospora, Sordaria* and *Ascobolus* are common saprophytes in soil, plant and animal remains (Figures 3 and 4). Some fungi such as *Lulworthia* and others are common



Figure 3. Ascospores of Aspergillus stellatus Curgi. 1000 ×.



Figure 4. Asci and ascospores of *Sordaria fimicola* (Rob.) Ces. & de Not. 400  $\times$ .

in estuarine environment. All such fungal forms are important in decomposition processes, because of their ability to degrade cellulose and other plant polymers. Truffels form ectomycorrhiza on forest trees and fruit bodies of fungi such as *Tuber* are a delicacy and highly prized commodity. Species of *Arthroderma* and *Nanninzzia* parasitize man and cause diseases. Species of *Ceratocystis*, *Claviceps*, *Erysiphe*, *Phyllactinia*, *Sphaerotheca*, *Taphrina*, etc. parasitize plants and cause huge losses.

Based on the published literature, it is estimated that the Ascomycetes form approximately 40 to 45% of the total fungi and this proportion is also true for Indian records.

#### Basidiomycotina

This group comprises largely of fleshy fungi which include toadstools, bracket fungi, fairy clubs, puff balls, stinkhorns, earthstars, bird's nest fungi and jelly fungi. They live as saprophytes however some are serious agents of wood decay. Some toadstools which are associated with trees form mycorrhiza, a symbiotic association<sup>30</sup> while others are severe parasites, e.g. Armillaria mellea which destroys a wide range of woody and herbaceous plants. Some fleshy fungi are notorious in being poisonous, however, a majority is harmless and some are good to eat<sup>31</sup>. Mushrooms occur in various shapes, size and colour and have attracted the attention of naturalists and are thus prized as drawings, paintings, sculptures, etc (Figure 5). In nature, mushrooms grow wild in almost all types of soils, on decaying organic matter, wooden stumps, etc. They appear in all seasons, however rains favour rapid growth when organic matter or its decomposition products are easily available. More than 2000 species of edible mushrooms are reported in the literature from different parts of the world. Singer<sup>32</sup> had reported 1320 species belonging to 129 genera under Agaricales.



**Figure 5.** *Psathryella gracilis* Fr.

#### Mushrooms and other macrofungi

Among fungi, basidiomycetes in particular have attracted considerable attention as a source of new and novel metabolites with antibiotic, antiviral, phytotoxic and cytistatic activity. About 10,000 species within the overall fungal estimates of 1.5 million belong to this group. Mushrooms alone are represented by about 41,000 species, of which approximately 850 species are recorded from India<sup>33</sup>. Besides extensive surveys of the Himalayan region that are compiled by Lakhanpal<sup>34</sup>, records from Punjab, Kerala and Western Ghats have been published during the last years<sup>35,36</sup>. What is noteworthy is the component of macrofungi that is mycorrhizal and therefore determines ecosystem dynamics of forests. For example, Lakhanpal<sup>37</sup> has recorded that in a survey conducted in the North-Western Himalayas during 1976–1987, 300 species of mushrooms and toadstools were recovered; of these, nearly 72 species in 15 fungal genera were observed to enter into mycorrhizal relationship with Abies pindrow Royle, Betula utilis D.Don, Cedrus deodara (Roxb.) Loud, Picea smithiana (Wall.) Boiss, Pinus roxburghii Sarg, Pinus wallichiana A.B. Jackson, Rhododendron arboreum Smith, Quercus incana Roxb. and Quercus semicarpifolia Smith. As many as 24 fungal species were found to be associated with Q. incana alone. Deshmukh<sup>33</sup> has compiled the folk medicine value of the Indian Basidiomycetes besides recording nearly 60 wild mushrooms, representing 54 species in 36 genera around Mumbai (18.55 N 72.54 E11 M altitude). Among the new targets used in their medicinal value are, antitumour and immunomodulatory actions of unusual polysaccharides of these macrofungi<sup>38,39</sup>.

### Rust and smut fungi

Rusts are the largest group of plant parasitic fungi in Basidiomycotina that cause severe diseases of economically important crop plants like wheat, corn, cereals, legumes, beans and grasses. They are obligate in nature except a few and produce more than one spore forms in their life cycle. More than 160 genera of rusts have been recorded, out of which 46 are monotypic comprising 7000 species, world over (Figure 6). Geographically, rusts are distributed



Figure 6. Rust spores of Marvalia ehinulata Omo. 1000 ×.

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all over the world except Antartica. The greatest number of species occur in temperate and near temperate regions. The most widely distributed species of economic significance are, *Puccinia graminis* var. *tritici* Pers., *P. recondita* Rob. Ex Desm., *Uromyces phaseoli* (Rabenh.) Wint. and *Tranzschelia discolor* (Fck.) Trans. and Litv. An important pathogen which is found around the globe is *Puccinia polysora* Underw, popularly known as Southern corn rust. More rust fungi occur on dicots than on monocot plants.

Rust fungi parasitize plants that range from ferns to orchids, and mints to composites. No rust fungus is however known to parasitize mosses or yet more primitive plants. Among the monocots, grasses support as many species as all other families combined together. Several smuts in nature also cause considerable economic loss to cultivated plants. Smut fungi are plant parasitic, mainly on angiospermic monocots. No smut fungus is known to occur in the plant family Orchidiaceae that has 1800 species. The number of recognized smut species is 1450, distributed in 77 genera and about 3500 synonyms. Host plant species numbers approximately 4100. About 800 species of smut fungi parasitize grasses alone (family Gramineae). Teliospore forming smuts (Ustilaginomycetes) are parasites on herbaceous, non-woody plants, while those lacking teliospores (Microstromatales, Exobasidiales) mostly parazitize woody plants.

#### Deuteromycotina

Deuteromycetes constitute an artificial group, which represents asexual phases of Ascomycotina and Basidiomycotina. The multiplication occurs by the production of mitotic spores or conidia from specialized hyphae called conidiophores. Conidial ontogeny forms the basis for identification and segregation of fungi imperfecti. Hughes<sup>40</sup> had visualized thallic and blastic, as the two basic developmental patterns of fungi. Louis Rene and Charles Tulsane wrote in 1811 at the end of their work 'In order to study the hidden marvels of these fungi, one must devote a great deal of labour and patience, but in gazing upon them when one discovers them, how much greater is the Joy'.

Deuteromycetes comprise 1700 genera of Hyphomycetes, and 700 genera of Coelomycetes that cover some 20,000 known species. They colonize, survive and multiply in air, litter, soil and other substrates and contribute extensively towards bio-degradation and recycling of organic matter, enzyme production, industrial production including antibiotics, immunoregulators, bio-control agents, besides causing profound mycoses, allergies and plant diseases. About 8000 Fungi Imperfecti are reported from India (Figure 7 *a–f*). These are indexed in volumes, *Fungi of India*<sup>1,41–44</sup>. A small number of important fungi reported from India fall in the category of thermophiles, with growth optima<sup>45</sup> around 45°C and others that cause disease on account of their growth on keratinaceous material including human skin<sup>45</sup>.

#### Fungal diversity and vascular plants

Fungi are known to colonize, multiply and survive in diverse habitats besides parasitizing plants as obligate parasites and biotrophs. There is substantial evidence of fungal diversity associated with a particular host plant. For example, around 40–60 fungal species are associated as endophytes with grasses<sup>95–97</sup>. While the study of tropical trees for sustenance of fungal diversity has not been carried out seriously, a single leaf, simultaneously supporting 6–10 fungal species as biotrophs and endophytes, has been recorded in *Eucalyptus*<sup>98–101</sup>. Various kinds of mycorrhizal associations are known to be formed with vascular plants, of which ectomycorrhizae (EM) and arbuscular-mycorrhizae (AM) have been studied extensively across the globe



**Figure 7** *a–f. a*, Speiropsis pedatospora Tubaki. 400 ×; *b*, Paecilomyces elegans (Corda) Mason & Hughes  $1000 \times$ ; *c*, Alternaria alternata (Fr.) Keissler. 400 ×; *d*, Curvularia eragrostidis (P. Henn.) J.A. Meyer.  $1000 \times$ ; *e*, Trichurus spiralis Hasselbring. 400 ×; *f*, Helicosporium sp.  $1000 \times$  (Anamorphic fungi).

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# Ectomycorrhizal fungi

EM fungi can contribute up to 25% or more of root biomass of forests, thus contribute effectively as a major structural component of the forest ecosystem. Diversity of macrofungi has been extensively investigated globally during the last decade or so<sup>46</sup>. But such fungal forms can help to develop management strategies of plants at community or local level, only if appropriate information with respect to species richness is known. While this information has been extensively generated for forests in North America, most studies in the Indian context<sup>34</sup> have looked at the distribution of various macrofungi without recourse to ecosystem dynamics. Recently, Pande et al.<sup>47</sup> however, have described the species diversity of epigenous EM fungi of Western Himalaya on oaks (Quercus leucotrichophora and Q. floribunda), pines (Pinus roxburghii and P. wallichiana) and deodar (Cedrus deodara). Species richness values for EM in oak and conifer forests were 43 and 55 respectively, which were close to midpoint range for similar other forests studied globally. In terms of the relative number of species, EM genera declined in the order; Amanita > Boletus > Lactarius > Hygrophorus > Cortinarius. There was clear-cut host specificity as well, with Amani-

tas primarily associated with conifers and *Boletus* and *Russula* with oaks; forests exhibiting dominance of EM hosts, however, had low tree diversity.

# Arbuscular mycorrhizal fungi

Arbuscular mycorrhizal (AM) fungi form obligate symbiotic association with many agricultural, horticultural,



Obligately mutualistic AM fungi have been studied extensively at a global scale, not only on account of their



Figure 9. Glomus macrocarpum Tulasne & Tulasne. 200 ×.



**Figure 8.** *Gigaspora margarita* Becker & Hall. 200 ×. CURRENT SCIENCE, VOL. 89, NO. 1, 10 JULY 2005



Figure 10. Sclerocystis microcarpa Iqbal & Bushra.  $400 \times ($ Arbuscular mycorrhizal fungi).

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ability to help plant withstand various kinds of abiotic and biotic stresses but also with their new found role in evolution, ecosystem dynamics, and plant community establishment. AM fungi comprise approx. 150 species, placed in Zygomycotina, order Glomales and on account of their beneficial effect on plant growth, fossil record of such fungi has helped unravel the origin of land flora. Simon et al.<sup>50</sup> have placed the origin of VAM (AM)-like fungi at 353-462 Myr ago, which concurs with the hypothesis that VAM were instrumental in the colonization of land by ancient plants. While root occupancy of nearly 85% land plants by AM fungi is known, van der Heijden et al.<sup>51</sup> proved the singular relationship among mycorrhizal fungal diversity, ecosystem variability and productivity. The data provided by these workers have reemphasized the need to protect AMF and to consider them in ecosystem management practices. On other front, Oehl et al.<sup>52</sup> examined the impact of land use intensity on the species of AMF in agroecosystems at eight sites in the, 'three-country corner' of France, Germany and Switzerland. These sites were representatives of low-input, species-rich grasslands; lowto-moderate-input farming with a 7-year crop rotation, and high-input maize monocropping; AMF spores and species numbers in the field samples declined in this order. However, some species were prevalent at all sites and were thus 'generalists' whereas those forming sporocarps were 'specialists' and appeared in grassland sites; only one species was restricted to high-input maize site. When trap cultures were raised with Plantago lanceolata, Trifolium pratense and Lolium perenne, root colonization by AMF was highest with inocula from the permanent grasslands and lowest with high-input monocropping sites; AMF spore formation followed a reverse trend, slowest with the former and fastest with the latter inocula. Increased land use intensity was therefore correlated with a decrease in AMF species richness. Considering the enhanced soil fertility of agroecosystems through organic farming, Oehl *et al.*<sup>53</sup> studied its influence on AMF diversity in a long-term (22 yr) organic trial underway in Switzerland. Field plots, carrying an 18-month-old-grassclover stand were examined in the 23rd year for AMF in both, organic and conventional plots. AMF spore abundance and species diversity was significantly higher in the organic than in the conventional systems. AMF communities differed in the two systems. For example, spores of Acaulospora and Scutellospora species were more abundant in the organic system. According to the authors, some AMF species present in the natural ecosystems are maintained under organic farming but are strongly depressed under conventional farming practices, indicating loss of ecosystem function in the latter. Studies of the central European agroecosystems have led to descriptions of not only new species of Glomus<sup>54,55</sup> but also establishment of a new genus Pacispora.

In view of their established significance in plant productivity and stress management, AMF diversity has been studied extensively in various natural and man-made ecosystems and some new forms discovered. However what is significant is the extent of diversity in a given ecosystem<sup>56</sup>.

### AM fungal diversity in India

In India, Bakshi<sup>57</sup> was the first to publish an account of 14 spore types: Glomus macrocarpum, Glomus macrocarpum Tul. and Tul. var. geosporum, G. mosseae, Glomus sp., Sclerocystis coremioides Berk. and Broome, Sclerocystis sp., Gigaspora calospora, Acaulospora sp., Endogone gigantea Nicol. and Gerd. Endogone microcarpum, Endogone 1, Endogone 2, Endogone 3. Gerdemann and Bakshi<sup>58</sup> reported two new species, viz. Glomus multicaule and Sclerocystis sinuosa. Bhattacharjee and Mukerji<sup>59</sup> described the species Glomus reticulatum from soils of Bangalore. Bhattacharjee et al.<sup>60</sup> reported the structure and hyperparasitism for Gigaspora candida while Bhattacharjee and Mukerji<sup>59</sup> described the ultrastructure of Sclerocystis coremioides sporocarp. Mukerji et al.<sup>61</sup> reported two species of Glomus, viz. Glomus multisubstensum and G. delhiense both from soils of Delhi. Till this date, 102 AM species have been reported from India.

The occurrence of AM fungi in a natural forest stand was recorded in the Old Delhi Ridge, Saraswati Range of Haryana<sup>62</sup>, forest soils and coastal regions of Andhra Pradesh<sup>63</sup>, Kodayar forest, Tamil Nadu<sup>64</sup>, forest plants of Nilgiris<sup>65</sup>, coastal tropical forest (Kodikkarai Reserve Forest) of Tamil Nadu<sup>66</sup>, Servarayan Hills of Tamil Nadu<sup>67</sup>, forest soils of Andhra Pradesh<sup>68</sup> and black pepper grown in the forest soils of Kerala<sup>69</sup>. The diversity of AM fungi has also been studied in the coastal regions of Konkan and Servaravan Hills of Tamil Nadu<sup>70</sup>, Coromandel coast of Tamil Nadu<sup>71</sup>, coastal sand dunes at Someshwara, Mangalore coast of Karnataka<sup>72</sup>, coastal sand dunes of the west coast of India<sup>73</sup> and western Ghats of Goa<sup>74</sup>.

Sengupta and Chaudhari<sup>75,76</sup> studied the occurrence of AM fungi in *Sueda maritima* (a pioneer mangrove) in terminal, seabound Gangetic delta in West Bengal. Mangrove of Muthupet estuary was surveyed by Selvaraj and Subramanian<sup>77</sup>. The occurrences of AM fungi in arid and semi-arid regions were studied in Tamil Nadu<sup>78</sup>, deserts<sup>79</sup>, arid zones of Rajasthan<sup>80</sup>, and semi-arid grasslands of Maruthamalai hills (off-shoot of the Western Ghats in Peninsular India)<sup>81</sup>.

The diversity of AM fungi in agricultural fields was reported on *Leucaena leucocephala* from Bangalore<sup>82</sup>, ornamentals and cultivated plants at Allahabad and adjoining areas<sup>83</sup>, crop fields of Konkan and Solapur<sup>84</sup>, tea plantation at Nilgiris, Tamil Nadu<sup>85</sup>, pearl millet, maize, wheat, pigeonpea and chick pea in Gwalior<sup>86</sup> and different agroclimatic regions of India<sup>87</sup>. The distribution of AM fungi in stressed ecosystems has also been reported from coal, lignite and calcite mine spoils of India<sup>64,88</sup>,

Kothagudam coal mine spoil, Andhra Pradesh<sup>89</sup>, heavy metal polluted soils of Tamil Nadu<sup>90</sup>, petro-effluentirrigated fields, soils polluted with industrial and sewage effluents<sup>91</sup>, tannery effluent polluted soils of Tamil Nadu<sup>92</sup> and stressed soils of Bailadila iron ore sites in Madhya Pradesh<sup>93</sup>.

These studies indicate that the genus *Glomus* is ubiquitous in various ecosystems in India. The distribution of other genera, i.e. *Entrophospora, Gigaspora, Sclerocystis* and *Scutellospora* is limited, indicating greater adaptability of *Glomus* to varied soil conditions.

# Tropical endophytes and the issue of fungal diversity

Fungal endophytes are microfungi that colonize living tissues of plants without producing any apparent symptoms or obvious negative effects<sup>94</sup>. Fungi that are biotrophic mutualists, benign commensals or latent pathogens are included under the broad term 'endophytes'<sup>95</sup>. Many endophytes produce unusual secondary metabolites of industrial importance<sup>2,4</sup>. Furthermore, some endophytes are known to contribute to the fitness of their hosts<sup>3–6,95–99</sup>.

Hawksworth<sup>16</sup> estimated that there are 1.5 million species of fungi; of these, only 75,000 species have been so far described. Several mycologists have tried to answer the question 'Where are the missing fungi?' by identifying the habitats that are to be studied for the presence of such fungi101,103. The internal tissues of plants harbouring endophytes may well account for a substantial number of new fungi<sup>104,105</sup>. Tropical plants are expected to support a high diversity of endophytes<sup>106</sup> and only a few of them have been screened for endophyte presence<sup>107,108</sup>. Arnold et al.<sup>109</sup>, based on the results of their study on leaves of two understorey tree species in Panama, suggested that tropical forests are hyperdiverse with reference to endophytes to such an extent that the figure of 1.5 million markedly underestimates fungal diversity. Such an argument is not untenable since the high plant diversity in the tropics is supposed to mirror endophyte diversity. However, some recent studies show that not all tropical forests are as hyperdiverse for endophytes<sup>110–112</sup>. Suryanarayanan et al.111 studied tropical forests in the Nilgiri Biosphere Reserve of the Western Ghats for endophyte assemblages based on host recurrence and spatial heterogeneity of their endophytes and concluded that the dry tropical forests had much less endophyte diversity compared to wet tropical forests.

Host specificity of endophytes concomitant with high host diversity is expected to increase the diversity of endophytes and consequently, of fungi in the tropics. However, in their study of 24 trees in the Western Ghats, Suryanarayanan *et al.*<sup>112</sup> observed neither host specificity among the endophytes nor association of distinct fungal communities with any host tree. Cannon and Simmons<sup>110</sup> obtained similar results for 12 tree species of Iwokrama forest reserve in Guyana. The low endophyte diversity in some tropical forests is attributed to the presence of dominant generalists and low frequency of occurrence of host specific forms among the endophytes<sup>111,112</sup>. Furthermore, molecular evidence shows that certain fungi, such as Phyllosticta capitalensis and Colletotrichum spp., have a very wide host and geographical range. For example, P. *capitalensis* occurs as an endophyte in South Africa<sup>113</sup>, Japan, Thailand<sup>114</sup>, India<sup>115</sup> and Brazil<sup>116</sup>, which suggests that this fungus could have been described several times as different species, especially since species name in case of the genus Phyllosticta is almost invariably based on the host from which it was isolated. This may be true for a few other coelomycete taxa<sup>117</sup>. Hence, such ubiquitous endophytes require reinvestigation using molecular techniques to avoid an exaggerated value of fungal diversity. Molecular studies on Colletotrichum endophytes isolated from trees of Guyana have also confirmed that at least in some cases fungal diversity may be inversely related to host diversity<sup>118</sup>.

Recent studies have demonstrated that fungal endophytes are neither passive residents<sup>7</sup> nor a mere assemblage of latent pathogens of their hosts<sup>119</sup>. They possibly represent a storehouse of new species of fungi, especially in the tropics<sup>120,121</sup>. Since, only a few plant hosts and habitats have been studied for endophytes, the importance of such studies cannot be overstressed<sup>108,122</sup>.

#### Zoosporic and conidial aquatic fungi

The zoosporic fungi predominant in moist soils have been surveyed extensively in the country<sup>123,124</sup>. Watermolds contributes towards the energy flow and productivity of aquatic and semi-aquatic systems through degradation of plant matter. On account of its climatic variability, the Kumaun region of the Central Himalaya exhibits significant diversity of zoosporic fungi, popularly termed water molds. Sati<sup>125</sup> has reported these fungi from diverse habitats, viz. water, agricultural fields, diseased root seedlings and diseased fish and their eggs. According to Sati, as many as 80 species of zoosporic fungi parasitic on cold water fish, belonging to families Blastocladiaceae, Pythiaceae and Saprolegniaceae were found in the Kumaun region. Another interesting group of aquatic fungi that occurs on air-water interface, the aquatic hyphomycetes completes their life cycle on submerged substrates, in foaming springs and posseses unique conidial morphology. The conidial aquatic fungi are involved in the first stage of leaf decomposition in freshwater streams<sup>126</sup>. Many workers have undertaken extensive surveys of such fungi in South India and near the line Kumaun Himalaya and have recovered 60 species from various habitats<sup>6,21,127,128</sup>. In a recent survey, Sati et al.<sup>129</sup> collected submerged leaves and water foam from streams located in the western part of the Central

Himalaya (20°22"–29"N lat. and 79°28"–30"E long.) around Jeolikot (1850 m asl), Khurpatal (1600 m asl), Niglat (1650 m asl), Gufa Mahadeu (1850 m asl) and Snow view (2250 m asl) and recorded 14 species of conidial aquatic fungi representing 10 anamorphic genera, all new to Indian mycoflora. These were, *Alatospora, Anguillospora, Dimorphospora, Dwayaangam, Flabellocladia, Leonniera, Mag-dalaenaea, Tetraladium, Tricladiopsis* and *Trinacrium*.

## Communities of fungi and ecology

Fungi are known to play a vital role as decomposers, symbionts of plants and animals and as parasites of plants in different ecosystems. Fungi interact with their hosts, and also with abiotic variables in the environment. They occur on rocks, in soil, in sea and freshwater, in extreme habitats, experiencing high and low temperature, on dry substrates and in concentrated nutrients. Members of mucorales are considered as ruderals since they survive in soil as long as nutrients are available although they are not capable of degrading cellulose or lignin. Fungi like Fusarium, Gliocladium, Penicillium and Trichoderma are stress tolerant. Majority of fungi are mesophiles with maximum growth between 25 and 30°C (Mucor mucedo, Mortierella, Penicillium chrysogenium) however Cylindrocarpon sp., Candida scotti are cold tolerant (psychrotolerant) and can grow near 0°C; others are thermotolerant and grow above 40°C (Rhizomucor, Thermomyces, Talaromyces). Xerotolerant fungi can grow on dry material (Aspergillus, Pencillium) with low matric potential (a<sub>w</sub>) while osmotolerants grow at very low osmotic potential (Pichia sp.). Dung of herbivorous mammals harbours a large number of fungi, termed coprophiles, of which Pilobolous, Ascobolus and Basidiobolus are famous for their special shot-gun dispersal mechanism!

An interesting ecological group of fungi captures and grows parasitically on nematodes, their cysts and eggs (nematophagous); over 150 species are reported<sup>130</sup> within the genera *Catenaria*, *Dactyella*, *Harposporium*, *Monacrosporium*, *Nematophthora*, *Rhopalomyces* and *Stylopage*. Such fungi are good trapping agents in the biological control of nematodes.

# **Keratinophilic fungi**

A specialized group of fungi that colonizes keratinaceous substrates such as human hair, skin, nails, feathers, hooves and horns have been studied extensively in India since several of these are cause of human and animal diseases. Soil is the reservoir of such forms that are distributed in genera *Acremonium*, *Arthroderma*, *Chrysosporium*, *Epidermophyton*, *Malbranchea*, *Microsporum*, *Myceliophthora* and *Trichophyton*<sup>131</sup>. The relative distribution of keratinophiles depends on the frequency of animals visiting a particular habitat but for species such

as *Chrysosporium tropicum* it could be considerably high. The other major habitat of keratinophilic fungi is, birds feathers and nests. Major developments concerning their occurrence and activities are compiled by Kushwaha and Guarro<sup>132</sup>.

Preponderence of keratinophilic fungi that are responsible for a variety of fungal infections (dermatomycoses) can be gauged by the fact that in a recent study of soils from agricultural fields, garden, forests and zoo, Kushwaha and Gupta<sup>131</sup> recorded 22 species of *Acremonium*, *Chrysosporium*, *Malbranchea*, *Microsporum*, *Myceliophthora* and *Trichophyton*; of these 14 species belonged to the genus *Chrysosporium*.

In a study of their distribution on feathers of Indian birds (chicken, pigeon, house sparrow, parrot, crow), Kushwaha and Gupta<sup>131</sup> recovered 30 keratinophilic fungal isolates that belonged to *Chrysosporium*, *Microsporum*, *Myceliophthora* and *Trichophyton*; 23 isolates belonged to the single genus *Chrysosporium* indicating its wider distribution and relative ability to colonize the keratinaceous substrates in nature. What is of greater significance is the occurrence of dermatophytes over keratinophiles since the former cause skin infections, viz. *M. gypseum* and *T. mentagrophytes*. This suggests that the distribution of keratinophilic fungi in birds depends upon their body temperature, feather fat and the prevailing environmental conditions.

# Fungal diversity and community dynamics in mushroom compost ecosystem

Mushroom compost, a complex man-made ecosystem, harbours a complete spectrum of microbial diversity mesophilic and thermophilic. From a purely microbial ecology point of view, this controlled ecosystem is indeed unique since conditions under which the crop is grown and relatively short time required to complete the successional cycles, is not matched elsewhere. Microbial community succession occurs at a very fast pace and changes with the temperature gradient of two phases. Mushroom compost fungi constitute a dominant component with respect to species richness, distribution and abundance. Straatsma *et al.*<sup>133</sup> reported a biomass ratio of 1.0:1.8 of bacteria to fungi after phase II while according to Weigant<sup>134</sup> the ratio was 1.0:0.9 in conventional compost and 1.0:2.3 in the experimental compost. In a recent study of mushroom composting, Rawat<sup>135</sup> found that the mesophilic fungal counts (log<sub>10</sub> CFU) in mushroom compost varied from 4.83 to 5.01 (zero day to drench) and that of thermophilic fungal counts, from 4.62 to 4.47. Maximum structural divergence and species richness amongst mesophilic fungi was high in zero day morphotypes (H' =1.55; RI = 2.54) and the least in peak-heat morphotypes (H' = 0.68; R1 = 0.64). For thermophilic fungi, fourth turning of phase I compost exhibited maximal diversity (H' = 2.14) and peak heat stage morphotypes (H' = 0.75),

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the least diversity. Aspergillus spp., Rhizopus oryzae, Trichoderma viridae, Chaetomium sp., Penicillium sp. and Alternaria sp. were observed as dominant mycoflora in the initial stages of composting. Acremonium sp. and Paecilomyces variotii were the additional mesophilic fungi in the later period of composting<sup>136</sup>. The recolonization of mesophilic mycoflora in the compost can occur at the time of spawning due to fall in temperature and if free cellulose and carbohydrates are still present in compost<sup>137</sup>. During the course of composting and at spawning stage most mesophilic fungi exert a deleterious effect on the growth of A. bisporus mycelium, with ultimate reduction in yield<sup>117</sup>.

The mesophilic microflora forms the pioneer community while thermophiles form the climax community. Thermophilic fungi grow extensively during the last phase of composting (phase II) from the spores that survive the pasteurization temperature<sup>138</sup>. Their presence throughout the course of composting is largely responsible for maintenance of biological equilibrium that ultimately leads to unique selectivity wherein A. bisporus multiplies without competition. Composts harbour up to 10<sup>6</sup> propagules of thermophiles<sup>139</sup> and these are therefore, primarily considered as compost fungi. At the end of the composting process, about 50-70% of the compost biomass is constituted by thermophilic fungi<sup>140</sup>. While most species are eliminated, S. thermophilum appears as near exclusive species after phase II composting and constitutes a climax species in the mushroom compost along with thermophilic actinomycetes<sup>133</sup>. The number of CFU of S. thermophilum in fresh matter of phase II is about 10<sup>6</sup> g<sup>-1</sup> compost<sup>141</sup>, however, actinomycetes and bacteria appear to play a decisive role in successful colonization by this thermophile.

Dominance of S. thermophilum has been reported by several workers<sup>133,134,138,142</sup> while *H. grisea* var. thermoidea and *H. insolens* have been described by others<sup>143</sup>. They are inherently close partners in the degradation processes in compost and provide selectivity to compost<sup>138</sup>. The RAPD analysis and sequence analysis of ITS region of rDNA show wide genetic variation in Torula-Humicola complex<sup>144</sup>. RAPD analysis of 34 geographically diverse isolates revealed two distinct groups showing differences in the banding pattern. An examination of the genetic distance matrix indicated differences between isolates from Scytalidium thermophilum cultural types 1 and 2. The sequence analysis of ITS1, 5.8S and ITS2 region of rDNA suggests high homology between the isolates with minor sequence variation. This could be correlated with differences between the isolates based on morphology and thermogravimetric characteristics<sup>145,146</sup>. The genomic DNA variations thus facilitate the differentiation of subgroup within the species. Study of in situ genetic diversity of fungi of mushroom compost by Rawat<sup>135</sup>, using SSCP as tool, has brought into focus presence of 95 distinct bands although only 34 fungal species were recovered.

Considering the fact that culturable microbial populations are limited on account of our poor understanding of their nutritional requirements, detailed *in situ* enzymatic investigations are likely to provide a better understanding of the relationship between structural and functional diversity of thermophilic fungal community. Iiyama *et al.*<sup>147</sup> observed that the loss of cellulose and lignocellulose and the increase in protein content during the composting period were a result of increased polysaccharolytic activity of the fungal biomass.

#### **Fungal conservation**

Threats to fungi throughout the globe is of concern since they are not only beautiful but also play a significant role in human welfare. Moore et al.<sup>148</sup> have suggested the following steps for fungal conservation: (i) Conservation of habitats, (ii) In-situ conservation of non-mycological reserves/ecological niches, and (iii) Ex situ conservation especially for saprotrophic species growing in culture. Fungi are very seldom legally protected however in Slovakia, 52 species have a special legal status, enabling managers to prevent damage to their habitat<sup>149</sup>. In the absence of legal protection, some effort needs to be made to have code of practice or suggestive documents stressing the importance of fungal conservation, a practice adopted in UK and Switzerland. One of the tools that would help in conservation is, inventorization. In most countries checklists of fungi are not available however such projects are now operative under the umbrella of IUCN.

To help culture collections centres maintain appropriate standards, the World Federation for Culture Collections (WFCC) has formulated guidelines which outline the necessary requirement<sup>150</sup>. The first service culture collection was that of Frantisek Kral in German Technical University in Prague that was established<sup>151</sup> in 1890. World data center<sup>152</sup> now has 350 registered culture collection centers in its database. The selection of preservation technique for fungi not only depends upon the success of the method but also upon the use of the organism, time, facilities and resources available. Long-term stability is considered together with the required availability of the culture without delay. A collector may select a continuous growth method which is to be backed up with one that reduces the possibility of change during storage. For example, growth techniques allow strain drift. Use of synthetic medium places selective pressure on the organism, allowing variants to dominate. Mineral oil storage is a simple method of storage that retains viability of fungi for many years but places strains under selective pressure because of the special conditions of storage. Water storage technique may allow growth depending upon the method adopted. The procedure is to cut agar plugs from the edges of actively growing cultures and placing them in sterile distilled water in screw cap bottles. The nutrients available in the agar will allow growth until oxygen is depleted in the storage container. Soil storage involves inoculation of spores or mycelium suspended in sterile distilled water into sterile soil of approximately 20% moisture content. This method of storage can retain viability for 10 to 20 years. Silica gel storage methods are suitable methods for fungal spores that remain viable for periods up to and over 20 years. Freeze drying entails freezing of the organism and its desiccation by the sublimation of ice under reduced pressure. Cryopreservation is the method of storage at ultra low temperatures, which is the most successful method for retention of both the viability and characteristics of fungi.

# Prospecting fungi and exploitation of fungal diversity

Fungi are known to colonize, multiply and survive in diversified habitats, viz. water, soil, air, litter, dung, foam, etc. Fungi are ubiquitous and cosmopolitan in distribution covering tropics to poles and mountain tops to the deep oceans. The kingdom of fungi contains 1.5 million fungal species, of which 74,000 species are named<sup>2</sup>. Many of the described species are known only as dead herbarium material and around 5% of species are isolated as pure cultures. Geographic location, climatic conditions, micro-habitat, substrate type, distribution of fauna and flora are all important factors contributing to fungal distribution around the world. Fungal flora of the United Kingdom, Korea, Cuba and other countries has been well explored for fungal species. Unlike prokaryotes, automated isolation techniques for fungi are not yet possible because of the extreme diversity in fungal mycelium, growth and texture. Studies of fungal distribution and mapping are challenging tasks due to lack of sufficient taxonomic knowledge and lack of mycologists around the world.

Nature represents a formidable pool of bioactive compounds and is more than ever a strategic source for new and successful commercial products. Recent advances made in genomics, proteomics and combinatorial chemistry show that nature maintains compounds that have already the essence of bioactivity or function within the host and in the environment. Microbial sources such as fungi are well recognized to produce a wide variety of chemical structures, several of which are most valuable pharmaceuticals, agrochemicals and industrial products. The world of fungi provides a fascinating and almost endless source of biological diversity, which is a rich source for exploitation.

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