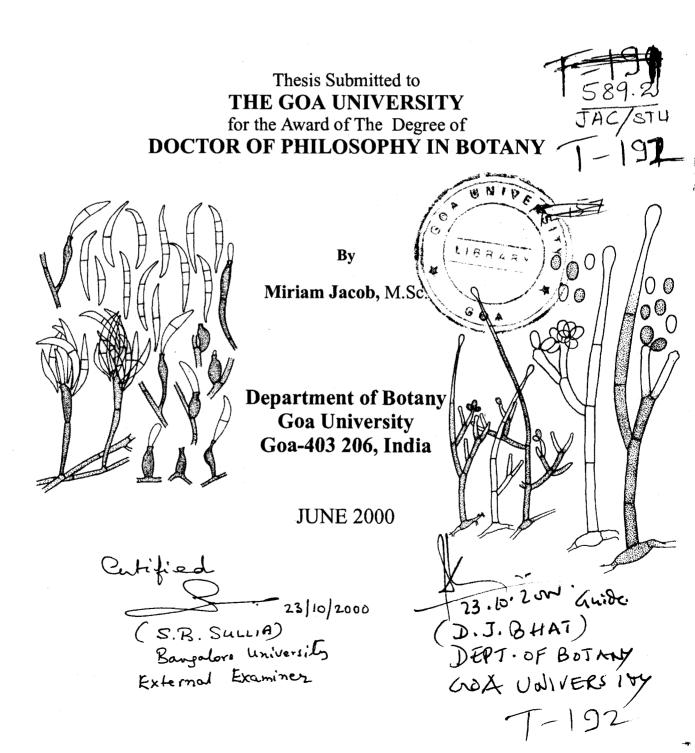
STUDIES ON DIVERSITY, ECOLOGY AND ACTIVITY OF THE MICROFUNGI ASSOCIATED WITH FICUS BENGHALENSIS Linn. AND CARISSA CONGESTA Wight FROM GOA STATE.



CERTIFICATE

I certify that the thesis entitled STUDIES ON DIVERSITY, ECOLOGY AND ACTIVITY OF THE MICROFUNGI ASSOCIATED WITH FICUS BENGHALENSIS Linn. AND CARISSA CONGESTA Wight FROM GOA STATE submitted by Ms. Miriam Jacob, is a record of research work done by her during the period from 1997-2000 when she worked under my supervision. The thesis has not formed the basis for the award of any degree, diploma, associateship or fellowship to Ms. Miriam Jacob.

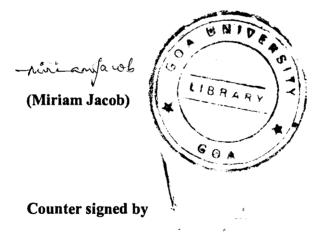
I affirm that the thesis submitted by Ms. Miriam Jacob incorporates the independent research work carried out by her under my supervision.

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DECLARATION

I hereby declare that the Ph.D. thesis entitled "Studies on diversity, ecology and activity of the microfungi associated with Ficus benghalensis Linn. and Carissa congesta Wight from Goa State"submitted to Goa University, forms an independent work carried out by me in the Department of Botany, Goa University, under the supervision of Dr. D. J. Bhat, Professor and Head, Department of Botany, Goa University and the thesis has not formed previously the basis for the award of any degree, diploma, associateship or other similar titles.



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CHAPTER I INTRODUCTION

Introduction:

About 18 million kinds of living organisms are known to exist on the earth's surface, of which fungi have been estimated to constitute 1.5 Million species (Hawksworth, 1991; Janzen and Hallwachs, 1994). Being heterotrophs, the fungi play several distinctive roles in the functioning of ecosystems: as saprophytes they subsist on dead and decaying organic remains, as parasites they exploit living plants and animals and as symbionts they cooperate and adjust with many phototrophic organisms (Kendrick, 1992). The saprophytic fungi are very versatile organisms and exist in a wide range of habitats; for example, as aquatics in fresh water and sea, as terrestrials in soil, air and plant litter, as coprophilous on dung of herbivores, as entomogenous on living insects, as endophytes in living plant and animals and so on. It is well known that with their amazingly diverse species composition, ability to produce a variety of extra-cellular enzymes and breaking down of organic substances and from simple to complex ecological association with plants and other organisms, the fungi act as regulators of the structure, function and dynamics of plant and animal community in nature (Dix and Webster, 1995).



Diversity of fungi:

An extensive and elaborate record of studies carried out on saprophytic, parasitic and mutualistic microfungi associated with plants is available. Such studies have resulted in the making of several monographic treatments of fungi (Ellis, 1971, 1976; Ingold, 1975; Lundqvist, 1972; Matsushima, 1971, 1975; Sivenesan, 1984; Subramanian, 1971, 1987; Sutton, 1980). A few studies were directed at measuring the abundance and diversity of microfungi that inhabit the plant litter (Heredia, 1993; Wicklow and Carroll, 1981). Several investigations were carried out to elucidate the

process of decomposition of plant litter in a variety of habitats (Barlocher, 1992; Dickinson and Pugh, 1974).

Recent studies have revealed that plant litter and habitats in the tropics harbour diverse microfungi in abundance (Hyde, 1997; Bills and Polishook, 1994). Being in the tropical belt, the natural substrates and habitats in our country are said to accommodate a very rich fungal gene pool. Relative to the understanding of the extent of biotic diversity, ecology, biogeography and biochemical functioning of terrestrial plants and animals, knowledge on the microfungi however remained underdeveloped.

Ecology of Litter fungi:

Studies on the succession of fungi on leaf-litter of angiosperms conifers and lower plants have been carried out (Dix and Webster, 1995). The pattern of development of the fungus flora on conifer needles and angiosperm leaf-litter is that the leaf-inhabiting phylloplane fungi persist on the needles/leaves for several months after, needle/leaf fall and later produce their sexual stages. A small group of leaf-inhabiting fungi which appear first on the living leaves in low numbers become more abundant once the leaves reach the ground. The decaying leaves are further invaded by true litter-inhabiting fungi which facilitate complete decomposition of the leaves. The decaying leaves are also colonized by typical soil-inhabiting fungi and these become more dominant as time passes and when the decomposed litter gets embedded in the deeper layers of the soil. It is not known, however, besides the epiphytes and endophytes of the living leaves, which other components in the vicinity of the plants contribute for the litter mycoflora.

Studies have been carried out on the fungi associated with decomposing plant litter in India. Yadav (1966) examined the frequency of occurrence of fungi on

decaying stems of *Heracleum sphondylium*. Vittal (1973) undertook qualitative and quantitative analyses of the fungi associated with *Atlantia monophylla* and *Gymnosporia emarginata*. Rai (1974) studied the succession of fungi on tropical grasses. Sudha (1978) examined the mycoflora associated with the leaf litter of *Ixora parviflora* and *Glycosmis cochinchinensis* and Dorai (1987) carried out investigations on the fungi associated with 13 species of *Eucalyptus*.

The qualitative and quantitative analyses of the structure of fungal communities have been worked out using theoretical models (Shearer and Webster, 1985). Aoki and Tokumasu (1995) attempted to analyze the dominance and diversity of fungal communities on fir needles using mathematical models and explained the community structure of microfungi.

Endophytic fungi:

Though initial researches on endophytic fungi were mainly explorative and taxonomic in nature, recent studies were aimed at search for novel and bioactive compounds (Bills, 1995b). These efforts have resulted in the documentation of endophytes from a large number of phanerograms, ferns, lichens and mosses (Petrini 1986, 1991). Endophytic fungi have now been reported from hosts as diverse in habitat and taxonomy as those from the tropics, the mangroves, the freshwater macrophytes, the desert plants, the arctic mosses and the angiosperms and conifers of temperate regions (Redlin and Carris, 1995). Endophytic fungi have been recovered and analyzed from aerial plant parts such as leaves, stem, bark, fruit, flower and seed and underground roots (Bills, 1995b).

Importance of fungi:

The fungi are considered important in the degradation and subsequent conversion of relatively indigestible cell wall substances of vascular plant tissues into carbohydrates and protein sources which are utilized by consumers in the food web. It is well known that the fungi, with their ability to secrete a variety of extracellular enzymes, play an important role in the decomposition process (Dix and Webster, 1995). Various enzymes such as amylase, cellulase, protease, pectinase, ligninase, is also a lightnase laccase and xylanases play significant part in the degradation of cellulose and other polysaccharides.

It is now becoming clearer that microfungi of the tropics are potential sources of biotechnologically significant and pharmaceutically valuable organic molecules (Bills, 1995a; Cuomo et al., 1995). Besides, the fungi are recognised as economically and ecologically sustainable biocontrol agents of harmful insects and pests, mycorrhizal biofertilizers and nutritionally rich food additives (Kendrick, 1992). Realizing these exciting revelations on the diversity, creativity and associations of fungi, Hawksworth (1991) and Rossman (1994) advocated for urgent and comprehensive inventory, recovery and investigations of the mycobiota of the tropical belt, since this region of the world is presently under serious ecological stress.

The Present Work:

Many gaps exist in our knowledge on the possible source and extent of fungal diversity and density on aerial leaves and fallen plant litter, ecological relation between the leaf endophytes and fungal gene pool in the litter and the possible activities of the mycota in this environment. In order to understand these complex interactions, the present investigation was carried out during the last two years with an

aim to elucidate the diversity, ecological association and activity of the fungi associated with two plant species commonly found growing in the State of Goa, *Ficus benghalensis* Linn. (F: Moraceae) and *Carissa congesta* Wight (F. Apocynaceae) and their environment. The results of the study are presented in three parts and compiled in the form of a thesis.

The Chapters II and III of the thesis cover the review of relevant literature and materials and methods used in the study. The findings of the investigation are presented and discussed in Chapter IV and summarized in Chapter V. An exhaustive bibliography is given at the end of the thesis.

It is believed that studies such as this, where the diversity, ecology and activity of the fungi associated with evergreen or deciduous plants elucidated over a period of time, are likely to throw some light on the role of saprophytic, endophytic and other fungi after senescent leaves reach the ground as leaf.

CHAPTER II REVIEW OF LITERATURE

The surface of plant leaves and stems generally serve as deposition sites of airborne microorganisms, pollen grains and inert particles (Barnes, 1968; Fokkema, 1971a, 1971b; Warren, 1972; Chou and Preece, 1968). Throughout the life of plants, fungal spores are continuously deposited on their surfaces by wind impaction, sedimentation and rain wash-out from the atmosphere and splash-dispersal (Dickinson, 1976; Dix and Webster, 1995).

The Phylloplane:

The first systematic study of the phylloplane biology was by Potter (1910). The term phyllosphere, meaning the external surface of the leaf, was first put to use by Last (1955). The term phylloplane was distinguished by Leben (1965) who also recognized two types of phylloplane microflora, the casuals and the residents. The casuals, consisted of organisms that are firmly lodged on the surface of the leaf but not in a position to germinate on or colonise the plant surface. The residents were those that are more acclimatized to the phylloplane where they thrive as saprophytes. The inability of the casuals to grow on the leaf surface was attributed to factors such as, surface texture, lack of essential nutrients, host specificity and competition between the resident organisms (Barnes, 1969; Ruinen, 1966; Last and Deighton, 1965).

Dickinson (1976) classified the phylloplane fungi into three categories: non-pathogenic, pathogenic and exochthonous. The non-pathogenic fungi consisted of those able to grow and sporulate in favourable and unfavourable conditions but triggered to grow only at the onset of senescence. The pathogenic fungi are wholly or partially restricted to the phylloplane and could survive long periods on the phylloplane prior to penetration. The phylloplane forms an essential link in the life

cycle of exochthonous fungi though the fungi do not derive any advantage from the habitat.

The nature of phylloplane mycoflora:

The leaf surface forms a host to diverse microbial population which mainly includes fungi and bacteria. The most abundant of the fungi on the surfaces of leaves are yeasts which included members of the Ascomycotina, Basidiomycotina and the Fungi Imperfecti (Last and Deighton, 1965). The genera such as *Candida*, *Cryptococcus*, *Rhodotorula*, *Sporobolomyces*, *Tilletiopsis* and *Torulopsis* were regularly encountered on the surface of the leaves (Dickinson, 1986).

The leaf surface has been looked upon as being home to a large number of filamentous fungi. Taxa belonging to filamentous Ascomycetes, the Zygomycetes, Basidiomycetes and Deuteromycetes have been recorded on the phylloplane of different plants (Dickinson, 1976). Amongst the filamentous fungi recorded on aerial plant surfaces, the sooty molds stand out as producing spectacular colonies. Epiphytic fungi such as species of *Erysiphe* which grow extensively on leaves and other surfaces develop physiological connections with the underlying host tissues. The biology of these pathogens is of interest to microbial ecologists as they influence the aerial plant surface ecosystems (Dix and Webster, 1995).

Several studies have been carried out on the phylloplane mycoflora. Hog and Hudson (1966) described the succession of fungi on leaves of *Fagus sylvatica*. Hog (1966) elucidated the factors determining the natural succession of fungi on beech leaves. Studies on the phylloplane mycoflora so far made included the plants such as *Halimone portulacoides* (Dickinson, 1965), *Pisum sativum* (Dickinson, 1967), *Cassia tora* (Mishra and Tewari, 1969), *Echinocloa crusgalli* (Mishra and Srivastava, 1971),

Northofagus truncata (Ruscoe, 1971), Typha latifolia (Pugh and Mulder, 1971), Triticum aestivum (Mishra and Srivastava, 1974), tomato (Mishra and Kanaujia, 1974), rye (Fokkema et al., 1975), barley (Dickinson and Skidmore, 1976), Brassica oleracea (Gingell et al., 1976), Hippophae rhamnoides (Lindsey and Pugh, 1976a,b), Hordeum vulgare (Mishra and Tewari, 1976), maize (Warren, 1976), Panicum coloratum (Eicker, 1976), Picea abies (Collins and Hayes, 1976), potato (Kumar and Gupta, 1976), poplar and plum (McKenzie and Hudson, 1976), larch (McBride and Hayes, 1977), Quercus robur (Cox and Hall, 1978); Populus tremuloides (Wildman and Parkinson, 1979), Ilex aquifolium (Mishra and Dickinson, 1981), Eucalyptus vaminalis (Cabral, 1985), muskmelon (Singh, 1995), Shorea robusta (Baruah and Bora, 1995), Citrus (Kalita et al., 1996), Quecus robur (Newsham et al., 1997) and Myristica fatua var. magnifica and M. malabarica (Bhat and Kaveriappa, 1999).

Two broad kinds of techniques have been employed to study the microfungi on leaf surfaces. Direct techniques include the impression films and surface stripping (Beech and Davenport, 1971; Lindsey and Pugh, 1976a), staining (Schimdt, 1973; Warren, 1972a), leaf clearing (McBride and Hayes,1977), scanning electron microscopy (Lindsey and Pugh, 1976b), infra-red photography (Purnell and Farell, 1969) and autoradiography (Waid et al., 1973). Indirect or cultural techniques include use of selective media (Beech and Davenport, 1971), impression plates (Apinis et al., 1972), thin agar film (Parkinson et al., 1971), dilution plate and leaf washing (Warren, 1976; Davenport, 1976; McBride and Hayes, 1977), spore-fall method (Lindsey and Pugh, 1976b), incubation in humidity chamber (Lindsey and Pugh, 1976b) and measurement of fungal products (Frankland et al., 1978). The methods employed to study the colonization of aerial organs of plants have been reviewed by Lindsey (1976), Beech and Davenport (1971) and Macauley and Waid (1981). The pros and

cons of the direct and indirect techniques are enumerated by Dix and Webster (1995) and they concluded that for gathering comprehensive data on the fungal colonization of plant tissues, different investigative techniques must be employed.

Last and Deighton (1965) pointed out that the season, age of the plant and nutritional status of leaf, control the nature of phylloplane mycoflora. Dickinson (1976) attributed the presence of phylloplane fungi to the availability of fungal inocula, nature of plant surface, factors such as temperature, rain, dew, humidity, wind, physiology and health status of the plant and the nature of the plant community. Macauley and Waid (1981) listed out the factors such as nutrients, the ability of the organism to survive in an exposed environment depending on their resistance to the extremes such as starvation, drought, high and low temperatures, UV radiation, presence of fungicides, the grazing population, mycolytic organisms living in association with the phylloplane fungi and the response of the host plant to the presence of fungi or the production of fungal metabolites are responsible for colonization of fungi on leaf surface. Dix and Webster (1995) attributed the factors such as cell leakage, competition, the pollen effect, interspecific competition, plant inhibitors and climatic factors as influencing the growth of microorganisms on plant surfaces.

Role of fungi in the decomposition of plant litter:

In terrestrial ecosystems, much of the energy fixed by photosynthesis finds its way to the soil in the form of dead organic matter which is decomposed by a host of microorganisms. The rates of breakdown of forest litter influences the nutrient uptake and the standing state of nutrients in the forest floor. Because of its role in nutrient

cycling and in supporting the saprophagic component of the ecosystem, the process of decomposition has received growing attention in recent years.

Decomposition is the process of separation of any substrate into its constituent elements. This would signify the mechanical disintegration of dead plant structure from the stage where it is still attached to the living plant, to the humus stage where the gross cell structure is no longer recognisable. Decomposition is essential for recycling the forest canopy and in determining the plant and animal communities that thrive on the forest floor (Dix and Webster, 1995).

Work done at Pine Lake Preserve, had some interesting observations on forest litter decomposition. The study showed that, the speed of decomposition of litter varied from forest to forest. It was slowest in the hemlock, fastest in the mixed hardwood, and proceeded at an intermediate rate in the beech and red pine forests. Tiny bags of litter were prepared from each forest and placed in each of the other forests and how fast the litter decomposed was measured. Hemlock litter didn't decompose faster than in the mixed hardwood forest. In fact the guest litter, no matter which forest it was from, decomposed pretty the way it would have at home. Although there was some interaction between the litter and the type of forest it was in, the decomposition rate seemed to depend on the litter itself.

In conclusion, forest litter is the basis for an elaborate detritus food web. Bacteria and fungi feed on the litter, and they, in turn, are eaten by small invertebrates such as springtails, mites, and nematodes. These are devoured by larger invertebrates, namely the earthworms, euchytraeid worms and insects, which in turn are eaten by vertebrates like the red-backed salamander, the top carnivore of the detritus food web in the temperate forest. It was also found that the decomposition process seemed to be sensitive to acidic conditions. More the acidic the litter and forest soil, the slower the

litter decomposed. The hemlock forest was the most acidic of the four forests on the Preserve, the hardwood forest the least. The study also showed that the decomposer bacteria were more sensitive to acidic conditions than the fungi

The role of the fungi in the decomposition process is well studied and documented (Hayes, 1965; Hering, 1965; Hudson and Webster, 1958; Hogg and Hudson, 1966; Kendrick and Burges, 1962; Macauley and Thrower, 1966; Minderman and Daniels, 1967). Fungal floristics has been the object of many of the studies, though in some there were attempts to relate the occurrence and succession of fungi to changes in the nutritional status of the leaf (Hering, 1967; Hudson, 1971). The ecology of fungi colonizing senescent and fallen leaves has also been the subject of some of these investigations (Hudson, 1971).

The layer of dead plant material not attached to a living plant and may be present on the soil surface is generally considered as litter. The making of litter however commences with senescence of leaves. Abscission of a leaf base follows the senescence when much of the mineral content is withdrawn to the stem and the phylloplane fungi already commenced the decomposition of available carbohydrates.

On young green leaf, yeasts and yeast like imperfect fungi such as *Auriobasidium pullulans* and *Cladosporium* sp. are prominent (Dickinson, 1973, 1976; Godfrey, 1974; Leben, 1965; Ruinen, 1963; Dickinson and Wallace, 1976; Last and Warren, 1972). The presence of filamentous fungi appeared to be less frequent on young green leaves than on older ones (Dickinson, 1976). About 100 genera of phylloplane fungi have been reported so far on over 35 different higher plants studied. Most species were known to occur infrequently. As the leaf matured hyphal development increased rapidly (Dickinson, 1976; Ruscoe, 1971; Pugh and Mulder, 1971; McBride and Hayes, 1977) to a point where at abscission, the leaf was

extensively colonized. These species were the primary colonizers of the dead tissue of the leaf (Hudson, 1968; Dickinson, 1976). Last and Deighton (1965) found out that the phylloplane fungi frequent the leaves more often than in the soil suggesting that they are well adapted to the micro-environment of the leaf and possession of pigments in sooty moulds is to survive the high light intensity at the leaf surface. It has been observed that the mycoflora changed as senescence occurred and to a certain extent mycoflora affected the rate of senescence (Dickinson and Wallace, 1976). At the stage of abscission, primary saprophytic species belonging to the genera such as *Ascochyta*, *Leptosphaeria*, *Pleospora* and *Phoma* along with other parasitic fungi which may or may not be host specific, inhabit the moribund leaf (Dickinson, 1976). After abscission, the role of these fungi changed to bring about the breakdown of organic matter and to prevent the accumulation of toxic substances to levels harmful to primary colonizers and leading to mineralization of essential elements out of the organic debris in order to maintain fertility and the productivity of the ecosystem (Witkamp, 1973).

Cabral (1985) made a detailed study of the phylloplane mycoflora of Eucalyptus viminalis. He recognised two groups of fungi, phylloplane or epiphyllic species and endophytes or endophyllic species. Phylloplane fungi colonised the interior of the leaf only occasionally and did not displace the endophytes. He also observed that ascomycetes and coelomycetes were better represented as endophytes than hyphomycetes. Alternaria alternata, Cladosporium cladosporioides, Epicoccum nigrum and Microsphaeropsis callista were phylloplane fungi isolated in high frequency, while Coccomyces maritiniae, Coniothyrium sp., Macrophoma smilacina and Zolleneria eucalypti were the common endophytes. A distinct seasonal pattern was observed for the phylloplane fungi wherein the maximum number was seen in

autumn-winter and minimum in summer in proportion to humidity and temperature. The endophytes were appeared to rely more on the age/or physiological conditions of the leaf. He also made an attempt to ecologically classify the phylloplane fungi into the ruderals, residents and primary saprophytes. Ruderals occurred sporadically in low frequencies and as inactive propagules. Residents were those that were more persistent and appeared in high frequencies. Primary saprophytes were those that disappeared before the leaf died. Residents were further subdivided into, 'specific' that did not actively indulge in the degradation of the substrate when the leaf died and 'unspecific' that participated actively in the primary degradation and did not diminish when the leaf ultimately died.

Several studies have dealt with the decomposition of plant matrices in different ecosystems and the changes in fungal saprophytic communities in the litter layers in time (Bills and Polishook, 1994; Chasseur and Beguin, 1990; Kjoller and Struwe, 1990; Sieber-Canavesi and Sieber, 1993; Aoki, *et al.*, 1990, 1995; Tokumasu *et al.*, 1994). Some studies were concerned with the composition of and seasonal variation in fungal species colonising the leaf litter of single plant species (Vardavakis, 1988; Marakis and Diamantoglou,1990; Mulas *et al.*, 1990,1995) whereas others were with mixed litter (Lunghini, 1993, 1994; Zucconi *et al.*, 1997).

Time-related changes in community structure are the so-called fungal successions (Dix and Webster, 1995). Many of the factors which influence successional changes have been identified and the sequence of events involved is now fairly understood. Colonisation of a dead organism leads to the immediate struggle amongst potential saprophytic colonists for establishment, what is called as the 'prior colonisation effect' (Barton, 1960, 1961; Bruehl and Lai, 1966). Weak parasites like *Pythium* and *Fusarium* species and harmless or mutualistic non-obligate endophytes

usually form prominent members of the pioneer communities, with their ability to germinate rapidly and grow fast. The reasons for the loss of pioneer colonisers as the community matures during succession is no longer attributed to nutritional hypothesis, wherein pioneer colonisers dependant upon simple organic sources, disappeared from communities when supplies of these became exhausted. The development of antagonistic phenomenon or the accumulation of staling and antibiotic toxins in the substratum could stop the growth of the coloniser.

Dix and Webster (1995) observed that the successional changes can be accepted if based on the presence or absence of actively growing mycelia since the appearance of sporulating structures bear little relationship in time to the appearance or disappearance of the mycelium. Actively growing mycelia may be present; but may never sporulate or the sporulation be delayed for a long period. The tendency for the climax of successions to become dominated by one or two highly antagonistic species may also result in changes in the rate of decomposition as the succession develops. Rich species diversity at the beginning of succession corresponds to highest rates of decomposition. Eventually, the rate of decomposition at the climax of succession becomes that of the most vigorous competitor. These have slow growth rates with lower metabolic activity and hence the rate of decomposition also becomes slow (Dix and Webster, 1995).

Phylloplane fungi persist on the fallen leaves of angiosperms and species of Cladosporium, Aureobasidium and others were isolated from the leaf litter for many months after the leaf fall (Hogg and Hudson, 1966); several also known to produce their sexual stages there (De-Boois, 1976). More enduring leaf litters typically develop a secondary flora of litter microfungi, the sporulating structures of which usually appear about a year after leaf fall (Hogg and Hudson, 1966). Once in the litter,

leaves become colonised by specis of typical soil-inhabiting fungal genera such as *Doratomyces*, *Humicola*, *Fusarium*, *Gliocladium*, *Penicillium*, *Trichoderma*, etc. and as time passes these become dominant as leaves are buried and get into the deeper layers of the litter (Dix and Webster, 1995). Some of the common autochthonous soil fungi associated with tree leaf litter appeared to play only a minor role in its direct decomposition (De-Boois, 1976).

Most studies on the succession of fungi on litter have been carried out in the temperate and some notable examples include *Pinus sylvestris* and *P.nigra* by Ward (1952), *P. sylvestris* by Gremmen (1957), *Quercus ruber* by Witkamp (1960), *Fagus crenata* by Garrett (1963) and Saito (1966), *Abies grandis*, *Picea sitchensis* and *Pinus sylvestris* by Hayes (1965a), maple, elm, and ash by Novak and Wittingham (1968), *Shorea robusta* by Mishra (1969) *Abies grandis*, *Pinus monticola* and *P. ponderosa* by Brandsberg (1969), *Fagus sylvatica* by Hogg and Hudson (1966), *Eucalyptus regnans* by Macauley and Thrower (1966), Oak, birch and hazel by Hering (1965), *Nothofagus truncata* by Ruscoe (1971), *Castanopsis cuspidata* and *Quercus phillyraeoides* by Tubaki and Yokoyama, (1971, 1973a, 1973b), *Eucalyptus maculata* by Eicker (1973), *Pinus taeda* by Watson *et al.*, (1974) and *Populus tremuloides* by Visser and Parkinson (1975).

The first detailed study of fungal succession on coniferous litter was by Kendrick and Burges (1962) who followed the colonisation of leaf litter of *Pinus sylvestris* by fungi and found out that in litter, fermentation and humus layers, of the pathogens present on living needles, viz. *Lophodermium pinastri*, *Coniosporium* sp. and *Fusicoccum bacillare*, *Coniosporium* sp. did not survive even on the litter; *Lophodermium pinastri* remained active and sporulated extensively up to 6 months after needle fall and then disappeared; *Fusicoccum bacillare* showed an extensive

development from the time of death and showed another heavy production of spores 3-5 months after leaf fall. After the needle fall, the *Verticicladium* stage of *Desmazierella acicola* was the common coloniser. *Aureobasidium pullulans* was replaced on the surface by *Helicoma monospora* and *Sympodiella acicola* which appeared on the needles even in the litter layer. In the fermentation layer the external colonisers were *Trichoderma viride* and *Penicillium* sp. and the internal colonisers were basidiomycetes and a sterile dematiaceous fungus.

Succession of fungi which occurs as the leaf ages could also be correlated to the changing nutrient status of the leaf (Macauley and Waid, 1981). The initial colonizers, the yeasts and yeast like fungi utilize simple carbon compounds or leachates which are exudated from the living leaf onto the leaf surface. As the leaf ages, the frequency of filamentous fungi increases in correlation with the increasing amount of exudate. At the stage where these exudates are exhausted, fungi subsist on other available substrates such as the cellulose that persist even on the dead tissue

For conifers, the pattern of development of the fungus flora on needles in litter had some general features in common with the mycoflora developing on the leaf litter of angiosperm trees (Dix and Webster, 1995). One similarity was that the leaf-inhabiting fungi of the phylloplane persisted on the needles in the litter for several months after needle fall and some went on to produce their sexual stages. Among these were a small group of needle-inhabiting fungi which appeared first on living needles in very low numbers but became more abundant as the needles reached the litter. In the litter, the decaying needles were invaded by litter-inhabiting fungi which completed the decomposition of the needles.

Dilly and Irmler (1998) studied the functional structure within the biota during the decomposition of leaf litter in a black alder forest in northern Germany. The succession of the food web was analysed at a dry and wet site close to a lake with eight, four, and seven functional groups of bacteria, fungi and fauna. The decomposition process was divided into two phases separated by the summer dryness. During the first phase cellulolytic bacteria, omnipotent and minor potent fungi were present together with mycetophagous, saprophagous and humiphagous soil animals.

Derived from trophic relationships between the functional groups, a food path was suggested by Dilly and Irmler (1998) for the first phase from litter via celluloytic bacteria to microphagous and saprophagous soil fauna and their predators. In addition, food paths led from litter via different fungal groups to mycetophagous soil fauna and their predators. During the second phase of decomposition the number of food paths was reduced. Only fungi without lignolytic potential persisted and saprophagous animals predominated. A retarded occurrence of nitrifying bacteria was observed which suggests increasing ammonium and nitrite concentration during decomposition. High correlation was found between general bacteria and proteolytic bacteria referring to an internal protein flux within these functional groups. The number of trophic links was higher during the first phase.

Rauni et al. (1999) studied the microbial composition in a primary successional sequence on the forefront of Lyman Glacier, Washington, United States. They sampled microbial communities in soil from nonvegetated areas and under the canopies of mycorrhizal and nonmycorrhizal plants from 20 to 80 year old zones. Three independent measures of microbial biomass were used: substrate-induced respiration (SIR), phospholipid fatty acid analysis (PLFA), and direct microscopic counts. All methods indicated that biomass increased over successional time in the nonvegetated soil. The PLFA analysis indicated that the microbial biomass was greater under the plant canopies than in the nonvegetated soils; the microbial

community composition was clearly different between these two types of soils. Over the successional gradient, the microbial community shifted from bacteria-dominated to fungi-dominated set up. Microbial respiration increased while specific activity (respiration per unit biomass) decreased in nonvegetated soils over the successional gradient. The maximal respiration rate and the total C released from the sample decreased sharply over the successional gradient. They proposed and recommended new parameters for estimating the carbon use efficiency of the soil microbial community. The study suggested that during the early stages of succession, the microbial community cannot incorporate all the added substrate into its biomass though rapidly increased its respiration.

Pennanen et al. (1999) studied the structure, biomass and activity of the microbial community in the humus layer of boreal coniferous forest stands of different fertility. The Scots pine dominated Calluna vulgaris type (CT) represented the lowest fertility, while Vaccinium vitis-idaea type (VT), Vaccinium myrtillus type (MT), and Oxalis acetocella-Vaccinium myrtillus type (OMT) following this order, were more fertile types. The microbial community was studied more closely by sampling a succession gradient at the MT site. The phospholipid fatty acid analysis (PLFA) revealed a gradual shift in the structure of the microbial community along the fertility gradient even though the total microbial biomass and respiration rate remained unchanged. The relative abundance of fungi decreased and that of bacteria increased with increasing fertility. The spatial variation in the structure of the microbial community was studied at a MT site. Semivariograms indicated that the bacterial biomass, the ratio between the fungal and bacterial biomasses, and the relative amount of PLFA were spatially autocorrelated within distances around 3 to 4 m. The total microbial and fungal biomasses were autocorrelated only up to 1m. The spatial

distribution of the humus microbial community was correlated mainly with the location of the trees, and consequently with the forest floor vegetation.

The succession in physiological capabilities of bacterial and fungal communities was studied during leaf litter decomposition within the first 12 months at a drier and a wet site in a black alder forest (Dilly et al., 1998). Eutrophic and proteolytic bacteria were positively and cellulolytic and lipolytic bacteria negatively correlated. In many cases, densities of bacterial populations were positively correlated with fungal enzymatic potentials indicating a concerted action of bacterial and fungal communities during degradation of litter constituents. Cellulolytic bacterial numbers were positively linked with polygalacturonic and lignolytic potential of the fungi indicating a fine-tuned mineralization. However, lipolytic bacterial numbers and the respective potential of fungi were negatively correlated which suggests shifting importance of bacteria and fungi for lipid degradation. The fungal communities seem to play a predominant role in the litter breakdown at early stages whereas bacteria succeeded later in order to complete the process of mineralization. The data were related to microbial carbon content, activities and abiotic properties.

The dynamics of fungal and bacterial potentials in the decomposition of leaf, branch and bark litter along a gap size gradient in a subtropical forest was determined using substrate-induced respiration (SIR) with antibiotics selective for fungi and bacteria, respectively (Zhang and Zak, 1998). Fungi had higher SIR than bacteria for each type of litter in any size of gaps. Decomposing leaf litter exhibited higher fungal and bacterial SIRs than branch and bark. Correlation analysis indicated that fungal SIR was a reliable index of decomposition rates. Fungal SIR was positively correlated with soil moisture whereas bacteria was not. The relationships among microclimatic factors, fungal and bacterial physiological activities and rates of plant litter

decomposition suggested that in the subtropical ecosystems, fungal activities were strongly and directly regulated by the environmental heterogenity within gaps and are important regulators of rates of plant litter decomposition.

There have been a few detailed studies of the fungal successions on lower plants. Kilbertus (1968) studied the moss, *Pseudoscleropodium purum*, and Frankland (1966, 1969) and Godfrey (1974) investigated *Pteridium aquilinum*. Dix and Webster (1995) indicated that there are differences in the mycoflora of the litter of lower plants. Fronds decayed more slowly than angiosperm leaves and were invaded early in fungal succession by basidiomycetes and deuteromycetes which become dominant by the end of the second year. This kind of succession resembled the decay of wood, probably due to similarities in the presence of low nitrogen level and high lignin content.

Herbaceous litter - Monocots:

Very few detailed comparative studies on herbaceous plant litter are known and in this respect, the early investigations by Webster (1956, 1957) on *Dactylis glomerata* are exemplary. His studies revealed several distinct observations. On upright stems, from the upper to lower internodes, four groups of fungi were recognised. Group I were the primary saprophytes of the leaves and stems, consisting of *Alternaria tenuis*, *Cladosporium herbarum*, *Epicoccum purpurascens*, *Leptosphaeria microscopica* and *Pleospora vagans*. Sporulation of primary saprophytes was first recorded in low frequency on leaves at lower internodes in May and progressed upwards as the season advanced. They persisted at the upper internodes for about 15 months until the stem collapsed in the second winter. Primary saprophytes that were recorded immediately after the flowering at lower internodes

which did not spread to upper internodes as senescence progressed made up group II, as typified by *Acrothecium sp*. Group III consisted of *Mollisia palustris* and *Tetraploa aristata* which appeared at the lower internodes. The sporulating structures of these secondary saprophytes were not recorded until the following spring. This was followed by the appearance of group IV consisting of *Helminthosporium hyalospermum* and *Tetraploa aristata* which fruited in the following summer.

Hudson and Webster (1958) studying Agropyron repens revealed a remarkably similar pattern of colonization which differed only in some qualitative aspects of the mycoflora. Agropyron repens showed major differences in the distribution of species at different levels on the stems and this suggested that moisture content or the nutritional status of stems are more important regulators of fungal growth than atmospheric humidity. The differences in fungal colonization of the upper and lower internodes were attributed to factors such as water content, nutritional status, host resistance and competition with the organisms.

Pugh (1958) discussed the distribution of fungi on *Carex paniculata* by studying the leaves from previous years and recently dead leaves. He found that the older litter harboured less number of fungal species than the recently dead leaves.

Webster and Dix (1960) worked on the culms of *Dactylis glomerata* to analyze the nutritional status of the upper and lower internodes and their ability to support fungal growth. They also looked into some of the factors controlling the pattern of colonization which was an extension of the experiments conducted by Hudson and Webster (1958). They found that upper internodes had a higher nutritional status in the early periods of colonization. Primary colonizers were capable of rapid colonization, the spores germinated rapidly and the mycelium spread at lower relative humidities than secondary colonizers.

Similar patterns of succession have been observed on plants growing in other climates. In warmer regions, there are differences in the species composition with a tendency for the species diversity to increase. Studying fungal succession on the leaves of sugarcane, Hudson (1962) observed much a pattern irrespective of the position of the leaves on the stem. He recognised three groups of fungi. Very early colonisers of green leaves (group I), viz. Guignardia citricarpa, Leptospaeria sacchari and other parasites. These were joined by Alternaria tenuis, Cladosporium herbarum, Curvularia lunata and Nigrospora sphaerica as the leaves senesced and all of them first appeared on basal leaves and then spread upward. The early colonisers were followed 2-3 months later by group II fungi consisting of Lacelliniopsis sacchari, Periconiella echinochloae and Pithomyces maydicus. These were in turn followed by group III of fungi which included Anthostomella minima, Apiospora camptospora, Didymosphaeria sp., Entosordaria deightonii, Lacellina graminicola, Lophodermium arundinaceum, Metasphaeria sp., Pleospora vagans, Spegazzinia tessarthra and Tetraploa aristata.

Meredith (1962) studied the mycoflora on collapsed and decaying banana (Musa sapientum) midrib, petiole and lamina. The primary colonisers consisted of Deightoniella torulosa, Gloeosporium musarum, Nigrospora sp., Pyricularia mussae and Verticillium theobromae. Verticillium theobromae and Deightoniella torulosa were most prominent on petioles and midribs. As the leaves dried, the primary colonisers were replaced by species of Acremonium, Alternaria, Aspergillus, Cladosporium, Fusarium, Paecilomyces, Penicillium and several others.

Khanna (1964) studied the succession of fungi on three decaying grasses, viz. Bothriochloa pertusa, Cynodon dactylon and Dichanthium annulatum. Fungal succession on decaying leaves of Saccharum munja was studied by Rai (1973) for over two years and observed a similar pattern of colonisation like that of Hudson (1968) on *S. officinarum*. Sharma and Dwivedi (1972) recorded the mycoflora colonising different portions of the shoot system of fodder grass, *Setaria glauca*, from early senescence onwards. Fungal flora of the air overlapped with a majority of the fungi isolated from the shoots of the grass. The number of fungal species recorded from stem segments was lesser than that on blades and sheaths. This they attributed to several morphological and anatomical features of different plant parts and ecological factors such as moisture content of the substrates, temperature and relative humidity of air and competition between colonisers.

Rai (1974-75) suggested a general scheme for fungal succession on decaying grasses of the tropics. All grasses that had been studied commonly showed the presence of dominant members, though they differed in the frequency of occurrence on different substrates. Deuteromycetes and a few ascomycetes were the prime colonisers of grasses. Phycomycetes and Basidiomycetes were not recorded on any of the grasses subjected to study.

Herbaceous litter - Dicots:

The green and moribund leaves of *Halimone portulacoides* was studied by Dickinson (1965). Three groups of phylloplane fungi were recognised: transient, lying on the surface of the leaf, consisted of the first group; fungi such as species of *Cladosporium* thriving and sporulating at ease form the second group; the third group consisted of species such as *Ascochytula obionis* that form pycnidium on moribund leaves. Though there was similarity between the mycoflora of *Halimone* and *Dactylis* (Webster, 1957), the frequency of occurrence of *Ascochytula obionis* made a prominent difference. Dickinson (1965) also stated that abundance of air-borne spores

such as Aspergillus sp. and Penicillium sp dictated their presence on the leaves.

Kerling (1964) found a similar trend in fungal colonisation on strawberry and rye litter.

Herbaceous plants of Calluna vulgaris, Festuca sp., Melandrium sp. and Vaccinium myrtilis were assessed for fungal colonisation at different stages of decomposition (Mangenot, 1966). Species of Cladosporium, Mucor and Rhizopus were dominant at the early stages while those of Chaetomium, Fusarium and Trichoderma were frequent during the later stages on Calluna and Vaccinium litter. Species of Penicillium such as P. aurantio-candidum, P. janthinellum and P. frequentans were also present in large numbers. Species of Fusarium were the dominant colonisers throughout the decomposition of Melandrium litter. Chaetomium globosum, C.indicum, Cladosporium sp., Mucor sp. and Rhizopus sp. were major colonisers on Festuca litter.

Yadav (1966) recognised five groups of fungi based on the frequency of their occurrence on decaying stems of *Heracleum sphondylium*. With senescence, *Alternaria tenuis*, *Cladosporium herbarum*, *Botrytis cinerea*, *Coniothecium* sp., *Epicocum nigrum* and *Phomopsis astericus* were first observed on the leaves and leaf sheaths. The lower internodes were then attacked by *Acremonium* sp., *Cladosporium herbarum*, *Dendryphion comosum*, *Epicoccum nigrum*, *Hormiscium* sp., *Periconia cookei*, *Phoma complanata*, *Stachybotrys atra* and *Torula herbarum* without any localised pattern of colonisation. He concluded that the primary mycoflora, which appeared on the stem in the year of their growth were possibly deposited there by wind. The secondary mycoflora which appeared in the winter following summer, characteristic of lower internodes, probably arrived from the soil and gradually spread upwards. His findings were parallel to those reported on *Dactylis* by webster (1957).

Dickinson (1967) working on leaves of *Pisum sativum*, leaves found that the major fungi on senescent and dead leaves were *Alternaria* sp., *Aureobasidium* sp, *Cladosporium* sp and *Stemphylium* sp. On stems of *Urtica dioica* Yadav and Madelin (1968) found out that *Alternaria tenuis*, *Botrytis cinerea*, *Cladosporium herbarum* and *Epicoccum nigrum* were the primary colonisers while the lower portions were colonised by these along with *Acremoniella atra*, *Alternaria tenuis*, *Cladosporium herbarum*, *C. sphaerospermum*, and *Phoma acuta*.

Sharma and Mukerji (1972) reported the results of taxo-ecological investigations on the mycoflora of leaves of *Gossypium hirsutum* L. at different stages of senescence, while still attached to the mother plant and after abscission. The effect of seasonal variations in temperature, relative humidity, soil pH and moisture content on the quality and quantity of the mycoflora were established. *Candida albicans* and *Phoma spp*. showed remarkable fluctuations in the number of propagules per gram dry weight of leaves when correlated with seasonal variations in temperature and relative humidity.

Vittal (1973) made a detailed study of the fungi colonising leaves and litter of two dicotyledonous plants, *Atlantia monophylla* and *Gymnosporia emarginata* collected from Vandalur, Madras, over a two year period. The fungi of leaves and litter of both plants were studied in the first year. In the following year, litter from each plant was graded into three on the basis of the extent of decomposition and the fungi on each grade of litter were analysed into five groups, viz. dominant, common, frequent, occasional and rare. The number of fungal species recorded were greater on *Atlantia* than on *Gymnosporia*. Deuteromycetes were the dominant members on both the plants; in addition, myxomycetes, phycomycetes and ascomycetes were also observed on *Atlantia*. *Beltaniella portoricensis*, *Sesquicillium setosum* and

Ophiognomonia sp. were dominant on Atlantia, while Beltrania rhombica, Idriella vandalurensis and Pestalotia theae were dominant on Gymnosporia litter. Quite a number of fungal species were common to both the plants although the frequency and percentage occurrence differed for both plant litter types.

Many species were common to all three grades of litter and some restricted to only a single grade of litter. Percentage occurrence of each species was considered to be an index of activity on litter. Among the species common to all grades of litter, some differences in activity of the species on each grade were found. For example, Beltaniella portoricensis, Sesquicillium setosum, Cladosporium herbarum, Volutina concentrica and Ophiognomonia sp. were the most active in that order, on grade 1 litter of Atlantia; Beltraniella portoricensis, Sesquicillium setosum, Gyrothrix circinata and Ophiognomonia sp. were the most active, in that order on grade 2; Pyrenochaeta sp., Sesquicillium setosum, Stachybotrys chartarum, Beltraniella portoricensis and Ophiognomonia sp. were in that order the most active on grade 3 litter of Atlantia. Similarly, Pestalotia theae, Idriella vandalurensis, Gyrothrix circinata and Cladosporium herbarum were the most active in that order on grade 1 litter of Gymnosporia and Idriella vandalurensis, Gyrothrix circinata, Pestalotia theae and Stachybotrys chartarum were the most active, in that order, on grades 2 and 3 litter of Gymnosporia.

Vittal (1973) also compared the fungi isolated on *Atlantia* and *Gymnosporia* litter collected from different localities. This highlighted the qualitative similarity in the mycoflora of litter of both plants. It was observed that *Beltraniella portoricensis* restricted to *Atlantia* litter at Vandalur was recorded on *Gymnosporia* litter from Kambakkam, and likewise, *Beltrania rhombica* restricted to *Gymnosporia* litter at Vandalur was recorded on *Atlantia* litter from Kambakkam.

Phylloplane mycoflora of *Atlantia* and *Gymnosporia* were also studied by Vittal (1973). *Drechslera hawaiiensis, Nigrospora oryzae* and *Pestalotia theae* were common to the phylloplane of both plant species. *Rhinocladiella* sp. was found to selectively colonise green and yellowed leaves of *Gymnosporia* but not *Atlantia*. For both plant species, over 50% of the phylloplane fungi continued to be frequent on grades 1 and 2 of the litter, but on grade 3 litter, their percentage was very much lesser. A survey of the air mycoflora showed an abundance of propagules of species found on the phylloplane of *Atlantia* and *Gymnosporia*.

Sudha (1978) studied Glycosmis cochinchinensis and Ixora parviflora from a scrub jungle at Thambaram, Madras, to obtain information on the nature of the mycoflora active during different phases of decomposition of litter. Litter samples were collected once a month for a period of two years. Each monthly sample was sorted out into 3 grades on the basis of extent of decomposition. Grade 1 being the freshly fallen leaves which have undergone little decomposition; grade 2 represented litter more decomposed than grade 1 litter and grade 3 represented highly decompsed litter. Mycoflora of each grade of litter of the 2 plant species was studied by moist chamber incubation and dilution plating techniques. Besides, senescent leaves of each plant species collected every month were allowed to undergo decomposition in a separate experimental set up constructed to simulate conditions found in nature, in which yellowed senescent leaves still attached to the plant were collected every month and were placed in layers one above the other separated by a nylon mesh, which maintained physical continuity between the 12 layers. The different layers were then assessed for mycoflora at the end of 1 year. In all, 118 species in 83 genera were recorded on both plants. This included myxomycetes (2 species), mucorales (6 species), ascomycetes (7 species), coelomyctes (11 species) and hyphomycetes (92

species). As a result of the study she obtained 57 fungal species on *Glycosmis* and 70 on *Ixora* litter. *Colletotrichum dematium*, *Linospora* sp, *Sesquicillium setosum* and *Volutina concentrica* were found exclusively on *Glycosmis* litter, whereas *Endophragmia alternata*, *Weisneiriomyces javanicus* and *Zygosporium masonii* were confined to *Ixora* litter. *Beltrania rhombica*, *Beltraniella portoricensis*, *Scolecobasidium constrictum* and *Trichoderma harzianum* were common colonisers on both substrates. *Endophragmia alternata*, *Helicosporium vegetum* and *Rhinocladiella* sp. occurred exclusively on grade 3 litter of *Ixora*. Similarity indices of the mycoflora of litter of the 2 plant species from the different layers of experimental set up showed that mycoflora on litter from adjacent layers had the greatest similarity, the similarity decreasing with increasing distance of the layers.

On the basis of the frequency and the colonising efficiency of the different species, Sudha (1978) proposed that for both plant species, the first colonisers on litter were predominantly a few weak parasites on living or senescent leaves, followed in succession by true litter fungi which were replaced in the final stages of decomposition by soil inhabiting fungi.

Dorai (1988), worked on the taxonomic and ecological aspects of the fungi colonising the leaf litter of *Eucalyptus* species in India. The examination of the leaf litter of 13 species of *Eucalyptus* resulted in the isolation of 264 species belonging to 170 genera. The majority of species belonged to the Deuteromycotina (84%), though members of myxomycetes, Zygomyccetes, Ascomyccetes and Basidiomycetes were also represented. Of the 264 species, 22, constituting 8.3% were undescribed species. Some fungi were specific to a particular host. Seven species were specific to *E. citriodors*; 13 to *E. deglupta*; 44 to *E. globulus*; 1 to *E. grandis*; 14 to *E. longifolia*; 1 to *E.maculata*; 53 to *E. tereticornis* and 4 to *E. torelliana*. No host specific species

were recorded from E. eugenioides, E. ficifolia, E. macrorhyncha, E. regnans and E. saligna. The number of host specific species recorded on E.globulus and E. tereticornis were greater when compared to the remaining 11 species of Eucalyptus. Dorai attributed this to the distribution of the different host species in South India; E.globulus was most commonly grown in the hilly tracts of Andhra Pradesh, Kerala and Tamil Nadu while E. tereticornis was grown in the plains of Karnataka, Kerala and Tamil Nadu. Hundred and four species were common to different species of Eucalyptus. Corynespora cassicola was recorded only from E. globulus and E. tereticornis, while Weisneiriomyces javanicus was recorded from 10 species. kakombensis, Cryptocoryneum Haplographium Cryptophiale rilstonii, helicocephalum, Hyphodiscosia jaipurensis, Parasympodiella laxa Pseudopetrakia kambakkamensis were reported for the first time from Eucalyptus litter. Comparing the microfungi from the studies of Vittal (1973) and Sudha (1978) on Atlantia monophyla, Gymnosporia emarginata, Glycosmis cochinchinensis and Ixora parviflora, Dorai (1988) listed the following fungi common to all these plants. Beltrania rhombica, Beltraniella portoricensis, Corynespora cassiicola, Curvularia eragrostidis, C. tuberculata, Cylindrocladium parvum, Gyrothrix circinata, Memnoniella echinata, Periconia hispidula, Periconia cookei, Weisneiriomyces javanicus, Kramasamuha sibika, Zanclospora indica. The last two fungi were a new genus and new species recorded from Vandalur. He concluded that the similarity in the mycoflora associated with leaf litter of plants belonging to unrelated but growing in the same locality suggests that while the nature of substrate is an important factor, the geographical location of the sampling area and its biogeoclimate also plays a major role in deciding the nature of the mycoflora of that particular area.

Dorai (1988) also made ecological investigations on the fungi colonising leaves and litter of Eucalyptus tereticornis collected at bimonthly intervals from a 17year old plantation at Vandalur over a period of two years. The objective of this study was to know the nature of the fungus flora of the phylloplane and to understand the sequence of fungal colonisation of the living leaves, senescent and dead leaves by using moist chamber incubation technique and dilution plating. Fungi were grouped as 'most frequent', 'common', 'occasional', and 'rare' depending on their periodicity of occurrence. A total number of 119 species belonging to 88 genera were isolated from all the three layers of litter, the greater number being isolated from F₁ layer than in L and F₂ layer. Arthrinium phaeospermum, Beltrania malaiensis, Chlamydomyces palmarum, Corynespora cassiicola, Monodictys castaneae and Torula herbarum were recorded exclusively from L layer. Cercosperma longispora, Coniella castaneicola, Hansfordia ovalispora, Harknessia ventricosa, Microdochium caespitosum, Mycotypha microspora and Rhinocladiella mansonii were specific to F₁ layer. Choanephora cucurbitarum, Chaetomium turgidopilosum, Dactylaria purpurella, Polyscytalum sp., Scolecobasidiella tropicalis, Spadicoides aggregata, Stachybotrys kampalensis and Stemonitis virginiensis were recorded from F, layer.

A clear pattern of fungal colonisation of leaves and litter of E. tereticornis was observed by Dorai (1988). The phylloplane mycoflora consisting of Alternaria alternata, Aspergillus niger, Cladosporium cladosporioides, C.oxysporum, Curvularia sp. and Penicillium funiculosum common to airflora were recorded. As the leaves became senescent and shed, these foliicolous fungi began sporulating on freshly fallen leaves represented by L layer. True litter fungi, Gyrothrix circinata, Parasympodiella laxa, Phragmocephala sp. and Weisneiriomyces javanicus appeared afresh in L layer and continued to be active in F_1 , where more true litter fungi such as

Cercospora longispora, Harknessia ventricosa, Helicoubisia coronata, Hyphodiscosia jaipurensis, Kramasamuha sibika, Pleurotheciopsis tax. sp., and Zanclospora indica were observed. Some of these fungi continued to appear in F_2 layer which consisted of litter in advanced stage of decomposition

Bhat and kaveriappa (1999) studied the phylloplane and surface mycoflora of aerial parts such as shoot bud, flower bud, flower and fruit of *Myristica fatua* var. *magnifica* and *M. malabarica*, two endangered tree species of evergreen forests of the Western Ghats in Uttara Kanada, in Karnataka, using serial dilution and blotter methods. A total of 83 species belonging to 48 genera were isolated. Among these, 61 species were recorded on *M. malabarica* and 72 species on *M. fatua* var. *magnifica*. Maximum number of species were recorded on mature leaves and minimum on flower buds. *Alternaria alternata*, *Aspergillus aculeatus*, *A. niger*, *Cladosporium oxysporum*, *Fusarium oxysporum*, *Penicillium chrysogenum* and *Trichoderma viride* were found on all parts of both plant species. Maximum number of fungal species were recorded on mature leaves and shoot buds during summer months (Dec-March), while minimum number of species were recorded during rainy season (June-Sept.)

Roberts et al. (1986) studied the fungi occurring in the achenes of Helianthus annus. About 28,000 samples of achenes from several production areas in the United states were subjected for analysis. Ninety-eight species in 38 genera were identified from graded samples of achenes stored at 20°C, and 63, 83 and 93% relative humidities. Sixty four fungal taxa were reported from sunflower aches for the first time. Forty-five potentially mycotoxigenic, five thermophillic and five new records of species of Microascus were isolated.

Litter in semi-aquatic habitats:

Fungi colonising aerial stems, leaves and roots of Salsola kali were categorised into three groups by Pugh and Williams (1968). Group I consisting of Acremonium sp. and Fusarium sp. were commonly associated with aerial parts, although these were frequently isolated from the roots. Group II such as Acremoniella atra, Alternaria tenuis, Botrytis cinerea, Camarosporium sp., Cladosporium herbarum, Epicoccum nigrum and Stemphylium sp. were more prominent on the aerial parts than on the roots. Group III had only a Chaetomium sp. that was isolated from buried stems and leaves. Pugh and Mulder (1971) traced the succession of fungi colonising Typha latifolia right from the time of its appearance, senescence and to its final decay. In the early stages, the leaf was colonised by typical leaf surface fungi, in the secondary stages pyrenomycetes dominated. In the final stages soil fungi were replaced by predacious fungi.

The mycoflora of submerged leaves of *Phragmites communis* in various stages of development, i.e. senescent, dead and decaying leaves from various habitats in England, were compared (Apinis *et al.*, 1972, 1975). A total of 49 species were recorded, *Acremonium* sp., *Alternaria tenuis*, *Cladosporium herbarum*, *Dasycyphus controversus*, *Diplosporium* sp. and *Oidiodendron fuscum* were common to all the 6 habitats studied. Young culms harboured few fungi while the number of species on the nodes and internodes increased with age

Van-Maanen and Gourbiere (1997) have studied the host and geographical distribution of *Verticicladium trifidum*, *Thysanophora penicilloides* and similar fungi on decaying coniferous needles and conclude that the coexistence of these dematious hyphomycetes on some samples and the colonisation of some Pinus litter by

Thysanophora penicilloides suggests that these distributions result from competition rather than strict host specificity.

The ecological mechanisms by which plant biodiversity and species composition are regulated and maintained are not well understood. Marcel *et al*, (1998) made an attempt to show that below ground diversity of arbuscular mycorrhizal fungi is a major factor contributing to the maintenance of plant biodiversity and to ecosystem functioning. It also shows that conservation of the fungal gene pool is likely to be prerequisite for maintenance of floristic diversity in grasslands, as well as in other ecosystems such as boreal forests, where the fungal web is known to influence allocation of resources between plant species (Read, 1998).

Raviraja et al. (1998) studied the fungal colonisation and processing of eucalyptus and banyan leaves in organically enriched reaches of the river Nethravathi in coastal Karnataka in southern India. They found that conidial production and species numbers of aquatic hyphomycetes were very low. Comparisons with their earlier studies (Raviraja et al., 1996) that is with geographically close but clean streams showed that pollution was the determining factor.

Abundance and diversity of microfungi in tropical litter:

Knowledge on plant-inhabiting tropical microfungi has been based on collections of fungi sporulating on their natural substrata in situ or those sporulating on plant debris incubated in moist chambers. Such collections have been studied in order to document fungal floristics, and to provide basic data for taxonomic monographs. These studies have revealed that tropical plant debris exhibits an enormous diversity of fungi and further provided countless descriptions of species with which workers can identify the microfungi of their region. Only few

investigators have so far attempted to measure the abundance of diversity of microfungal species that inhabit tropical litter (Heredia, 1993; Bills and Polishook, 1994).

Based on particle-filtration technique, Bills and Polishook (1994) estimated how many species of saprobic microfungi could be expected on decaying leaves of a single plant species, *Heliconia mariae* in the lowlands of south-eastern Costa Rica. Pulverized decayed leaves were separated into fine particles and repeatedly washed. When 0.1 ml particle suspensions were plated onto 4 petri plates of two selective media, a total of 1676 isolates were recovered, ranging from 310 to 599 isolates/plant. The number of species /plant ranged from 56 to 98.

Bills and Polishook (1994) devised the particle filtration technique based on the conventional soil washing method, to determine if the characteristic fungal flora of leaf litter could be preferentially isolated while minimising recovery of soil and common saprobic fungi. They also made preliminary measurements of the magnitude of fungal species richness in tropical forest litter. Rarefaction curves based on the number of species expected in random subsamples were used to compare species richness among samples. From their study many uncommon genera of litter fungi belonging to coelomycetes, sterile strains, endophytes and phytopathogens were recovered. Typical soil fungi were a relatively minor component of the total isolates. Species abundance distribution showed that there were few abundant species and a high proportion of rare species. Species present in all samples belong to genera *Cylindrosympodiella, Glomerella, Lasiodiplodia* and *Pestalotiopsis*. Hyphomycetes and coelomycetes were the most abundant type of fungi. Some of the true genera of litter fungi isolated included *Beltrania*, *Chalara Chloridium*, *Cordana*, *Cryptophiale*,

Dendrosporium, Dichtyochaeta, Gyrothrix, Kutilakesopsis, Leptodiscella, Speiropsis, Tetracladium, Thozetella, Trinacrium, Volutella and Zygosporium.

Limited sampling of endophytes from leaves and stems of trees at the same locations of litter samples by Bills and Polishook (1994) indicated that some of the endophytic species appeared in the litter layer as well. For example, *Cylindrocarpon* sp., *Fusarium decemcellulare*, *F. solani*, *Glomerella cingulata*, *Lasiodiplodia theobromae*, *Nodulisporium* sp., *Phomopsis* sp., *Tubercularia lateritia* and *Xylaria* sp. and many species of coelomycetes. This observation was similar to the studies made by Boddy and Griffith, (1989), Parkinson and Kendrick (1960) and Wildman and Parkinson (1979) wherein they found that endophytic and phytopathogenic fungi are commonly recovered from the upper layers of forest litter and are associated with litter decomposition.

Fungus flora of the air over a wheat field was studied by Misra and Tewari (1975). Air was sampled at heights of 15, 30 and 45 cm, by exposing nutrient plates; simultaneously the wheat leaf samples were also collected from the corresponding heights for assessment of leaf surface mycoflora. They found that the number of spores on the leaf surface was nearly proportional to the number of spores in the air. The fungal population was highest at 15cm and decreased with increasing height. The population also increased from November to March in both the cases. Seventy percent fungal species were trapped from both the environments whereas only 3.5 and 26.4% were restricted to air and the leaf surface respectively.

The Endophytes:

Detailed study of the fungal endophytes commenced only in the middle of the 1970's (Berstein and Carroll, 1977; Carroll and Carroll, 1978), although their presence was first discovered by Sampson in 1935 within the plant tissues of *Festuca rubra* (Petrini1991) microorganisms are now known to interact with surfaces as well as interior tissues of plants. All living plants so far investigated have been shown to harbour fungi inside their tissues (Petrini1991).

The term endophytes was originally used by De Bary (1866) to refer to any organism occurring within plant tissues, distinct from the epiphytes that live on plant surfaces. Microbes living within the interior tissues of healthy plants, without causing any disease symptoms, are called endophytes (Wilson, 1993). It is now known that the fungi in grasses and trees living asymptomatically within the host plant give the host acquired resistance against herbivores (Carroll, 1988; Clay, 1988; Isaac, 1992). Some of these fungi are considered to be mutualistic, because they afford host plants a degree of protection from herbivory. Although, the term endophyte has been used to describe mycorrhizal fungi (O'Dell and Trappe, 1992), because of their characteristic external hyphae extending into the soil surrounding the infected root tips and such fungi necessarily residing only partly inside plant tissues, the taxonomic limit to the definition of an endophyte now remains an ongoing biological debate. In addition, several bacterial endophytes have also been recognized (Chanway, 1996).

Petrini 1991 consideres all organisms inhabiting internal tissues of plant organs at some time in their life without causing apparent harm as endophytes. This consideration also includes latent pathogens which are found as endophytes during stages of their life cycles. This definition obscured the boundaries between epiphyte, endophyte and latent pathogen. Some fungi persist within the plant as endophytes and

in order to facilitate the infection of other plants, release its spores into the air. During this stage, the fungus might be seen as an epiphyte living on the surface of the plant leaves, where spore dispersal into the air may be achieved. In this form, an endophyte with external structures can be seen as an epiphyte with hyphae growing into the plant (Clay, 1991). Endophytes may also be weak plant pathogens, for example some smuts which are systemic and inhibit host growth (Clay, 1991). A species of *Rhabdocline*, a weak pathogen of Douglas fir leaves, can cause no signs of infection for up to two years and, according to Carroll (1988), during this latent period, the fungus may be referred an endophyte.

Chapela (1989) used the term 'xylotropic endophytes' for fungi that were found within host trees and have the tendency of growing into secondary xylem upon drying of the wood, thereby emphasizing their relatedness to endophytic fungi.

Grass endophytes:

Taxonomy and biology of fungal endophytes of grasses (Poaceae) and sedges (Cyperaceae and Juncaceae) have been the subject of extensive studies, mainly because of the impact fungi on the ecology of grass populations. Diehl (1950) investigated the Balansiae (Clavicipitaceae), a group of fungi that parasitize grasses and sedges, both taxonomically and ecologically. A high degree of host specificity has been shown by *Balansia strangulans* which was found in a given site almost invariably on only one host, although other grasses known as hosts may be growing in the immediate vicinity of the infected plant. Other species of the Balansiae are also host-specific atleast at the tribe level, and infect only closely related host plants.

The first report of endophytes of grasses was published in 1924 by Lewis (Petrini, 1991). There have since been extensive reviews of fungi inhabiting terrestrial

grasses (Clay, 1991; Carroll, 1986). Species of grasses previously known as poisonous are now known to be endophytically infected by fungi (Clay, 1991). Fescue toxicosis, caused by the consumption of the ergot toxin ergovaline by grazers of tall fescue grass, was found to be correlative of the ergosterol content of grass seeds to the endophyte content of the seeds. The endophyte impact of fine fescue seed samples was confirmed from the ergosterol and microscopic analysis. The ergosterol analysis can now be used in both daignostic and research applications to predict endophyte content in samples (Richardson and Logendra, 1997).

Endophytic fungus, Acremonium spp., infections were detected from wild populations of Lolium spp., examined from 15 of 20 European countries. Of the 523 populations studied, 38% contained no infection, 48% contained 1-50% infection and 14% contained 51-100% infection. Significant correlations were obtained between the level of infection and 5 climatic variables, the highest being with evapo-transpiration and water supply deficit. Groups of Lolium populations with a high level of infection were located mostly in Mediterranean regions, where stress from summer drought is common (Lewis et al., 1997). Clement et al., (1997) underscored the potential of endophytic fungi in conferring insect resistance in wild barley. They conducted experiments to compare the expression of Diurophis noxia (Homoptera: aphididae) resistance in four plant lines of wild barley (Hordeum sp.) infected with different species of endophytic fungi [tribe Balansieae, family Clavicipitaceae, Neotyphodium gen. nov. (formerly Acremonium sp.)]. Aphid densities were significantly lower on endophyte-infected plants (H. bogdanii and H.brevisubulatum), compared with densities on endophyte-free plants of both species in population growth experiments. This endophyte-associated resistance was attributed to antibiosis effects or starvation. Allelopathy of endophytic fungi, Fusarium sp. and Colletotrichum sp., was evaluated

as factors affecting the biological control of marsh reed grass, a weed of boreal reforestation areas in a study carried out by Winder (1997).

It is now known that endophytic fungi living within some grasses have contributed to the increase in resistance of their host plants to insect herbivores. Boning and Bultman (1996) have proved that endophytes mediate induced resistance by a grass to a herbivorous insect. Tall fescue, both infected and uninfected with an endophyte, *Acremonium coenophialum*, was artificially damaged by clipping a tiller from each plant four weeks after germination. They observed that eight day old Fall armyworm (*Spodoptera frugiperda*) larvae weighed less and took longer duration to develop into adults when fed endophyte-infected vs. endophyte-free plant material. In contrast, pupae weighed more when fed infected vs. endophyte-free plant material and the interaction between infection status and damage had a marginally significant effect on pupal mass. Pupae reared from damaged infected plants weighed less than those reared from undamaged infected plants. No pattern with damage was apparent for insects reared on endophyte-free plants. The results suggested that the clipping damage could have resulted in an induced response in plants infected with the fungal endophyte.

Cheplick (1997) tried to examine whether endophytic fungi influence plastic responses of host genotypes to variable soil nutrients and whether or not endophyte infection and host genotype interact to determine the extent of this plasticity. He observed that responses to nutrient conditions in relation to fungicide treatment were genotype specific. High levels of endophytic fungi appeared to reduce plasticity. The potential for microscopic symbionts to affect phenotypic plasticity in genetically variable populations has not often been recognized. However, the clandestine effects of symbionts on the plasticity of host genotypes could impact microevolutionary

processes occuring within plant populations that occupy heterogenous environments.

Differences in species composition, infection frequencies and fungal colonization were observed in asymptomatic leaves and culms of an annual and three perennial Juncus species in western Oregon by Cabral et al., (1993). They observed that infections limited to a single host epidermal cell were characteristic of Drechslera sp., Stagonospora innumerosa and an unidentified endophyte of Juncus bufonius. Infections originated in the substomatal cavity followed by limited intercellular colonization of the mesophyll. Alternaria alternata and Cladosporium cladosporioides were isolated in low frequencies and further found restricted to substomatal chambers. Marks and Clay (1996) attempted to analyse the physiology underlying the enhanced growth rates of several grass species infected by fungal endophytes. Carbon exchange rates (CER) and leaf conductances of 13 genotypes of tall fescue infected by the fungal endophyte Acremonium coenophialum were measured. At leaf temperatures above 35°C, infected tall fescue plants photosynthesized at a significantly greater rate (20-25%) than uninfected plants. This resulted from a decrease in the CER of uninfected plants, not an increase in the rate of infected plants, at high temperature. There were also significant infection and genotype interactions, indicating that the response to infection was specific to a given genotype. The results indicated that physiological responses of host plants to fungal endophyte infection depended both on the physical environment and the genotype of the plants.

Diversity of endophytic fungi:

Several authors have suggested that evolution of land plants has been intimatedly related to that of their endophytes (Bernard, 1916; Pirozynski and Malloch, 1975; Boullard, 1979; Atsatt, 1988). According to Chapela (1989), floristic differences in the xylotropic endophytes of separate plant families might provide information on their phylogeny. Hawksworth (1991) estimated that there are 1.5M species of fungi and of which 70,000 fungal species have been described worldwide. However, this may be a vast underestimate, especially in light of the large numbers of new fungal endophyte species recovered from almost every plant species sampled. Fungi may turn out as one of the most undescribed group of organisms on earth. The major difference between the two is that most of the worlds undescribed insects are believed to reside only in the tropics whereas world's endophytic fungi still await discovery almost in every climatic zone within the leaves and stems of both common and rare plants (Wilson *et al.*, 1997).

Fisher et al. (1992) isolated fungal endophytes from five *Thymus* species collected in the mountains of Austria and in Spain. The frequency of colonisation for stems and leaves was approximately the same in the alpine samples, in contrast to the Mediterranean species, where the leaves showed low colonisation frequencies when compared to the stems. A total of 30 species had been isolated. The dominant fungi in the stems of four species of *Thymus* sampled in the Mediterranean were *Alternaria* alternata.

Fungal endophytes have been isolated from a wide range of evergreen and deciduous plants. (Carroll *et al.*, 1977; Fisher *et al.*, 1986; Petrini, 1986; Petrini and Fisher, 1986). Of the examined, 21 evergreen plants from Ishigaki and Irimote islands in Okinawa, some of the endophytic fungi were found in all the plants examined;

Xylariaceous fungi and *Phyllosticta* spp. were isolated from about half of the plants tested, *Pestalotiopsis* from 7, *Phomopsis* spp. and *Colletotrichum gloeosporioides* from 6 plants each. *Acremonium* spp., *Alternaria alternata*, *Cladosporium* cladosporiodes, *Coccomyces* sp., *Curvularia* sp., *Gliocladium roseum*, *Nigrospora oryzae* and *Phoma sp*. were also isolated from several plants (Okane et al., 1997). Bayman et al., (1997) isolated fungal endophytes from roots and leaves of seven species of epiphytic and lithophytic *Lepanthes* from rainforests in Puerto Rico. *Xylaria* spp. and *Rhizoctonia*-like fungi were the most dominant, though their differences in frequency were negligible in the root and leaf. However, differences in number and types of endophytes among orchid species were distinct. Heterogenity of endophytes in single plants and plant organs was greater than differences between species. Many *Lepanthes* species are restricted in distribution and knowledge of their intractions with endophytes is said to be helpful in species management.

Assessment of diversity, species richness and intraspecific variation within a habitat was often difficult when morphological criteria were used for identification and classification of isolates. Michael and Hallaksela (1998) have showed how combined fatty acid and sterol profiles (FAST-profiles) can be used for classification of fungal isolates into FAST-groups (i.e. operational chemotaxonomic units) according to a defined upper variation limit. They used endophytic fungi of Norway spruce needles as a model system. The endophytic fungi of *Eucalyptus viminalis* phyllosphere was studied by Bertoni and Cabral (1988). They observed that the highest level of infection is in the blade and the basal half of the leaf, followed by the midrib and petiole, and the upper half of the leaf. The more frequently isolated species recorded were *Coniothyrium* sp., *Coccomyces martiniae* and *Mycosphaerella* sp. and less frequent ones were *Macrophoma smilacina* and *Nigrospora* sp.. The

distributions showed that the infections probably developed from deposited propagules rather than systemic infection.

Petrini and Fisher (1988) aimed to evaluate host specificity of fungal endophytes in a mixed stand of two distantly related *Fagus sylvatica* and *Pinus sylvestris* and assess specificity of the endophytes with respect to whole stem and xylem. Cluster analysis showed that *Pinus* tissues can be separated from that of *Fagus* on the basis of their endophyte populations, and a K-means cluster analysis revealed that eleven of the isolated fungi were mainly responsible for this separation.

Twelve species of endophytic fungi were isolated from the leaves and stems of Suaeda fruticosa, a Mediterranean plant from England, by Fisher and Petrini (1987). They found out that Colletotrichum phyllachoroides was entirely confined to the leaves. Two species of Camarosporium were mainly isolated from the stems and a higher incidence of colonization was found for complete stems as compared with xylem. They also made a qualitative comparison of the epiphytic fungi growing on unsterilized host species with the endophytic population of complete stems and xylem. Their study showed that the most frequently occurring endophytes were not present among the epiphytes, and correspondingly, epiphytes were uncommon among the endophytic fungi.

Endophytic fungi isolated from five species of broad-leaved evergreen shrubs from 16 sites in western Oregon by Petrini *et al.* (1982) showed different rates of infection in these plants. A pattern of species dominance was with the most common endophyte of a given host when isolated less frequently from other hosts; less commonly isolated endophytes appeared to be less host specific. Site and climate related differences in the endophytic fungal assemblages of leaves, xylem and bark of *Eucalyptus nitens* from Australia and England were analyzed by Fisher *et al.* (1993).

Sixty-four fungal taxa were isolated with a relative importance of more than 5% in any of the tissues examined. Australian and British samples were clearly separated according to their geographic origin..

Fungi inhabiting healthy stems and branches of American beech and aspen were induced to respond to drying of wood. The two tree species were similar in that the water content of the wood strongly determined fungal development, with a high water content preventing fungal growth for at least 25 weeks, fast drying resulting in poor development and slow drying inducing very fast growth of fungi within the wood. The fungi, dominated by ascomycetes and coelomycetes, were clearly different for tree species, even though samples of each were obtained from the same site and the experimental conditions were identical for both. Hypoxylon fragiforme was most frequently and abundantly isolated from beech (Chapela, 1989). Xylem and bark from stems of Alnus glutinosa and whole stems of A.incana and A. viridis from England and Switzerland were screened for endophytic fungi (Fisher and Petrini, 1990). Multiple colonisation frequency was comparatively higher for bark and xylem but colonization of segments by more than two fungi were rare. Fungal communities mainly composed of a small number of dominant species accompanied by a cohort of rare isolates. Cluster analysis showed that plant organs and tissues can be separated on the basis of their endophytic fungi

Suryanarayanan and Rajagopal (1998) studied fungal endophytes from the leaves of some South Indian trees, viz; Acacia malanoxylon, A. dealbata, A. decurrens, Dalbergia latifolia, Grewia tiliaefolia, Michelia champaca, M. nilgirica, Pterocarpus marsupium and Rhododendron arboreum and Eucalyptus globulus from two places in the Nilgiri Biosphere Reserve, Tamil nadu. A total of 60 different species of endophytic fungi were isolated from the lamina and petiole of ten trees. Of

these, 37 were sterile forms, 17 belonged to Hyphomycetes, 4 were Ascomycetes and 2 belonged to Coelomycetes. Acacia melanoxylon and Rhododendron arboreum harbored more number of endophytic fungi. In all the trees a larger number of endophytes were isolated from the petiole than the lamina. Nine endophytes were unique to the lamina, 24 to the petiole and 27 occurred both in petiole and lamina tissues. Alternaria alternata was isolated from all the ten trees while Curvularia lunata occurred in nine trees. Chaetomium indicum, C. globosum, Pestalotiopsis sp., Phoma sp. and Phyllosticta sp. were also isolated, but their CF was low. They also found that the ethylacetate extract of the culture filtrates of five of the endophytic fungi increased the mitotic index of onion root considerably.

Andrews *et al.* (1985) used leaves to study the species dynamics of microbial epiphytes on apple. They suggested that leaves represent an ideal model system to examine the macroecological principles such as the theory of island biogeography because leaves were easily quantifiable units, well defined in time and space, easily replicable, subject to frequent immigration and emigration through wind and rain, and the entire population of microbes can be sampled. Observation of lists of endophytes suggests that each species of vascular plant is infected by at least two to four endophyte species that are specific to that plant species (Bills, 1996). According to Dreyfuss (1989) endophytic fungi represent one of the largest reservoirs of undescribed fungal species.

Ecology of endophytes:

Recent extensive surveys in a wide variety of plants indicate that endophytes are apparently ubiquitous, at least within plants growing in humid or mesic conditions (Petrini, 1986). Many of them have a rather reduced host range which in some cases

may be confined to a single plant species (Carroll and Carroll, 1978; Bacon *et al.*, 1986) whereas others are widespread (Petrini, 1986). The ecological roles of endophytic fungi are varied. They may be dormant saprobes (Chapela and Boddy, 1988), latent pathogens (Verhoeff, 1974; Carroll, 1986), mutualists (Clay *et al.*, 1985), antagonizing plant enemies (Latch *et al.*, 1985) or inducers of growth and competitive ability (Bose, 1956; Clay, 1986).

Results from a study on the species composition of endophytic fungi in healthy needles of Austrian pine (Pinus nigra Am.) investigated at eight locations in Slovenia showed that ecological factors have the most pronounced effects on species composition and on frequency of colonisation (Jurc et al., 1996). Eighty species of microfungi isolated from October 1994 and January 1995 when compared with analyses of macronutrients, sulphur and lead content of the needles, showed frequency of isolated fungi the lowest in the site with the highest amount of lead in needles. Similar study was carried out in poor growth/polluted and good growth/unpolluted stands of symptomless green needles of Sitka spruce and its infection level by endophytes Lophodermium piceae and Rhizospaera kalkhoffii by Magan (1996). In general, both the endophytes increased with the age of the needles but a higher isolation frequency of R. kalkhoffii was obtained from the polluted site rather than the unpolluted site. Complementary in vitro studies showed that R. kalkhoffii was more tolerant of elevated sulphur dioxide, lowered water availability and had a lower temperature optimum than Lophodermium piceae. Thus Magan (1996) tried to highlight the potential of endophytic fungi as possible bioindicators of tree vitality in polluted areas.

Several mutualistic roles of endophytic fungi have been demonstrated (Bacon et al., 1986; Carroll, 1988; Clay, 1991; Rowan, 1993; Wilson, 1993, Gange, 1995).

However, the exact nature of the interaction and the strength of the proposed mutualism, still remain an enigma, because testing hypotheses on the ecological role of these fungi is difficult as manipulating micro-organisms in the field or greenhouse is not easy. Wilson (1996) described a method which involves placing bags (composed of clear PVC tops and polyester netting bottoms and sides) over branches to protect newly emerging leaves from infective propagules. Leaves can then be infected with target fungi by repeated spraying of spore suspensions onto the leaves.

Although the role played by individual endophytes are well speculated, the significance of endophytic communities in plant ecology has been assessed very little. Espinosa-Garcia and Langenheim (1990), isolated leaf endophytic fungi from 1 to 12 year old leaves of mature trees and basal sprouts of coastal redwood Sequoia sempervirens in a redwood forest in Central California. The two most frequent species were Pleuroplaconema sp. and Cryptosporiopsis abietina. Species composition in leaves of progressing age in single branches revealed a patchy pattern of leaf colonisation without an obvious sequence of succession. This kind of patchiness may be important for plant interactions with herbivores and pathogens and could result from factors like microclimate, previous infections and changes in the host chemistry. The endophytic communities from leaves of trees and sprouts were generally similar, but differed in species richness and in the distribution of Pleuroplaconema sp. and Pestalotiopsis funerea. Principal component analysis based on endophytic frequency indicated closeness of trees and sprouts as groups, but clearly separated each tree from its sprout. Thus, the distribution patterns within and among plants, as well as posible consequences of their presence, reinforces the idea that not only single endophyte species but whole endophytic communities may be important for the plants that harbour them.

Endophytes as mutualists:

Most endophytes show limited growth within host tissues, in many cases such growth limitation probably results from activation of the same localized host defense mechanisms. Reserves of fixed C and nutrients in plants fluctuate seasonally. In perennial herbaceous plants and in trees, leaves represent a significant fraction of the plants accessible nutrient capital. Typically a portion of these reserves are mobilized and recovered prior to leaf abscission (Chapin and Kedrowski, 1983). Fungal domains in senescing deciduous leaves appear as green spots against a background of red, yellow or brown. These green islands develop through the elaboration of cytokinins and other metabolites by the asymptomatic endophytic fungi, metabolites which locally retard senescence and impede the mobilization of fixed carbon and other nutrients (Goodman *et al.*, 1986).

A number of endophytes have now been shown to function as antagonists to plant diseases and insect pests (Carroll, 1990). The protective effects of endophytes are apparently not confined to plant shoots and their targets may be broader than insect pests. Grass endophytes may be active against soil nematodes (Pederson *et al.*, 1988). A number of leaf and stem pathogens of crops have been reported to persist as endophytes in weeds (Hartman *et al.*, 1986; McClean and Roy, 1988). While such fungi cause little damage to their weed hosts, they may debilitate the commercially important hosts which are in competition with the weeds. Endophytic fungi are diverse and abundant in woody plants and are thought to increase resistance of host trees to invertebrate and vertebrate herbivores (Faeth and Hammon, 1997a).

Though endophytic fungi have their advantages, they may introduce genetic level changes on host plants. It is speculated that the DNA isolated and amplified from higher plants may originate from symbiotic microbes occupying the plant

tissues. A recent report on the phylogeny of *Picea* contained sequence data that on later analysis proved to originate from filamentous ascomycetes (Camacho *et al.*, 1997). Isolates of endophytic fungi from *Picea* foliage collected from the same location, when examined to identify the source of the contaminating DNA, showed a DNA sequence originally attributed to *Picea engelmannii* as that of *Hormonema dematioides*, an ubiquitous foliar endophyte of conifers.

Economic importance of endophytic fungi:

Following the discovery of taxol from the endophytic fungus, *Taxomyces andreanae*, originally isolated from *Taxus brevifolia*, other fungi are also screened for potential drugs. In a recent study Pulici *et al.*, (1997) found that two strains of *Pestalotiopsis* spp., endophytic fungi of *Taxus brevifolia*, produced several new sesquiterpenes including three caryophyllenes, and pestalotiopsin A, B and C.

Substrate utilization studies conducted with fungal endophytes by Carroll and Petrini (1983), Sieber-Canavesi et al. (1991) and White et al. (1991) have conclusively demonstrated that most endophytes are able to utilize, at least in vitro, most substrates present on the surfaces or in the cell wall of the host. Most of the endophytes investigated are able to utilize xylan and pectin and produce non-specific peroxidases and laccases. Production of extracellular cellulases and hemicellulases other than xylanases are widespread but usually limited to organisms derived from selected hosts or even host tissues. However the utilization of starch is limited to small number of endophytes (Petrini et al., 1991). Production of both pectin and polygalacturonic acid degrading enzymes, responsible for the degradation of cell wall middle layer is also extremely widespread among endophytes (Petrini et al., 1992b).

Isolates of a given species derived from the same host were generally more homogenous with respect to their enzymatic activities (Leuchtmann et al., 1992).

The production of enzymes and secondary metabolites in endophytes is closely related to their ecological significance. The secretion of extracellular enzymes needed for cell wall degradation supports the hypothesis that fungal endophytes represent a group of organisms specialized to live within plant tissues (Carroll, 1988). The general tolerance of endophytes to phenolic compounds (Carroll and Petrini, 1983) and the differential reactions shown by certain redwood endophytes against terpenoids produced by their host (Espinosa-Garcia and Langenheim, 1991a, 1991b) suggests the potential ability of host-specific endophytic fungi to cope with compounds produced by the pathogens.

Production of secondary metabolites by endophytes:

Carroll (1986) presumed that conifer needle endophytes might produce toxins that effect defoliating insects. Larry et al, (1992) screened fungal strains for metabolites toxic to spruce budworm larvae. Five percent of the strains produced extractable compounds that resulted in mortality or reduced rate of development in the larvae. Fermentations of three strains yielded relatively potent extracts. This is said to be the first report of the identification of toxins from fungal endophytes of woody plants. Polishook et al., (1993) reported the isolation and antibiotic activity of Preussomerin D from the endophytic fungus Hormonema dematioides recovered from living plant tissue of a coniferous tree.

Endophytes in other plants and plant parts:

Fungal endophytes were isolated from the pinnules, leaf vein, rachis and rhizome of spring and autumn plants of Pteridium aquilinum in Devon, U.K. (Petrini et al., 1992a). Barrow et al. (1997) tried to determine the nature and incidence of root endophytes on fourwing saltbrush, Atriplex canescens. They found that the root cortex cells in arid rangelands of Southwestern United States were regularly colonized with three types of endophytic fungi: The widespread occurrence of these non-destructive fungal associations with plants implied that they have an important role in plant survival in arid environments. Fisher and Petrini (1989, 1990) studied the fungi that inhabit the bark and the xylem of mature roots of Alnus glutinosa and found that alder roots are colonised by a comparitively large and diverse community of fungal endophytes. They isolated nearly 40 species of endophytic fungi from long roots of mature trees of Pinus sylvestris. Most species were predominantly bark colonisers, some able to penetrate deep into the host tissue. They suggested that root endophyte communities may not be host specific and are probably influenced by the environment. Aquatic hyphomycetes were isolated from living root tissues of spruce, birch and maple in a woodland stream of Nova Scotia (Sridhar and Barlocher, 1992). The results suggested that roots of different species may be colonized by different fungal species.

Enzyme activity of fungi:

In order to grow, fungi require sources of C, N, a supply of energy and certain essential nutrients such as potassium and phosphorus. Supplies of nitrogen may be obtained from proteins and other organic sources or from simple inorganic substances such as nitrates and ammonium salts. Energy and most of the carbon required for

growth, however, are obtained by fungi directly from living organisms or indirectly from their waste and dead tissue. The latter two provide a great range of different substrata for the growth of saprophytic fungi and include such diverse resources as animal faeces, cast-off skins, hooves, fur, feathers, nails, and horns of vertebrates, the exoskeletons of arthropods, dead bodies of animals, plant litter and the mycelia and fruit-bodies of other fungi (Kendrick, 1992).

Plant litter is composed of six main categories of chemical constituents: cellulose, hemicellulose, lignin, water-soluble sugars, amino acids and aliphatic acids, ether- and alcohol-soluble constituents including fats, oils, waxes, resins and many pigments and proteins. The break down of these constituents is effected as a sequence of specific reactions with the enzyme systems of specific organisms.

The decomposition of leaf litter follows an enymatic degradative sequence as follows. Initially, the phylloplane fungi attack the easily decomposable sugars exuded from the leaf surface or released by aphids and other insects from sub-cuticular tissues. Melezitose, glucose and fructose were identified as the main organic constituents in the through fall from the canopy of a *Quercus* woodland. As the leaf becomes senescent, the phylloplane fungi which individually or in combination possessing cutinase, pectinase and cellulase, penetrate the cuticle, attack the middle lamella and degrade the cell walls (Dickinson and Pugh, 1974). When species diversity is richest, this is likely to correspond with the higher rate of decomposition, greater genetic diversity and greater the enzyme diversity (Dix and Webster, 1995).

Ecological adaptation is achieved through a range of enzymes produced and the multiple forms of individual enzymes. The full range of these and hence the substrates that can be utilized depends upon species. Extracellular enzymes are extremely stable glycoproteins that operate in the fluids of the substratum. Enzymes may diffuse through the substratum but, if pore sizes are limiting, enzymes will not be able to move into the substratum and reactions will then be restricted to interfaces between the substratum and the penetrating hyphae (Dix and Webster, 1995). Enzymes have applications in many fields, including organic synthesis, clinical analysis, pharmaceuticals, detergents, food production and fermentation. The application of enzymes to organic synthesis is currently attracting more and more attention. The discovery of new microbial enzymes through extensive and persistent screening will open new, simple routes for synthetic processes and consequently, new ways to solve human problems (Ogawa and Shimizu, 1999).

Hydrolysis of starch:

Starch is the commonest of food reserves in plants, and fungi, with the notable exception of most yeasts, produce amylases which catalyse starch hydrolysis. Chemically starch is made up of two polymers of glucose: amylose and amylopectin. These are present in varying proportions according to plant species but invariably amylopectin is in the greater amount, and is usually about 75-85% of most starches. Both polymers consist of chains of glucose molecules linked by α 1-4 glucosidic linkages but an important difference is that amylopectin is highly branched and carries side-chains which are linked to the main chain through α 1-6 bonding.

The starch-hydrolysing enzymes and their distribution in microorganisms have been described by Fogarty and Kelly (1979). α -amylase is the commonest starch-hydrolysing extracellular enzymes found in fungi. Fungi also produce extracellular amyloglucosidase (glucoamylase), an enzyme which seems to be exclusive to fungi. α -amylase hydrolyses both amylopectin and amylose to maltose and higher molecular

weight fractions, by-passing α -1-6 linkages and randomly cleaving chains in the fashion of an endozyme. Amyloglucosidase hydrolyses α -1-4 and α -1-6 glucose residues to glucose, working on the ends of chains in the manner of an exoenzyme and is also capable of hydrolysing amylopectin, amylose and glycogen almost completely to glucose. Since α -amylase cannot hydrolyse α -1-6 linkages it cannot attack the branch points in amylopectin; thus in fungi which produce no amyloglucosidase, high molecular weight dextrins tend to accumulate when starch is hydrolysed (Fogarty and Kelly, 1979). All the maltose produced by the hydrolysis of starch is finally split into two glucose molecules by the catalytic action of intracellular α -glucosidase.

Degradation of cellulose:

Cellulose is the most abundant substance in plant litter and as a major constituent of all the layers of plant cellwalls it forms about 30-40% of the dry weight of wood and can be as high as 45% of the dry weight of cereal straw.

Cellulose is a straight-chain β -1-4 glucan polymer containing as many as 10,000 glucose molecules linked together by the removal of water from two hydroxyl groups. Glucan chains join to form microfibrils, bundles of which run in the matrix of the plant cell wall as strengthening components. Each microfibril consists of about 40-100 glucan chains linked together by hydrogen bonding between adjacent hydroxyl groups.

In parts of the microfibril the glucan chains are regularly arranged in a parallel fashion forming cellulose with crystalline characteristics. Crystalline cellulose is the more resistant to decay, possibly because the close packing of the molecules prevents the penetration of microbial enzymes. Hydrolysis of cellulose is catalysed by an

enzyme complex called cellulase that consists of a number of extracellular β -1-4 glucanases, some of which are endohydrolases randomly disrupting linkages throughout β 1-4 glucan chains, producing glucose, cellobiose and high molecular weight fractions, while others are exohydrolases or β 1-4 cellobiohydrolases, which act only on the ends of β 1-4 glucan chains releasing the disaccharide cellobiose (Halliwell, 1979). Glycohydrolases that release single glucose units from glucan chains are also part of the cellulase complex of some microorganisms. The decomposition of cellulose is finally completed by the transformation of trisaccharides and disaccharides to glucose by the action of β 1-4 glucosidases within the hyphae.

All wood-rotting fungi degrade cellulose as do apparently many microfungi from soil and litter as measured by their ability to hydrolyse carboxymethyl-cellulose and pure cellulose in the laboratory (Domsch and Gams, 1969; Flanagan, 1981). However, in nature cellulolytic activity depends upon a number of substratum-related factors, notably pH and mineral composition. The ability to hydrolyse cellulose is very variable. Some fungi have very low rates of utilization and others are unable to degrade cellulose at all.

The ability to decompose cellulose (or other plant polymers) has been used to classify fungi into several substrate-related ecological groups (Garrett, 1966). Theodorou *et al.* (1980) observed cellulase and β-glucosidase activities 50 h after inoculation of *Trichoderma reesei* in an artificially structured ecosystem, simulating soil conditions like leaching and the attachment of microorganisms to a solid substrate and the activity was still present in the effluent collected 300 h later.

An extensive review of the ecology of microbial cellulose degradation has been produced by Ljungdahl and Eriksson (1985).

Hydrolysis of Pectin:

Pectin is a polyuronide of plant origin and is of variable composition depending on the source. Pectin occurs chiefly in the middle lamella (intracellular layer) of plant tissue and may be looked on as the cementing material lending rigidity to the tissue. Many fungi, including well known plant pathogens, secrete enzymes which solubilize by hydrolysis the pectin in situ causing the softening characteristics of rotting. In the plant, pectin exists in the form of a labile combination either with cellulose, hemicellulose or other material known as protopectin. The enzyme complex besides protopectinase also consists of pectase and pectinase. Pectase, is an esterase (pectinesterase) which hydrolyses the methoxy groups off from the esterified carboxyl groups of the galacturonic acid residues in the soluble pectin molecule. Methyl alcohol results and in the presence of calcium ion, the soluble pectin is converted into a gel. Pectase action is a necessary prerequisite for pectinase action, for only deesterified pectin is attacked by the latter enzyme. Though these two enzymes are distinct and separable, in virtually every case they are produced together by fungi attacking pectin. Pectinase is the enzyme responsible for the complete rupture of the polymerized pectin molecule into its structural components. This enzyme is extremely widespread in fungi both, parasites and saprophytes (Foster, 1949; Osagie and Obuekwe, 1991).

Pectinases have applications in the food industry; they also play an important role in the degradation of cell wall material by plant pathogens and have been associated with fruit development, ripening and cell wall extention (Fogarty and Kelly, 1993; Ward and Moo-Young, 1989). Aguilar & Huitron, (1993), found that intact conidia of *Aspergillus* sp. were able to degrade pectin *in vitro* even when

protein synthesis was inhibited, thus indicating the presence of cell bound pectinases.

They also found an exo-pectinase, present in the mycelium.

Hydrolysis of Protein:

Proteins are the most abundant nitrogen-containing constituent of living organisms. Soluble proteins, of about 30 amino acids or less in chain length, can pass through hyphal walls; insoluble proteins are hydrolysed externally before utilized by fungi.

Most fungi have extracellular proteolytic activity against proteins over a range of environmental conditions. Peptide endohydrolases (proteases) cleave internal peptide bonds, releasing soluble peptides. These when taken into the hyphae can be degraded to their component amino acids by a range of different peptidases. Four broad classes of proteinases have been detected in fungal cultures, serine; aspartic; cysteine and metallo-proteinases. Multiple forms of serine and aspartic proteinases appear to be the proteinases most widely produced by fungi (North, 1982). Fungal proteinases have a low substrate specificity and are very durable under extreme environmental conditions.

Degradation of Lignin:

A significant proportion of the carbon in plants is in the form of complex aromatic polymers, such as tannin, lignin and related phenolics. Lignin is most abundant in woody plants where it accounts for up to about 30% of the carbon content, providing rigidity and resistance to biological attack. Microorganisms in soil ultimately oxidize these compounds to carbon dioxide and water. The essence of this

process is that in the final stages of degradation, fission of the benzene ring must occur to produce straight-chain aliphatic substances which can be completely respired.

Certain fungi, mostly basidiomycetes, are able to extensively biodegrade the lignin; white-rot fungi can mineralize lignin, whereas brown-rot fungi merely modify lignin while removing the carbohydrates in wood. Several oxidative and reductive extracellular enzymes (lignin peroxidase, manganese peroxidase, laccase, and cellobiose: quinone oxidoreductase) have been isolated from lignolytic fungi; the role of these enzymes in lignin biodegradation is being intensively studied (Reid, 1995; Zhao et al., 1996). The dissimilation of lignin by fungi can be conveniently thought of as occurring by three mechanisms: (i) depolymerization by cleavage of bonds within the polymer; (ii) removal and modification of side-chains with substitution on benzene rings; and (iii) fission by ring-splitting enzymes to convert aromatic nuclei into respirable aliphatic compounds. About 15 separate enzymes are required for the complete oxidation of lignin polymers (Tuor et al., 1995).

The economic consequences of lignin biodegradation include wood decay and the biogeochemical cycling of woody biomass, degrade a variety of pollutants in wastewaters and soils, to increase the digestibility of lignocellulosics, and possibly to bioconvert lignins to higher value products (Reid, 1995, Youn *et al.*, 1995; Raghukumar *et al.*, 1999).

The enzyme laccase is a copper-containing oxidase; it does not require peroxide. Like Mn peroxidase, it normally oxidises only those lignin compounds with a free phenolic group, forming phenoxy radicals. However, in the presence of the artificial substrate 2,2'-azinobis (3-ethylbenzthiazoline-5-sulphonate) (ABTS), laccase can also oxidise certain non-phenolic compounds, probably by hydrogen abstraction from benzyl carbons. ABTS also enhances the ability of laccase to degrade the

residual lignin in Kraft pulps; other synthetic mediators reportedly have a similar effect. Laccase is not produced by all white-rot fungi (Kirk and Kelman, 1965; Setliff and Eudy, 1980) and many microfungi from soil and litter that cannot degrade lignin produce abundant laccase (Dix, 1979).

Fungal laccases have been implicated in sporulation, rhizomorph formation, pathogenesis and formation of fruity bodies and lignin degradation (Thurston, 1994; Bourbonnais and Paice, 1990; 1992; Yaropolov *et al.*, 1994; Heinfling *et al.*, 1998). Thus laccases appear to have a significant role in fungal biology (Dittmer *et al.*, 1997) and is widely distributed in fungi found on decaying lignocellulosic materials in the marine environment (Raghukumar *et a.l.*, 1994). Li *et al.* (1999), compared the ability of four different fungal laccases for the oxidation of lignin model compounds in a laccase mediator system. They have also suggested the criteria for better laccase utility and more effective laccase-mediator systems for pulp bleaching.

Hydrolysis of xylan:

D-xylans are the major components found in the hemicellulosic fraction in the cell walls of higher plants (Monti *et al.*, 1991). Natural xylans are heterogenous polysaccharides consisting of a backbone chain of β-1,4-linked D-xylopyranosyl residues and side chains of different substituents. The complete hydrolysis requires the action of several enzymes, probably analogous to the synergistic enzyme action involved in crystalline cellulose degradation (Kluepfel *et al.*, 1990). Xylanases are produced by hemicellulose-degrading fungi (Dekker and Richards, 1976). Common microfungi may degrade xylan more actively than carboxymethyl-cellulose or pectin (Domsch and Gams, 1969). The xylanase complex is known to consist of four endohydrolases, two capable of attacking branch points and branches, reducing the

size of side-chains, and two that only reduce the size of the main chain (Reilly, 1981). Endoxylanases randomly cleave the β -1,4 bonds in the polyxylose backbone, yielding oligosaccharides of varied chain lengths. β -Xylosidase activity generates D-xylose from both short chain oligosaccharides and xylobiose (Bachmann and McCarthy, 1989; Huang *et al.*, 1991; Alconada and Martinez, 1994). The action of exoxylanases is less frequent (Kluepfel *et al.*, 1990) although it is not clear whether this exoenzyme is a separate entity from the β -xylosidase (Hayashida *et al.*, 1988; Puls and Poutanem, 1989).

CHAPTER III MATERIALS AND METHODS

The samples of microfungi associated with two experimental plants, *Ficus benghalensis* and *Carissa congesta*, were simultaneously gathered at monthly intervals during January 1997 to December 1999 from two places in Goa, viz. (i) the Goa University campus at Taleigao Plateau located in North Goa and (ii) Senaulim at Verna in South Goa. These places are separated by about 25 kms (Fig. 3.1).

The study site:

- 1. The Goa University campus (73° 50'N and 15° 27'E), an area of over 170 hectares, is an east-west extending table-top plateau, bordered on the southern side by the river Zuari and by valleys in the north and Arabian Sea along the West. The plateau, 50-60 meters above MSL, is mainly lateritic covered by a thin layer of soil and bears patchy scrub vegetation interspersed by trees. The site harbours a number of annual shrubs and a few perrenial trees. The dominant vegetation in the campus includes Alistonia scholaris, Bombax ceiba, Carissa congesta, Caryota urens, Ficus benghalensis, Memycylon umbellatum, Tilia sp., and Zizyphus jujuba. The laterite is thickest at the northern side extending to about 3-4 m, below which lies a thick laterite clay, followed by fresh philite rock. Although the annual rainfall is over 3000 mm, lack of permeable fissures on the surface and insufficient percolation time result in the total run off of rain-water to the sea (Sharma, 1994).
- 2. Senaulim in Verna covers an area of over 400 hectares (73° 56' and 15° 22'E) and is bordered by Majorda and Nuvem on the South, Utorda and Cansaulim on the East, Nagoa and Quelossim on the North and Loutolim on the West. The soil is lateritic with moist deciduous vegetation and shrub jungle. The dominant plants of the area include Calycopteris floribunda, Carissa congesta, Ficus benghalensis,

Mimusops elengi, Sterculia urens, Strychnos nux-vomica, Tectona grandis and Thespe&ia populnea.

The weather data of the collecting sites for the years 1997 and 1998 is given in Table 3.1 and Table 3.2 (Source: Goa Observatory, Govt of Goa)

The experimental plants:

The two plants used in this study, *Ficus benghalensis* L. a semi-deciduous tree of the family Moraceae, and *Carissa congesta* Wight, an evergreen thorny shrub of Apocynaceae, were the dominant plant species found growing in the study area. Sampling was always done from the same trees and shrubs.

Carissa congesta (syn. C. carandas L.). or the 'Karanda', a berry yielding plant, is native and common to much of India, Burma and dry areas of Ceylon. The karanda, also known in sanskrit as 'Karamarda', has attracted much interest as a source of wild fruit and as medicinal. C. congesta occurs in the humid secondary scrub jungles of north and south Goa and Uttara Kannada and Dakshina Kannada districts of Karnataka State (Daniels, 1991; 1993).

Carissa congesta is a straggerly climbing evergreen shrub, attaining a height of 3-5 m, sometimes ascending to the tops of tall trees. The plant is branched, with pairs of divaricate simple or branched thorns at the nodes and is rich with white gummy latex. The leaves are opposite, ovate, elliptic or obovate, obtuse or mucronate, 2.5-7.5 cm long, dark green, coriaceous, glossy on the upper surface, lighter green and dull on the underside. The fragrant flowers are white tubular with hairy lobes which are twisted to the left in the bud. The flowers are in terminal cymes, often tinged with pink, bloom and bear fruits between early February and early August. The fruit, a berry, is in clusters of 3-10, oblong or round, 1.25-2.5 cm long, with fairly thin

but tough, purplish-red skin turning dark-purple or nearly black when ripe; smooth, glossy; enclosing very acid to fairly sweet often bitter juicy pulp, exuding flecks of latex. The fruit has 2-8 small, flat, brown seeds. Fig. 3.3(a, b)

Analyses of the ripe fruits show the following values: calories, 745-755/kg; moisture, 83%; protein, 0.4-0.6%; fat, 2.5-4.6%; carbohydrate, 0.5-1%; sugar, 7.5-11.5%; fiber, 0.6-1.8%; ash, 0.7-0.8 %. Ascorbic acid content has been reported as 9 to 11 mg per 100 g (Morton, 1987).

Green, unripe, sour fruits are used in chutney and pickles in India. The sweeter types may be eaten raw but the more acid ones are best stewed with plenty of sugar. The ripe fruits are utilized in curries, tarts and puddings in Asia. Besides, the fruits have been employed as agents in tanning and dyeing. Karanda leaves have furnished fodder for the tusser silkworm. A paste of the pounded roots serves as fly repellent. The white or yellow wood is hard, smooth and useful for preparation of fashioning spoons, combs and household utensils (Morton, 1987; Wealth of India, 1950).

Ficus benghalensis L., the banyan tree, pollinated by wasps and dispersed by birds and mammals, offers evolutionary and ecological insights. Conserved as a sacred tree in India and elsewhere in Asia and Africa, the banyan with its peculiar architecture and mysterious life cycle has prompted people to respect this tree. The tree possesses hanging roots that reach the ground and support the canopy. The leaves are 6-15 cm long, coriaceous, ovate to elliptical with rounded or subcordate base; nerves on the leaves are usually prominent, looping near the margins and hairy beneath. Numerous minute gland-like dots are usually raised on the upper surface of the leaf. The inflorescence, a syconium, is round, oval, fleshy, stalkless, 0.5-4 cm in diam, situated in axil of the leaves on the branches, hanging roots or on the trunk.

Green when young and red when ripe, the inflorescence is generally mistaken for the fruit. It possesses an ostiolar opening at the end opposite to the stalk. Numerous tiny flowers cover the inner surface of the syconium. Fig. 3.2(a, b).

The banyan is known to dot the landscape. Due to its unique ability to absorb calcium and water from stones, the tree is often the sole coloniser of rocky cliffs as well as walls. Trees of all *Ficus* species together constitute 2 to 3% of the trees in forests. This rarity, coupled with their disproportionately high consumption value for the frugivores has lead ecologists to term fig as keystone resource species in the ecosystem (Utkarsh and Almeida, 1999).

The leaves of banyan are loped for fodder. A coarse rope is prepared from the bark and aerial roots of the tree. Various parts of the plant are considered medicinal. The milky juice is externally applied for pains and bruises and as an anodyme in rheumatism and lumbago. A remedy for toothache, the leaves are heated and applied as poultice to abcesses. The bark is astringent and is used in dysentry, diarrhoea and diabetes. The seeds are considered cooling and tonic. The wood of aerial roots, stronger and elastic, is used as tentpoles, cart yokes and carrying shafts (Wealth of India, 1956)

Type of experimental samples:

The following types of samples were gathered from both plants and subjected to fungal recovery processes.

- (a) Partially dried and decomposed fallen 'leaf litter' from beneath the tree/bush canopy.
- (b) Young and mature disease-free, 'fresh leaves', flower and fruits.
- (c) Top 'soil' beneath the litter bed.

(d) 'Air', about one meter above the ground level underneath the tree/bush canopy and within the tree ambiance.

The samples were stored in fresh paper or polythene bags in a refrigerator at 4°C until processed.

Processing of the samples:

The leaf-litter was subjected to two methods of observation.

Particle plating technique: In this method (Bills & Polishook, 1994), partially decayed Ficus benghalensis (5) and Carissa congesta (20) Fig. 3.4(b, d) leaves were thoroughly washed in tap water, cut into pieces with a disinfected scissor and wethomogenized in an electric blender for 4 min. The pulverized sample was filtered through two alcohol disinfected sieves (250 μm and 100 μm mesh size). The particles trapped in the 100μm filter (of size between 100-250 μm) were thoroughly washed in sterile distilled water. About 1g of particles were resuspended in 5 ml sterile water and 0.05ml of the dilution was plated out into 5 petri dishes each containing (MEA) Halt extract with antibiotics. The plates were incubated at 23°C.

A. Moist chamber incubation method:

Moist incubation chambers (Hawksworth, 1974) were prepared as follows. A thin layer of absorbant cotton superimposed by a circular piece of blotting paper (20cm diam.) was placed in the petri plate (20cm diam.) and drenched with distilled water. Five sterile microslides were placed on the surface of the filter paper. The plates were sterilized at 121°C and 15lbs/cm³ pressure in an autocalve. Partially decomposed whole or cut leaves of *F. benghalensis* and *C. congesta* were thoroughly

washed in tap water and incubated separately in the sterile moist petriplates at room temperature (Fig. 3.5)

B. Plating of soil sample:

The soil sample (1 g) collected from directly underneath the canopy of the test plants, was plated as described above in particle-plating technique. However, these were not pulverised in the homogeniser.

C. Trapping air-borne particle:

Five MEA petri plates with antibiotics were exposed to ambient air in the proximity of both plants, at a height of about 1m from the ground for 5 min. The plates were incubated at 23°C.

Processing for Endophytes:

Five randomly selected fresh disease-free mature and young leaves of the experimental plants (Fig. 3.4a, c) were gathered in polythene bags and processed immediately or maintained in refrigerator at 4°C. Leaves were thoroughly washed in tap water, followed by deionised water and surface sterilized first, 1minute in 70% ethanol, and second, 3 min in 4% sodium hypochlorite and third for 30 sec in 70% ethanol. Each surface sterilized leaf was thoroughly rinsed in sterile distilled water for 5mins and cut into 1 mm² bits with a sterile razor blade. Bits from young and old leaves were plated separately.

The leaf bits were considered in three lots: i) the basal, which included the petiole and basal part; ii) the middle portion of vein and lamina; and iii) the terminal, which included the terminal part of vein and leaf blade. About 9 pieces were aseptically placed equidistant in rows in a 10cm diam. petri plate containing antibiotic incorporated malt extract agar medium. The plates were incubated at 23°C. As the

fungal colonies appeared from the edges of the leaf bits and extended into the agar medium, (Fig. 3.6 a, b) these were aseptically transferred onto MEA slants without antibiotics (Petrini, 1986).

Fungal culture medium used:

Malt extract agar (MEA) [malt extract 5 g, agar 20 g, 1 L distilled water - prepared and made available in dehydrated form by HiMedia Pvt. Ltd, Mumbai], was the major medium used in this study. A cocktail of antibiotics (a) consisting of bacitracin 0.02 g, neomycin 0.02 g, penicillin G 0.02 g, polymixin 0.02 g, streptomycin 0.02 g and terramycin 0.04 g dissolved in 10ml of distilled water and (b) cyclosporin A 0.001 g dissolved in 1ml of methanol and membrane filtered were added to 1 L of MEA medium The medium for culture maintainance tubes consisted of 2% MEA without the antibiotic.

Observation of Fungi:

The fungal colonies appearing in the petridishes with leaf-litter, soil and air samples were counted as 'colony forming units' (CFU). On the 2nd, 8th and 15th day of incubation, five, ten and five colonies were randomly picked up and aseptically transferred to MEA petriplates where the medium was cut into sectors before they were finally transferred to agar slants (Fig. 3.7 a, b). The isolates were grouped into sporulating and nonsporulating forms. The cultural characters of the fungi in MEA were recorded as stipulated in Hawksworth *et al* (1995). Semipermanent slides of the sporulating structures such as sporangiophores and sporangia (Zygomycetes - Mucorales), conidiophores and conidia (Hyphomycetes), pycnidia, conidiogenous

cells and conidia (Coelomycetes) and ascocarp, asci and ascospores (Ascomycetes) from the colonies were prepared using lactophenol mountant (Hawksworth, 1974).

The sporulating structures are the diagnostic characters used in fungal identification. The sporulating isolates were identified and assigned to respective genera and species using standard taxonomic keys and monographs. Description of the fungi was written in diagnostic form. Illustrations of fungi were made using prism-type camera lucida apparatus fitted to the straight tube of a monocular microscope. Photomicrographs were taken using an automatic Minolta camera fitted to a Leitz microscope. The non-sporulating forms, the 'morphotypes', were graded in this work as 'dematiaceous' (coloured) and 'moniliaceous' (colourless) forms.

The description of species is based on definite specimens, materials or cultures of fungi, as prescribed in the International Code of Botanical Nomenclature (Hawksworth, 1974). The microslides containing fungal diagnostic structures, on which the descriptions based, are neatly sealed, labelled and housed in the herbarium of Goa University Fungus Culture Collection (GUFCC). The pure cultures of fungi recovered in this study are properly labelled, numbered and maintained in the collections of the Goa University Fungus Culture Collection (GUFCC). Holotypes are maintained for all new taxa. Where the new taxon is based on a live culture, dried and dead culture mats of these are preserved in herbarium sheets and maintained in the GUFCC, to satisfy the nomenclatural rules.

Enzyme Assays

A large number of fungal isolates were obtained from the substrates such as leaf-litter, fresh leaf, soil and from the ambient air of the experimental plants. Of these, 60 different individual fungi that appeared commonly on leaf-litter, fresh leaf

and soil samples and recovered in culture were randomly selected for screening experiments in order to detect the presence of degradative enzymes such as amylase, cellulase, protease, pectinase, ligninase, laccase and xylanase.

Discs of 5mm, from edge of 7day old colonies grown at room temperature on MEA were used as the source of inoculum for the following enzyme assays.

1. Amylolytic activity:

The ability to degrade starch was used as the criterion for determination of ability to produce amylolytic enzymes. The medium used contained malt extract 20g/l, agar 5g/l, soluble starch 0.2%. The mineral salts solution contained per litre: (NH₄)₂SO₄, 2g; KH₂PO₄, 4g; Na₂HPO₄, 6g; FeSO₄.7H₂O, 0.2g; CaCl₂, 1mg; H₃BO₃, 10µg; MnSO₄, 10µg; ZnSO₄, 70µg; CuSO₄, 50µg; MoO₃, 10µg; pH 6. After 5-7 days of incubation the plates were flooded with 1% iodine solution. The yellow zone around the colony in an otherwise blue medium indicated amylolytic activity (Hankin and Anagnostakis, 1975) (Fig. 3.8 b)

2. Cellulolytic activity:

The medium used contained malt extract 5g/l, agar 20g/l, carboxymethylcellulose 10g/l and mineral salt solution as described above. After 5-7days of incubation, the plates were flooded with 1% Congo red solution. A clear zone around the colony indicated cellulolytic activity (Carder, 1986). The plate was then flooded with 20ml of 1N NaCl to let the clearance zone stay for a longer period (Fig. 3.11)

3. Pectinolytic activity:

This medium contained 500ml mineral salts solution, pH 7, 1g yeast extract, 15g agar, 5g pectin and 500ml distilled water. This medium at pH 7 was used to detect pectate lyase production. Plates were incubated for 5-7 days and then flooded

with hexadecyltrimethylammoniumbromide. This reagent precipitates intact pectin in the medium and thus clear zones around a colony in an otherwise opaque medium indicated degradation of pectin (Hankin and Anagnostakis, 1975) (Fig.3.9a)

4. Proteolytic activity:

A medium that contained casein as the protein substrate as described by Cruickshank *et al.*, (1975) was used to detect the production of proteolytic enzymes. This medium consisted of casein 1g/l, malt extract 5g/l, agar 20g/l and mineral salt solution, pH 6. After incubation for 5-7 days, the plates were flooded with 1% mercuric chloride which enhanced the clearance zone around the colonies in a more or less opaque agar (Fig3.9 b)

5. Lignolytic activity:

The low nitrogen (LN) medium (Dass and Reddy, 1990) contained the following per liter: 10g glucose, 2g KH₂PO₄, 1.45g MgSO₄.7H₂O, 0.132g CaCl₂.2H₂O, 1mg thiamine hydrochloride, 0.5g tween 80, 1.2mM ammonium tartrate, 20mM sodium acetate (pH 4.5), and 0.4mM veratryl alcohol and 40ml of 0.02% Poly-R dye. The following trace elements were also added (per litre): 0.14g nitrilotriacetate, 0.07g NaCl, 0.007g FeSO₄.7H₂O, 0.033g MnSO₄, 0.013g CoCl₂.6H₂O, 0.007g ZnSO₄.7H₂O, 0.0011g H₃BO₃ and 0.0007g Na₂MoO₄.2H₂O. The ability to decolorize Poly-R is believed to be positively correlated with the production of ligninase. (Tien and Kirk, 1988) (Fig. 3.8 a).

6.Xylanase assay:

This medium contained 2% birchwood xylan as the sole carbon source supplemented to LN medium as described before. Glucose, Tween 80 and Veratryl alcohol were emitted from this medium. Xylan degrading fungal strains gave clearance zone around the colony in this medium (Haack, 1992).

7. Laccase plate assay:

Malt extract agar medium as mentioned before was supplemented with 2mM ABTS [2,2'-azino-bis-(3-ethylbenzothiazoline-6-sulfonic acid]. Green or greenish-blue color under and around the fungal colonies was considered a positive reaction. (Raghukumar *et al.*, 1994) (Fig. 3.10a, b)

8. Laccase enzyme assay:

Laccase positive cultures were grown in ME broth (10ml/100ml conical flasks) for 7days at room temperature. The mycelial mats were washed once with 100ml of sterile distilled water and macerated using sterile glass beads. The inoculum thus prepared was used for inoculating LN medium. Aliquots of culture filtrates were tested for laccase activity at an interval of 2 days up to 15 days. Culture supernatents were assayed for laccase activity by the method of Niku-Paavola *et al.* (1988). The laccase reaction mixture contained 2mM ABTS in 100mM glycine-HCl buffer at pH 3.0 in a final volume of 1.0ml. The reaction was monitored by measuring the change in absorbance at 405 nm using a spectrophotometer at room temperature. The enzyme units were expressed as nanokatals per litre using the following formula (Niku-Paavola *et al.*, 1988).

Activity = ΔA_{405} x total volume(in ml) x 10⁹ nkat/liter Σ (ABTS) x dt x sample volume(in ml)

where, Kat= mol/sec

E(ABTS) = 35000 (molar extinction coefficient of the radical cation ABTS)

Dt = seconds (=60)

Statistical Analyses of the Data:

The data on fungal isolates obtained during the 24 month study were subjected to '3- factor factorial completely randomized design analysis' defined in the 'Statistical Package for Social Sciences'. For endophytes, the '3² factor factorial completely randomized design analysis' of the SPSS was followed. To analyse the enzyme activity, 'Joining Tree Clustering' from "Statistica ver.5" was used.

Square root transformations of the values obtained during the study was subjected to Analysis of Variance (ANOVA) Test, after adding 0.5 (Samuels, 1989).

Fig. 3.1 Map showing location of sampling sites

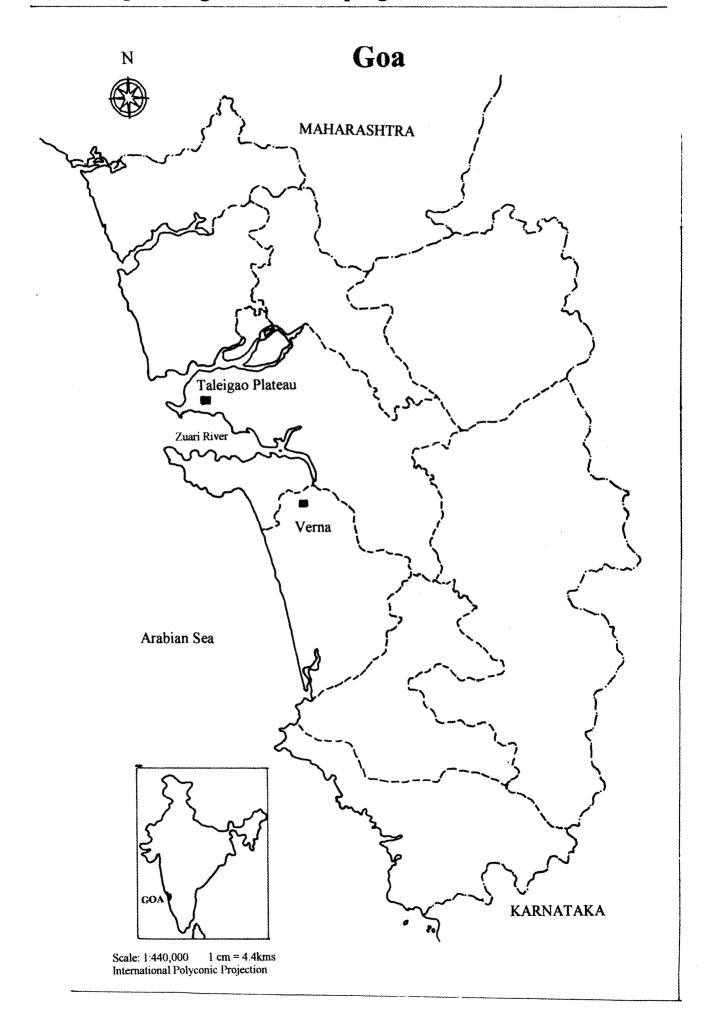
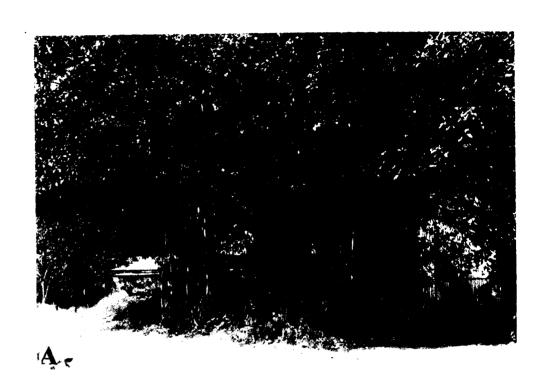


Table 3.1. Weather data for the year 1997:

MONTHS	RAIN	TEMPERATURE OC			AV. WIND	SUNSHIN	RELA	ATIVE
	FAL				SPEED FOR 24	Е	HUMIDITY	
	L				HRS. (KMPH)	(TOTAL	(%)	
	(MM)					HR)		
		MAX	MIN	MEAN			0830	1730
	·		٠				HRS	HRS
January	7.6	31.5	19.9	25.1	8.3	307.6	81	57
February	0.0	31.3	18.9	25.1	9.0	292.7	77	57
March	0.1	32.4	23.7	28.1	8.0	256.8	84	69
April	0.0	33.0	24.3	28.6	10.0	291.2	72	63
May	0.0	34.0	26.4	30.2	10.0	299.5	71	65
June	1057	31.2	25.0	28.1	12.0	170.0	86	75
July	1210	29.3	24.8	27.1	15.0	096.2	90	86
August	854	28.9	24.2	26.5	14.0	086.2	92	87
September	57	31.3	24.5	27.9	08.0	219.4	88	74
October	58	33.7	24.7	29.2	08.0	267.4	83	73
November	051	33.0	24.4	28.7	08.0	233.3	83	71
December	070	31.8	22.7	27.3	08.0	268.6	85	65

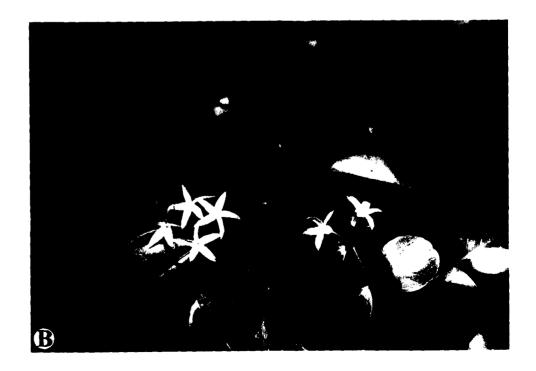
Table 3.2: Weather data for the year 1998

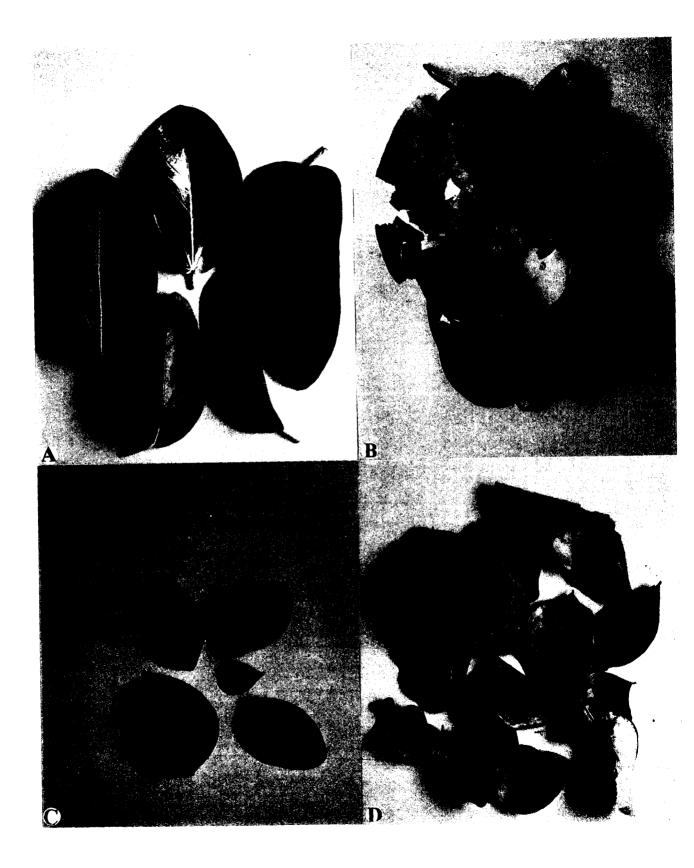
MONTHS	RAIN	ТЕМР	ERATI	URE OC	AV. WIND	SUNSHINE	RELA	TIVE
	FALL				SPEED FOR 24	(TOTAL	HUM	IDITY
	(MM)				HRS. (KMPH)	HR)	(%	(0)
		MAX	MIN	MEAN			0830	1730
							HRS	HRS
January	0.0	32.2	21.4	26.8	8.258	289.4	80	60
February	0.0	32.5	20.7	26.6	9.0	285.2	81	57
March	0.0	31.8	22.9	27.3	8.0	284.5	85	61
April	0.0	33.9	26.1	30.0	10.0	286.3	77	68
May	068.4	34.6	27.9	31.3	8.0	277.2	77	67
June	1069.7	31.0	25.6	28.3	6.3	132.0	90	82
July	751.0	29.8	25.1	27.4	14.2	121.5	91	87
August	549.8	30.0	24.8	27.4	9.4	148.6	96	87
September	427.3	29.5	24.3	26.9	21	108.3	96	85
October	167.5	30.8	24.2	27.5	7.3	217.8	92	82
November	44.9	32.2	22.3	27.2	6.9	275.1	85	73
December	3078.9	32.6	20.7	26.7	7.0	266.1	73	55

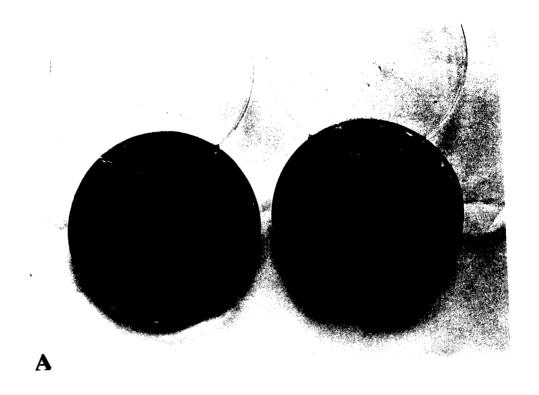


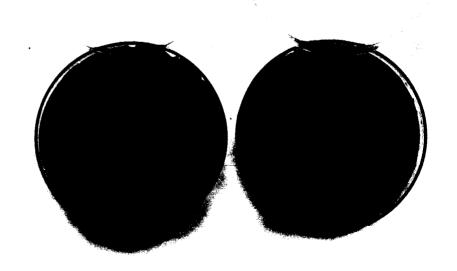




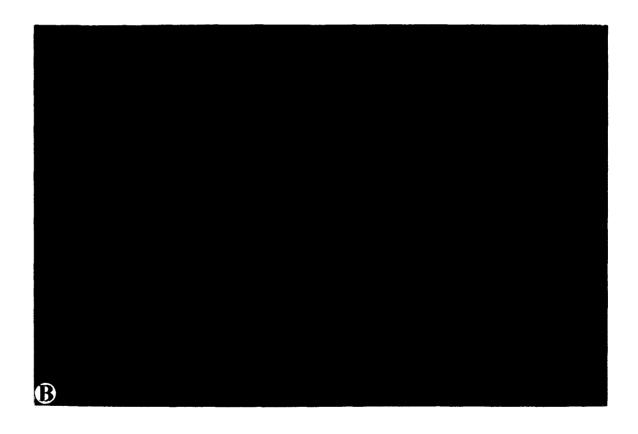


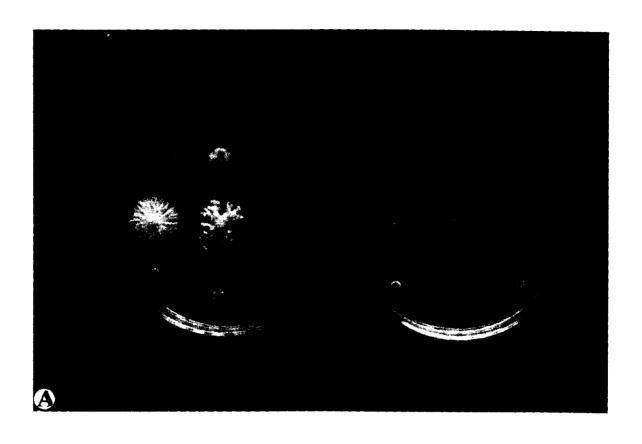




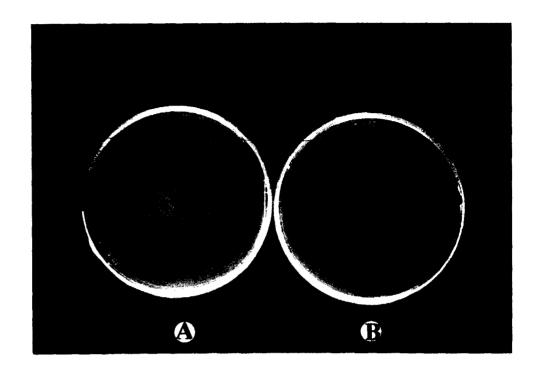


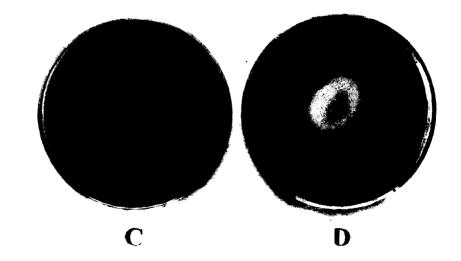


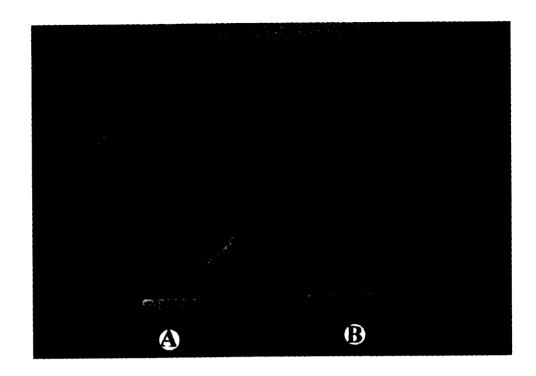


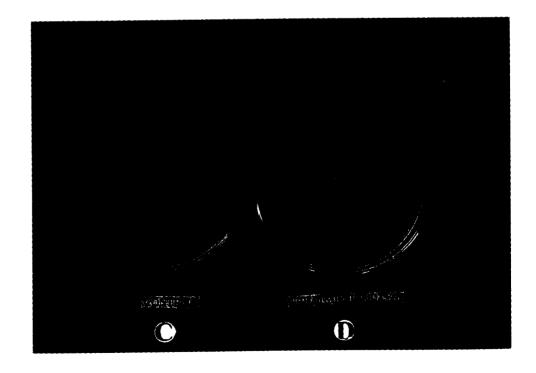


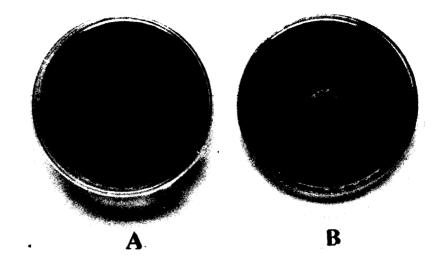


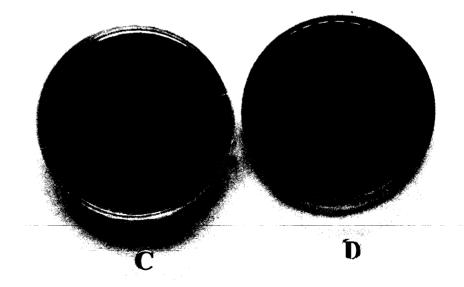


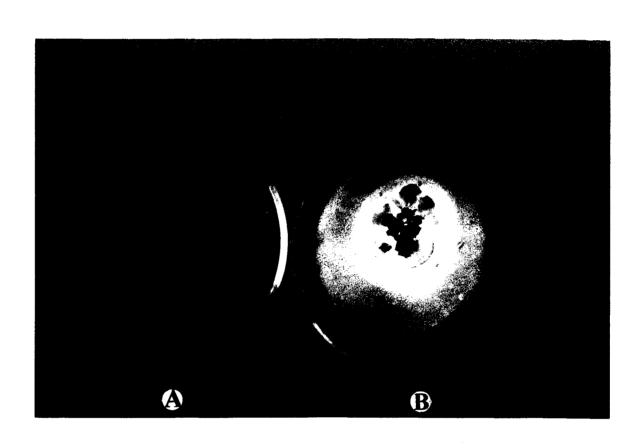












CHAPTER IV RESULTS AND DISCUSSION

RESULTS

PART 1. DIVERSITY OF THE FUNGI ISOLATED DURING THE STUDY:

Amongst the microfungi isolated from varied substrates and habitats and so far recorded in literature, maximum species diversity was on decomposing plant litter which included decaying leaves, twigs, logs, bark, roots, fruits, seeds, flowers and other plant produces (Hawksworth et al. 1995).

Several studies in the form of floristic investigations were carried out on the fungi of India and these resulted in the documentation of more than 10,000 species of litter fungi belonging to over 2200 genera (Sarboy et al. 1986, 1996; Hawksworth et al. 1995). The explorative studies however were fragmentary, both in space and time (Sarboy et al. 1986, 1996; Rao and De Hoog, 1986; Subramanian and Bhat, 1987; Bhat and Kendrick, 1993). Detailed mycofloristic investigations on specific plant substrata over a period of time were only a few (Vittal, 1973; Sudha, 1978; Rai, 1985; Dorai, 1988).

In the present study, investigation of the fungi occurring in association with two plant species, namely *Ficus benghalensis* and *Carissa congesta* and their neighbourhood, that is, the underneath soil and ambient air at a height of 1 M from ground level, was carried out at monthly intervals over a period of 24 months, i.e. Jan. 1997 to Dec. 1999, from two places, namely Taleigao Plateau and Verna in Goa, following moist-chamber and particle-plating recovery techniques.

The study resulted in the documentation of 228 species of fungi belonging to 120 genera which included Mucorales, Hyphomycetes, Coelomycetes and Ascomycetes (Table 4.1.). Of these, 177 taxa belonged to Hyphomycetes and are

accommodated in 81 genera. Sixteen hyphomycetous fungi were not assigned to any known taxa since these were unlike any known species of fungi and relevant literature was not available for identification. Four species belonging to 2 genera of Mucoraceous fungi encountered during the study are described. A total of 12 species belonging to 10 genera of Ascomycetes and 35 species belonging to 27 genera of Coelomycetes are only listed out.

A total of 121 isolates did not sporulate in culture or on the substrate and therefore they are not described in detail but recognised only as 'nonsporulating morphotypes'. These are recognised as morphotypes based on cultural characters such as colour, shape and size of the colony and presence or absence of exudates, but in the absence of any sporulating features, the taxonomy of these fungi remained undetermined.

The detailed description of the mucoraceous and hyphomycetous fungi are given below. Camera lucida illustrations of the diagnostic features, viz. the conidiophores and conidia in case of Hyphomycetes and the sporangiophores and sporangia in case of Mucorales of all the described taxa are appended in the text. The taxa belonging to the Coelomycetes and Ascomycetes listed out here are not described in detail.

The description of species is based on definite materials, specimens or cultures of fungi. The microslides containing the fungal diagnostic structures, on which the descriptions based, are neatly sealed, labelled and placed in the herbarium of Goa University Fungus Culture Collection (GUFCC). The pure cultures of fungi recovered in this study are properly labelled, numbered and maintained in the collections of Goa University Fungus Culture Collection (GUFCC). Holotypes are maintained for all new taxa. Where the new taxon is based on a live culture, dried and dead culture mats

of these are preserved in herbarium sheets and maintained in the GUFCC, to satisfy the nomenclatural rules.

The diagnoses of the new taxa described below are not latinized, though detailed descriptions are given and holotypes designated. The names are latinized. In the absence of latin diagnosis, as per Article 36 of the International Code of Botanical Nomenclature (Hawksworth, 1974), the novelty of these taxa at this stage remains provisional.

Table 4.1.1: The fungi that appeared on different plant parts of Ficus bengalensis and Carissa congesta:

Plants	Substrate	Muc	orales	i	Hyph	omycetes	Coele	omycetes	Asc	omycetes
		T	V		T	V	Т	V	Т	V
F. benghalensis	Leaf-litter	2	2		83	83	15	16	3	2
	Endophytes	0	2		26	20	12	5	2	2
C. congesta	Leaf-litter	0	1		66	77	15	14	3	4
	Endophytes	0	2		.20	27	8	9	3	3
	Soil	3	2		43	42	11	7	2	0
	Air	1	1		42	49	2	2	0	4

Table 4.1.2

Taxonomy:

The fungi recovered during the study are listed in Table 4.1.2 below:

1. Hyphomycetes:

Acronidiellina indica sp. nov.

Alternaria alternata

Arxiella terrestris

Aspergillus sp. 1-10

Bacillispora aquatica

Beltrania rhombica

Beltraniella buloloensis

Botryosporium diffusum

Brachysporiella gayana

Cercospora celosiae

Cercospora carriseae sp. nov.

Circinotrichum maculiforme

Cirrenalia indica sp. nov.

Chlamydomyces palmarum

Cladosporium sp. 1-9

Corynespora smithii

Curvularia brachyspora

Curvularia clavata

Curvularia crepinii

Curvularia lunata

Curvularia ovoidea

Curvularia pallescens

Curvularia richardiae

Curvularia senegalensis

Curvularia trifolii

Curvularia tritici

Cylindrocarpon ianthothele.

Cylindrocladium ilicicola

Cylindrocladium parvum

Dichtyochaeta assamica

Dicyma carisseae sp. nov.

Doratomyces indicus sp. nov.

Dreschlera tripogonis

Epicoccum purpurascens

Exosporium bryophylli

Fulvia fulva

Fusarium culmorum

Fusarium decemcellulare

Fusarium phragmitis

Fusarium semitectum

Fusarium solani

Fusarium tabacinum

Gliomastix murorum

Gliomastix uniseptata

Gonatobotryum bimorphospora sp. nov.

Helicoma dennisii

Helicomyces roseus

Helicosporium guianensis

Helicosporium lumbricoides

Helminthosporium dalbergiae

Helminthosporium mauritianum

Helminthosporium microsorum

Heteroconium solaninum

Humicola brevis

Humicola grisea

Humicola verrucosa

Hyaloscolecobasidium indicum Gen. et sp. nov.

Hyphodiscosia jaipurensis

Idriella fertilis

Idriella lunata

Idriella mucoidea sp. nov.

Idriella multiseptata sp. nov.

Idriella ramosa

Kumbhamaya indica Gen. et sp. nov.

Memnoniella echinata

Monodictys nigra

Monodictys lepraria

Monodictys fluctuata

Monodictys glauca

Monodictys cultura

Monodictys levis

Moorella ficusensis sp. nov.

Mycovellosiella perfoliata

Neocercosporella indica Gen. et sp. nov.

Neottiosporella triseti

Nigrospora endophytica sp. nov.

Nigrospora sphaerica

Nodulisporium honiaraense

Paracylindocladia indica Gen. et sp. nov.

Parahumicola endophytica Gen. et sp. nov.

Paecilomyces sp.1-3

Periconiella musae

Periconiella smilacis

Penicillium sp. 1-3

Periconia byssoides

Periconia lateralis

Periconia saraswatipurensis

Phaeotrichoconis crotalariae

Phialocephala carisseae sp. nov.

Phialocephala nephrospora sp. nov.

Phialocephala xalepensis

Phialomyces microsporus sp. nov.

Pithomyces chartarum

Pithomyces graminicola

Polyshema clavulata

Pseudobotrytis terrestris

Ramichloridium fasciculatum

Rhinocladiella cellaris

Septonema secedens

Sclerographium goanensis sp. nov.

Scolecobasidium constrictum

Scolecobasidium humicola

Scolecobasidium triangularis sp. nov.

Scolecobasidium variabile

Scolecobasidium verrucosum Scolecobasidium longisporum Scytalidium lignicola Septonema sp. Spadicoides indicus sp. nov. Spadicoides bina Speiropsis pedatospora Sporidesmium adscendens Sporidesmium altum Sporidesmium eupetoriicola Stachybotrys atra Stachybotrys nephrospora Stachylidium bicolor Sympodiella gracilispora Sympodiella multiseptata Tetraposporium sp. Trichobotrys effusa Trichoderma sp.1-2 Trichocladium cylindroclavatum Trichocladium opacum Tritirachium album Tritirachium orvzae Torula herbarum Vanakrippa parva Vermispora obclavata Veronea botryosa Volutella roseola Wiesneriomyces javanicus Zygosporium gibbum Zygosporium masoni Trichothecium roseum Undetermined Hyphomycete No.1 Undetermined Hyphomycete No.2 Undetermined Hyphomycete No.3 Undetermined Hyphomycete No.4 Undetermined Hyphomycete No.5 Undetermined Hyphomycete No.6 Undetermined Hyphomycete No.7 Undetermined Hyphomycete No.8 Undetermined Hyphomycete No.9 Undetermined Hyphomycete No.10 Undetermined Hyphomycete No.11 Undetermined Hyphomycete No.12 Undetermined Hyphomycete No.13 Undetermined Hyphomycete No.14 Undetermined Hyphomycete No.15

Mucorales:

Cunninghamella echinulata Mucor flavus Mucor javanicus Mucor silvaticus

Undetermined Hyphomycete No.16

Ascomycetes:

Chaetomium nigricolor Ames
Diatrype carisse De Not.
D. nigerrima Ell.& Ev.
Diatrypella indica Sathe & Sriniv.
Hypoxylon michelianum Ces.& de Not.
Sclerophaerum Berk. & Curt.
Guignardia sp.
Lophiostoma lecanthi Tilak
Nectria haematococca Wollenw.
N. pseudotrichia Berk. & Br.
Neocosmospora sp.
Xylaria sp.

Coelomycetes:

Ajrekarella polychaetriae Kamat & Kalani Ascochyta caricae Pat. Ascochytula sapindae Vyas & Purohit. Botryodiplodia theobromae Pat. Camarosporium indicum Rao & Rao Chaetomella cycadina Rao & Bahekar Coniochaeta fuckelii Sacc. Diplodia sp. Discosia atroceras (Tode ex Fr.) Fr. D. poonensis Kalani Gleosporium carissa Agarwal Hendersonia epileuca Wehmeyer H. palmigena Ponnappa Lasidiploidea theobromae (Pat.) Grif. & Maubl. Leptodothiorella creberrima Srinivasa Macrophomina sp. Monochaetia breviformis (Sacc.) Allesch. Neottiospora sp. Pestalotia paraguareiensis Maub. P. stictica Berk. & Curt Pestalotiopsis japonica (Syd.) Stey. P. palmarum (Cooke) Stey.

Phoma nebulosa (Pers.ex S.F. Gray) Berk. Phomopsis filiformis Mandal & Das Gupta P. herbarum West. Phyllosticta ficicola Pat. Robillarda sp.

Seimatosporium subulatum Sutton Septoria arcuata Ell. & Ev.

P. versicolor (Speg.) Stey.

S. pipulae Cooke
S. nilgiriensis Tilak
Stagonospora brideliae Thirum. & Narasimhan
Trichosprerma sp.
Vasudevella sporoboli Chona

The Hyphomycetes and Mucorales are described in alphabatical order with notes on taxonomy, cultural characters, morphology, substrate relationship and habitat from which they were recovered. The new taxa are briefly discussed. A total of 16 species of well sporulating hyphomycetous fungi described and illustrated in detail are not named. They are treated here as 'Undetermined hyphomycetes'.

1. Acremoniula sarcinellae (Pat. et Har.) Arnaud ex Deighton, 1969. Mycol.Pap.

118:3-5 (Fig. 1)

Colonies on MEA effuse, moderately brown, sparsely granular, 1.8 cm diam in 7 days, reverse brown. *Mycelium* light brown, interspersed with tiny groups of light brown spores, with smooth, thin-walled, septate, branched, hyaline, 1.5-2.2 μm wide hyphae. *Conidiophores* mononematous, smooth, unbranched, straight to flexuous, hyaline, cylindrical, 10-20 x 1.5-2.6 μm. *Conidiogenous cells* monoblastic, integrated, terminal or intercalary, determinate, cylindrical to flexuous. *Conidia* solitary, dry, simple, spherical to elongate, when elongate broadly rounded at the apex, narrowly truncate at the base, brown, 5.0-11 x 5.6-7.5 μm.

Specimen examined: On decaying leaf litter *C. congesta* and soil; Verna, and GU Campus, Taleigao, Goa; 17/10/1998; Miriam, J.; GUFCC No. 501 (live culture); 101 (Herb. slide). Isolated by particle plating.

Acremonium Link

Several isolates of the genus Acremonium Link, typified by A. alternatum Link (Carmichael et al. 1980), were recovered during the study. These were quite apart in their colony and morphological characteristics. Identification of these morphotypes into distinct species was difficult in the absence of appropriate literature to differentiate those several microscopic characters which distinguish the taxa at species

level. Therefore, besides a common generic diagnosis, the species in the genus *Acremonium* are referred here in numericals.

Colonies on MEA generally white, varied in colour and shape, visible after 7 days, granular, slimy. Mycelium thin, flat, with thin-walled, septate, smooth, hyaline, hyphae. Conidiophores mononematous, straight to flexuous, unbranched, cylindrical, smooth, hyaline, varied in shape and size. Conidiogenous cells monophialidic, integrated, terminal. Conidia solitary, simple, septate, hyaline, smooth, varied in size and shape.

Acremonium	Colony	Conidiophores	Conidia	Habitat/	GUFCC	Fig.
species	(diam. in cm)	(size in µm)	(size in µm)	Locality/	No.	No.
				Substrate		
1	irregular,	with vase-like	1-septate,	Leaf litter of		
	pinkish at the	swellings at the	4.3-4.7 x 1.5	both plants	502	2
	centre, 4.0	centre,				
		10-15 x 2.2-2.6				
2	regular,		in false	Leaf litter of		
	with concentric	21-33 x 1.5	chains,	both plants	503	3
	zonations, 1.6		ellipsoidal,			
			aseptate,			
			3-5.2 x 1.5			
3	regular, 1.5	branched,	curved,	Leaf litter		
		30-90 x 1.7-3	aseptate	of F.	504	4
			5.2-7.4 x 1.7-	benghalensis		
			3			
4	pink, 2.0	branched,	ellipsoidal,	Leaf litter		
		94-100 x 1.5-2.5	aseptate	of C.	505	5
			4.3-6.5 x 1.5-	congesta		

2.4

5	white, 2.0	in fascicles,	ellipsoidal,		
		unbranched,	aseptate	506	6
		31-50 x 1.6-2	2.5-3.7 x		
			1.5-1.63.		

6. Acrodictys goanensis Miriam et Bhat sp. nov.

(Fig. 7)

Colonies effuse, hairy, olive brown. Mycelium partly immersed, composed of smooth, branched 1.5-2 μm wide hyphae. Conidiophores mononematous, simple, moderately to dark brown, hyaline towards apex, 3-5-septate, smooth, 64-470 μm long, 10-12 μm wide 7.5-20 x 1.5-2.5 at the base, 2-4 μm wide at the tip. Conidiogenous cells terminal, monoblastic, elongated, hyaline, smooth, 10-20 x 4 μm, truncate at the tip after conidial secession. Conidia solitary, dry, cerebroid-like, with two rows of transverse and 4-8-rows of longitudinal septa, pale- to moderately dark brown, smooth, with basal cell protuberant, 24- 40 μm wide, 10-12 μm high.

Holotype: On leaf-litter of *F. benghalensis*; Verna, Goa; 18/9/1997; Miriam, J.; GUFCC 102 (Herb. slide); Isolated by moist incubation.

The genus Acrodictys M.B. Ellis, typified by A. bambusicola M.B. Ellis, has about 26 species so far recognized (Hawksworth et al. 1995). A. goanensis is unique in the genus with its hyaline conidiogenous cells and cerebroid-like conidia.

Acronidiellina indica Miriam et Bhat sp. nov.

(Fig. 8)

Colonies on MEA effuse, olive green, convex, irregular, 1 cm diam in 7 days, reverse olive green. Mycelium moderate, greyish, with slightly verruculose, thinwalled, septate, branched, light brown, 1.7-3 µm wide hyphae. Conidiophores mononematous, straight to flexuous, geniculate, unbranched, smooth, dark brown,

decreasing in intensity towards the tip, upto 100 μ m long, 2.6-4.2 μ m wide above the base. *Conidiogenous cells* integrated, terminal, polytretic, sympodial, with cicatrized scars. *Conidia* solitary, elongate, cylindrical, with rounded ends, aseptate, rarely 1-septate, olivaceous brown, echinulate, 5-20 x 2-2.5 μ m.

Holotype: On decaying leaf litter *Ficus bengalensis*; Verna Goa; 17/10/1998; Miriam, J.; GUFCC No. 508 (live culture); 103 (Herb. slide). Isolated by particle plating.

Of the several species known in the genus *Acronidiellina* M.B. Ellis, typified by *A. loudetiae* M.B. Ellis, *A. indica* is the only species with cylindrical conidia. In other species of the genus, the conidia are ellipsoidal or clavate (Ellis, 1971, 1976).

Alternaria alternata (Fr.) Keissler, 1912. Beih. Bot. Zbl. 29: 434 (Fig. 9)

Colonies on MEA, dark green, irregular, granular, with faintly concentric zonations,

3.3 cm diam in 7 days. Mycelium white, interspersed with dark brown bunches of conidia, dark green, with smooth, thin-walled, septate, branched, hyaline, 1.5-2.5 μm wide hyphae. Conidiophores mononematous, straight to flexuous, unbranched, hyaline to pale golden brown, smooth, up to 50 μm long, 3-6 μm wide, with one to several scars. Conidiogenous cells polytretic, integrated, terminal or intercalary, determinate. Conidia catenate, obclavate, obpyriform, oval, often with a conical beak, mid- to dark brown, verruculose, with up to 5 transverse septa and several longitudinal oblique septa, 20-60 x 9-18 μm.

Specimen examined: On decaying leaf litter and fresh leaf of *C. congesta* and soil; Verna, and GU Campus, Taleigao, Goa; 17/10/1998; Miriam, J.; GUFCC No. 509 (live culture); 104 (Herb. slide). Isolated by particle plating and moist chamber incubation.

Colonies on MEA, regular, pinkish purple, flat with mycelium observed more clearly in centre, 3 cm diam in 7 days, reverse pinkish purple only at centre. Mycelium thin, centre, with thin-walled, septate, smooth, 1.0 μm wide hyphae. Conidiophores smooth, branched, straight, dark brown, 1.5-4 μm wide. Conidiogenous cells monophialidic to polyphialidic, integrated, determinate, cylindrical with serrated at the opening, left behind as cylindrical denticles, 2-7.6 x 1-6.5 μm. Conidia simple, l. shaped, wedge, solitary, smooth, hyaline, 1-septate, 6.5-13 x 2-3.2 μm.

Specimen examined: On fresh leaf and leaf-litter of *C. congesta and F. benghalensis*; Verna, and GU campus, Taleigao, Goa; 18/9/1998; Miriam, J.; GUFCC No.510 (Live culture); 105 (Herb. slide); Isolated by particle plating.

Aspergillus Micheli ex Fries: Micheli (Ref: Micheli, 1729. Nova Pl. Gen.: 212-213; Fries 1821. Syst. mycol., 1: 45; Carmichael et al. 1980)

Several species of the genus *Aspergillus* Micheli ex Fries: Micheli, typified by *A. glaucus* Link ex Gray, were isolated during the study. The species are recognised based on differences in their colony morphology, size, colour, exudates and other characters and conidiophore and conidial morphology and size. However, the specific nomenclature could not be ascertained for each of them in the absence of relevant literature. The generic diagnostic feature is given below. The differences at the specific level are enumerated in Table 4.2.

Colonies on MEA visible in 7 days, effuse, coloured, granular, reverse of the colony coloured. Mycelium thin, with septate, branched hyphae. Conidiophores mononematous, straight, smooth, hyaline, bearing a hyaline vesicle at the tip and

phialides in uni-, bi- and tri-series. *Conidiogenous cells* monophialidic, terminal, discrete, ampuliform, cylindrical, elongate, smooth, hyaline. The first formed phialide and later formed phialides vary in size. *Conidia* catenate, hyaline, spherical to elliptical, smooth or verrucose, with visible attachment between adjacent conidia.

Aspergillus	Colony	Conidiop	Vesicles	Phialides	Conidia	Habitat/	Fig. No.
Species	(in diam	hores	(in diam	(in µm)	(in diam μm	Substrat	
	cm)	(in μm)	μm)			į	
1.	Pale	650-1460	20-22,	Biseriate;	spherical,	Soil	11
	yellow,	x 6.5-9.5		5-7 x 2-4.5	smooth,		
ļ ļ	9.0	smooth,	i	1.8-2.5,	1.8-2.5, brown		
:		hyaline		brown			
2.	white, 2.7	80-330 x	11-13	Biseriate;	spherical,	Soil	12
	concentric	3.2-6.2		2.5-5.0 x	spinulose		
	zones	medium		2.8-4	3.75, brown		
		brown					
3.	creamish,	72-170 x	5-6.25	Biseriate,	Spherical,	Soil, air	13
<u> </u>	2.1cm,	3 -4		3.8-5 x 2.5	smooth,		
	concentric				2.5-3.8		
	zones						
4.	bright	192-864 x	7.5-10	Biseriate,	Spherical,	Soil,	14
	green, 1.9,	24-32,		2.5-5 x	spinulose,	air	
	concentric	smooth,		1.6-2.5	2.5-3.13		
	zones	hyaline					
5.	Yellow	12.5 x	2.6-3.0	Biseriate,	spherical,	Soil	15
	brown,	2.5-3.5,		2.5-6.9 x	spinulose,		
	4.6,	smooth,	•	2.5	2.5-2.7		
	concentric	hyaline					
	zones						·

		,					
6.	White, 2.0	2.5-15 x 1.25-2.5 smooth, pale brown	1.25-2.5	Uniseriate, 3.8-6.5 x2- 2.5	spherical, spinulose 2.5-3.2	Soil	16
7.	light	675-1400	30-70	Biseriate	spherical,	Soil	17
	green, 3.0	x 6.5-9.5		3.8-10 x	spinulose,		
		smooth,		3.8-9	3.2-5.0		
		hyaline					
8.	dark	30-57 x	5.2-7.4	Uniseriate	eliptical,	Soil	18
	green, 0.3	2.2-2.6		7-9 x 2.2-	smooth		
		smooth,		3.0	4.4-8.3		ļ
		pale					
		brown					
9.	Light	144-615 x	5.6-12.5	Biseriate	Spherical,	Soil	19
	green, 0.6	2.5-5		5-6.3 x	spinulose		
		smooth,		1.3-2.5	2.5-3.2		
		pale					
		brown					
10.	Creamish	165-250 x	3.8-6.3	Triseriate	spherical,	Soil	20
	white, 3.0	3.8		2.5-5 x	smooth		
		smooth,		1.3-2.5	1.6-2.5		
		hyaline					

Colonies on MEA effuse, granular, smooth in the margin, white, radial, with concentric zonations towards the edge, 1.5 cm diam. in 7 days; reverse white. Mycelium thin, interspersed with punctate conidial masses, with smooth, thin-walled, septate, branched, hyaline, 1.7-2 μm wide hyphae. Conidiophores mononematous, smooth, elongated, sometimes developing from a single point, branched, slightly broader at the base, tapering towards the tip, hyaline, 30-90 x 1.5-2 μm. Conidiogenous cells monophialidic, integrated, determinate, cylindrical. Conidia solitary, slimy, cylindrical with rounded ends, sometimes one end apically pointed, 1-3-septate, hyaline, smooth, 3.7-11 x 1.5 μm.

Specimen examined: On leaf litter of *C. congesta*; Taleigao, Goa; 13/6/1998; Miriam, J.; GUFCC No.521 (Live culture); 116(Herb. slide); Isolated by particle plating.

Beltrania rhombica O. Penzig, 1882. Nuovo G. bot. ital. 14: 72-75. (Fig. 22)

Colonies on MEA effuse, circular, regular, brown, 5 cm diam. in 7 days, reverse of the colony dark brown. *Mycelium* moderate, brown, with thin-walled, septate, smooth, 1.5 μm wide hyphae. *Conidiophores* mononematous, flexuous, brown, smooth, septate, branched, 15-175 x 2.5-6.5 μm. *Setae* simple unbranched, erect, thick-walled, septate, dark brown, 56-100 x 5-6.5 μm at the base, smooth, arising from a lobed, flat basal cell. *Conidiogenous cells* mononematous, integrated, terminal, polyblastic, sympodial, denticulate, denticles cylindrical; separating cells swollen, elongate to globose, smooth, very pale brown, 7.5-13.7 x 3.7-6.5 μm. *Conidia* solitary, biconic, appendiculate, the free end drawn into a tiny spine like structure, 0-septate, smooth, pale olive brown, with a distinct hyaline transverse band

immediately above the widest part, 22.5 –30 x 8.5-10 μ m; the appendage 10-11.5 μ m long, 1.5 μ m at its base.

Specimen examined: On decaying leaf litter and fresh leaf of *C. congesta* and soil; Verna, and GU Campus, Taleigao, Goa; 17/8/1998; Miriam, J.; GUFCC No. 522 (live culture); 117 (Herb. slide). Isolated by particle plating and moist chamber incubation.

Beltraniella buloloensis Matsushima, 1971. Microfungi of the Solomon Islands and Papua New Guinea: 7 (Fig.23)

Colonies on MEA effuse, olivaceous to dark brown, velvety, 2.5 cm diam. in 7 days; reverse of colony dark brown. *Mycelium* brown, with thin, smooth, septate, branched hyphae. *Conidiophores* mononematous, straight, unbranched, smooth, pale brown, arising at the base of the seta, 37.5-75 x 2.5-3.8 μm. *Setae* unbranched, erect, thick-walled, dark brown, 85-200 x 3-3.5 μm. *Conidiogenous cells* terminal, discrete, polyblastic, ampulliform, globose-subglobose, denticulate, light brown, 8-10 x 3-4.5 μm. *Conidia* solitary, acropleurogenous, 0-septate, with a transverse band just above the centre, obclavate, turbinate with the base drawn out to a fine point, hyaline, smooth, 20-25 x 5-6.5 μm. *Conidia* attached to the conidiogenous cells by separating cells which are hyaline, smooth, fusiform to limoniform, 12.5-13.8 x 2.5 μm.

Specimen examined: On decaying leaf litter and fresh leaves of *C. congesta* and on soil, Verna, Goa; 4/12/1997, Miriam, J.; GUFCC No. 523 (Live culture); 118 (Herb. slide). Isolated both by particle plating and moist chamber incubation of leaves.

Botryosporium diffusum Corda (Ref: Zhang and Kendrick, 1990. Acta Mycol. Sin. 9:31) (Fig.24)

Colonies effuse, extensive, cottony, white. Mycelium superficial, thin, with hyaline, septate, branched, smooth, 2.5 μm wide hyphae. Conidiophores mononematous, very long, cylindrical, 12.5-16 μm wide at the base, with several side branches developing on the main stipe, denticulate where the branches break off; denticles 1.5-3 μm wide; side branches swollen to a diamond shaped at the tip, smooth, hyaline, 47-82 x 3-5 μm, 9-10 μm wide at the broadest part of the diamond-shaped tip; the apex of the side branches bear numerous irregularly shaped, hyaline, lobed, 15.5-16 μm wide, fertile branches. Conidiogenous cells polyblastic, denticulate, discrete, present all over the lobe. Conidia dry, ellipsoidal, with pointed base, hyaline, smooth, 7.8-9.5 x 3.5-4 μm.

Specimen examined: On leaf-litter of *C. congesta*; Verna, Goa; 14/12/1998, Miriam, J.; GUFCC No. 119 (Herb. specimen/slide); Isolated by moist chamber incubation.

Brachysporiella gayana Batista, 1952. Bolm Secr. Agric. Ind. Com. Est. Pernambuco

19: 109

(Fig. 25)

Colonies effuse, dark brown. Mycelium partly immersed. Conidiophores mononematous, straight to flexuous, sometimes with short branches near the apex, brown to dark brown, smooth, up to 120 μm long 3-6 μm wide. Conidiogenous cells monoblastic, integrated, terminal, percurrent, cylindrical, doliform. Conidia solitary, acrogenous, simple, clavate, obpyriform, 2-3-pseudoseptate, medium to dark brown, smooth, cells unequally coloured and with dark bands at the septa, 17-34 x 7-14 μm; conidiogenous cells often remaining attached to the base of conidia when seceeded.

<u>Specimen examined</u>: On decaying leaf litter of *F.benghalensis*; GU Campus, Taleigao, Goa; 17/10/1998; Miriam, J.; GUFCC No. 120 (Herb. slide). Isolated by moist chamber incubation.

Colonies effuse, dark brown, stubs-like. Mycelium partly superficial, partly immersed. Conidiophores, mononematous, caespitose, straight to flexuous, geniculate, unbranched, dark brown, paler towards the apex, smooth, septate, up to 210 μm long, 3-4 μm wide. Conidiogenous cells integrated, terminal, polyblastic, sympodial, cylindrical, with cicatrized scars. Conidia solitary, acropleurogenous, simple, obclavate to elongate, colourless, 5-8-septate, smooth, pointed at the tip, truncate at the base, 46-95 μm long, 2-2.5 μm wide at the base, 1 μm wide at the tip.

Specimen examined: On decaying leaf litter and fresh leaf of *C. congesta* and soil; Verna, and GU Campus, Taleigao, Goa; 12/8/1998; Miriam, J.; GUFCC No. 524 (live culture); 121(Herb. slide). Isolated by particle plating and moist chamber incubation.

Cercospora carisseae Miriam et Bhat sp. nov. (Fig. 27)

Colonies effuse, dark brown, hairy. Mycelium partly superficial, partly immersed. Conidiophores mononematous, caespitose, erect, straight to flexuous, unbranched, dark brown, paler towards the apex, smooth, septate, up to 450 μm long, 3-5 μm wide. Conidiogenous cells integrated, terminal, polyblastic, sympodial, cylindrical, with cicatrized scars. Conidia solitary, acropleurogenous, simple, elongate, cylindrical, colourless, many-septate, smooth, up to 460 μm long, 2-4.5 μm wide at the base, 2-2.5 μm wide at the tip.

Holotype: On decaying leaf litter and fresh leaf of *C. congesta*; Verna, and GU Campus, Taleigao, Goa; 21/7/1998; Miriam, J.; GUFCC No. 525 (live culture); 122(Herb. slide). Isolated by particle plating and moist chamber incubation

The species so far in the genus *Cercospora* Fresenius, typified by *C. apii* Fresenius, are generally named based on the host specificity (Ellis, 1971, 1975). So far no *Cercospora* species has been described on *Carissa congesta* and therefore a new species is recognised in the genus.

Chlamydomyces palmarum (Cooke) Mason, 1928. Annotated List of Fungi received at the Imperial Bureau of Mycology, List 2(Fascicle 1): 37 (Fig. 30)

Colonies on MEA effuse, brown, 2.2 cm diam in 7 days, reverse brown. Mycelium light brown, with smooth thin-walled, septate, branched, hyaline, 1-1.5 μm wide. Conidiophores mononematous, straight to flexuous, unbranched, smooth, hyaline, septate upto 4.7-54.3 x 1.25 μm wide. Conidiogenous cells monoblastic, integrated, terminal. Conidia solitary, 1-pseudoseptate, ellipsoidal, with rounded apex, narrow and truncate at the base, light brown, cells unequally coloured and darker brown at edges, rough-walled, 11-15 μm long, 2.5-3.75 μm wide at the base and 7.5-11.5 μm at the top; conidiogenous cells often remain attached to the base of conidia.

Specimen examined: On decaying leaf litter of *C. congesta* and soil; Verna, and GU Campus, Taleigao, Goa; 12/10/1998; Miriam, J.; GUFCC No. 526 (live culture); 123 (Herb. slide). Isolated by particle plating.

Circinotrichum maculiforme C.G. Nees ex Per., 1822. Mycol. eur., 1:19 (Fig. 28)

Colonies effuse, dark brown to black, velvety. Mycelium partly superficial and partly immersed; setae present, simple, circinate, dark brown, paler at the apex, verrucose, septate, 145-200 µm long, 5-14 µm wide at the base, 3-6 µm wide in the middle tapering to 1-2 µm wide at the tip. Conidiophores prostrate, flexuous, irregularly branched, crowded at the base of the setae, subhyaline to pale brown,

smooth, 2 μ m wide. *Conidiogenous cells* erect, solitary, polyblastic, discrete, lageniform, pale brown, 5-6 μ m long, 2-4 μ m at middle, tapering to 1 μ m. *Conidia* solitary, dry, elongated, arranged in circles just below the apex of the conidiogenous cell, simple, 0-septate, colourless, 10-14 x 1-1.5 μ m.

Specimen examined: On leaf-litter of *F. benghalensis and C. congesta*; Verna, Goa; 18/9/1997; Miriam, J.; GUFCC No. 124 (Herb. slide); Isolated by moist chamber incubation.

Cirrenalia indica Miriam et Bhat sp.nov.

(Fig. 29)

Colonies on MEA effuse, moderately brown, granular, 2 cm diam in 7 days, reverse brown. Mycelium light brown, with smooth, thin-walled, septate, branched, pale brown, 1-2 μm wide hyphae. Conidiophores light brown, cylindrical, smooth, 4 -20 μm long, 1.5-2.5 μm wide. Conidiogenous cells monoblastic, integrated, terminal, determinate. Conidia solitary, dry, simple, elongated, curved, 1-2-septate, smooth, thick-walled, dark brown, slightly constricted at the septa, with terminal cell often larger than others, 14-27 μm long, 6.5-11.7 μm wide at the tip, 1.5-4.5 μm wide at the base.

Holotype: On decaying leaf litter of *C. congesta*; Verna, and GU Campus, Taleigao, Goa; 11/11/1998; Miriam, J.; GUFCC No. 527 (live culture); 125 (Herb. slide). Isolated by particle plating.

Several species of the genus *Cirrenalia* Meyers et Moore, typified by *C. macrocephala* (Kohlmeyer) Meyers & Moore, are known (Ellis, 1975) and all species except *C. donne* Sutton have marine affinity. *C. donne* was isolated from the bark of *Abies* in Canada. *Cirrenalia indica* differs from the known species in its conidium size and morphology and its terrestrial affinity.

Cladosporium Link (Ref: Ellis, 1976. More Dematiaceous Hyphomycetes, 325-344)

Several species of the genus *Cladosporium* Link have been recovered during the study. These are described here in a tabular form indicating the characteristic features along with a common generic diagnosis.

Colonies on MEA effuse, circular, green brown, velvety, granular, growth visible in 7 days, reverse of the colony coloured. Mycelium moderate, hyaline golden brown with thin walled, smooth, very dark septa, branched. Conidiophores mononematous, branched, moderately brown with tiny granular surface very faint, branched. Ramoconidia present, smooth, moderately brown. Conidiogenous cells polyblastic, integrated, terminal and intercalary, cylindrical, cicatrised scars prominent. Conidia catenate, ellipsoidal, with a distinct scar at its base, light brown, smooth, septate.

Cladosporium	Colony	Conidiopho	Ramoconidia	Conidia	Fig. No.			
sp.	Diam. in cm	res	in µm					
		in µm						
1		granular,	3-18x1.6-2.3	smooth, aseptate,	31			
olive green,		165-169 x		ellipsoidal,				
	frilled margin,	6-8		3 x 2-2.5.				
	3.6							
2	Dark green	Verrucose,	4-10x2.5-3	ellipsoidal to	32			
	brown, radial,	121-212 x		spherical,smooth				
	patches in	2-4		2.5-4 x 2-2.5				
	colony, 2.8							
3	Olive green,	Granular,	5-18x2-3	smooth, aseptate,	33			
	granular,	10-50 x 4-5		2-4				
	1.8							
4	Very dark	Verrucose,	Uniserriate,	smooth, aseptate,	34			
	green, single	12-19 x 1.3	3.8-8.8 x 1.6-2	1.3 x 2-2.5				
	concentric zone							
	just before the							
	periphery,							
	radial, 3							

5	Green yellow,	Verrucose, 5-100 x 1.3	Uniserriate, 1.3 x 2.5-7.5	1.4-x1.6	35
6	Green,	Verrucose, 19-200 x 1- 3	4-19x2.5-4	verrucose, aseptate, 2.5	36
7	Dusty olive green, Single concentric zone just before the periphery, 1.7	Verrucose, 19-25 x 3	0-2 septate, 6-16 x 2.3-3	verrucose, aseptate, 9 x 6	37
8	Dusty brown, 2.5	Unbranche d, 38-97 x 3	Verrucose, 4.7-18 x 2.3	spherical, verrucose, aseptate, dark brown deposition seen in patches,	38
9	Moderate brown, 2.1,	Branched, Smooth	spherical to ellipsoidal, 0-1-septate, smooth, 5-13.5 x 2.5- 4.7	ovate to ellipsoidal, aseptate, smooth, light brown, 4.4-5 x 2.5-3.7	39

Corynespora smithii (Berk. et Br.) M.B. Ellis, 1957. Mycol. Pap. 65: 3-6 (Fig. 40)

Colonies on MEA, effuse, with irregular margin, light pinkish brown, 4.8 cm diam in 7 days, velvety reverse of the colony brown. Mycelium moderate, greybrown, loose, raised over in the colony, 1.3-2 μm wide hyphae. Conidiophores mononematous, straight to flexuous, unbranched, smooth, light brown. 53-83 μm long and 1.5-2.2 μm wide Conidiogenous cells monoblastic, integrated, terminal, percurrent, cylindrical. Conidia solitary, dry acrogenous, simple, obclavate, light

brown to straw coloured, pseudoseptate, smooth, 28-58 μ m long, 3-5 μ m wide at its broadest part, 1.5-2.2 μ m wide at its truncate base.

Specimen examined: On leaf-litter of *C. congesta*; Verna, Goa; 18/9/1997; Miriam, J.; GUFCC No.537 (Live culture); 135 (Herb. slide); Isolated by particle plating.

Cunninghamella echinulata Thaxter (Ref: Gilman, 1957. A Manual of Soil Fungi, 66)

(Fig. 41)

Colonies on MEA irregular, white, 7.3 cm diam. in 7 days. Mycelium thin, transversely sparse, with thin walled, non-septate, smooth, hyaline, 6 μm wide hyphae. Sporangiophores solitary, branched, hyaline, smooth, up to 120 μm long, 4-5 μm wide. Sporangia spherical, smooth, hyaline, 13-15 μm diam. Spores spherical, hyaline, distinctly appendaged with long bristiles, 5.6- 10.8 μm diam.

Specimen examined: On leaf-litter and fruit of *F. benghalensis*; Verna, Goa; 13/5/1997; Miriam, J.; GUFCC No.538 (Live culture); 136 (Herb. slide); Isolated by particle plating.

Curvularia brachyspora Boedijn (Ref: Ellis, 1971, Dematiaceous Hyphomycetes: 454)

(Fig. 42)

Colonies on MEA, dirty brown with patches in between, granular, regular, concentric circles, reverse brown, 5.7cm diam. in 7 days. Mycelium moderate, dirty brown forming a soft mat, interspersed with dark brown tiny spore masses, with thin walled, smooth, septate, hyaline, 1.5-2.0μm wide hyphae. Conidiophores mononematous, straight to flexuous, nodose, light, smooth, septate, 100-180 x 3-3.5μm. Conidia solitary, simple, curved, 3-euseptate, smooth with scar, 15.6-27 x 6.2-12.5μm.

Specimen examined: On leaf-litter of *C. congesta*; Verna, Goa; 18/11/1997; Miriam, J.; GUFCC No.539 (Live culture); 137 (Herb. slide); Isolated by particle plating.

Curvularia clavata Jain, 1962. Tran. Br. mycol. Soc. 45: 542. (Fig. 43)

Colonies on MEA effuse, dark brown, in concentric zones, lighter in the centre becoming darker towards the periphery, 6 cm diam. in 7 days, reverse dark brown. Mycelium moderate, hyaline, interspersed with dark brown spores, with thin walled, smooth, septate, 2.35-3.2 μm wide hyphaae. Conidiophores mononematous, straight to flexuous, nodose, moderately brown, smooth, septate, 62-237 x 2.35-3.12μm. Conidiogenous cells polytretic, integrated, terminal, intercalary, sympodial, with cicatrized pores. Conidia solitary, simple, slightly curved at centre, ellipsoidal to clavate, 3-4-pseudoseptate, central three cells moderately brown, smooth, 25.7-31.2 x 11-14 μm.

Specimen examined: On leaf-litter of *C. congesta*; Verna, Goa; 8/9/1997; Miriam, J.; GUFCC No.540 (Live culture); 138 (Herb. slide); Isolated by particle plating.

Curvularia crepinii (Westend.) Boedijn (Ref: Ellis, 1971, Dematiaceous Hyphomycetes: 454) (Fig. 44)

Colonies on MEA dark brown with patches in between, granular, irregular, concentric circles, reverse of the colony brown with dark patches. 6.6cm diam. in 7 days. *Mycelium* thin, light brown, interspersed with dark brown spores, with thin walled, smooth, septate, hyaline, 1.5-2.0µm wide hyphae. *Conidiophores* mononematous, straight to flexuous, nodose, light brown, smooth, septate, 210 x 3-3.5µm. *Conidia* solitary, simple, curved at the top, 3-pseudoseptate, darker than moderate brown, smooth with scar outside, 23-40 x 13-20 µm.

Specimen examined: On leaf-litter of *C. congesta*; Verna, Goa; 21/6/1997; Miriam, J.; GUFCC No.541 (Live culture); 139 (Herb. slide); Isolated by particle plating.

Curvularia lunata (Wakker) Boedijn, 1933. Bull. Jard bot. Buitenz. 13: 127 (Fig.45)

Colonies on MEA, effuse, brown with patches in between, granular, irregular, concentric circles, reverse of the colony brown with dark patches. 6.6cm diam. in 7 days. *Mycelium* thin, brown, interspersed with dark brown spores, with thin walled, smooth, septate, hyaline, 1.25-2.0μm wide hyphae. *Conidiophores* mononematous, straight to flexuous, nodose, moderate brown, smooth, septate, 170-190 x 4.5-5μm. *Conidia* solitary, simple, curved, ends straight, 3-septate, true septate, second cell from the tip where the spore is curved is very dark brown, smooth with scar within, 25-27 x 9.4-12.5μm.

Specimen examined: On leaf-litter of *C. congesta*; Verna, Goa; 14/9/1997; Miriam, J.; GUFCC No.542 (Live culture); 140 (Herb. slide); Isolated by particle plating.

Curvularia ovoidea (Hiroe et Watan.) Muntanola (Ref: Ellis, 1971. Dematiaceous Hyphomycetes: 456) (Fig. 46)

Colonies on MEA dirty brown with patches in between, granular, irregular, concentric circles, reverse of the colony brown with dark patches. 6.2cm diam. in 7 days. *Mycelium* thin, light brown, interspersed with dark brown spores, with thin walled, smooth, septate, hyaline, 3.2µm wide hyphae. *Conidiophores* mononematous, straight to flexuous, nodose, moderate brown, smooth, septate, 3.12 x 63-103µm. *Conidia* solitary, simple, slightly curved at centre, ends straight, 1-euseptate, dark brown, smooth, with an scar outside, 9.4-12.5 x 4.7-6.4µm.

Specimen examined: On leaf-litter of *C. congesta* and *F. benghalensis*; Verna, Goa; 16/5/1997; Miriam, J.; GUFCC No.543 (Live culture); 141 (Herb. slide); Isolated by particle plating

Curvularia pallescens Boedijn, 1933. Bull. Jard bot. Buitenz. 13: 133 (Fig. 47)

Colonies on MEA dark brown with patches in between, granular, irregular, few concentric circles towards the periphery, 6.5 cm diam in 7 days, reverse green brown. Mycelium thin, white, interspersed with dark brown spores, with thin walled, smooth, septate, hyaline, 1.5-2.0μm wide hyphae. Conidiophores mononematous, straight to flexuous, nodose, light brown, smooth, septate, 143-150 x 6-7 μm. Conidia solitary, simple, slightly curved, 3-pseudoseptate, light brown, smooth with scar outside, 16-22 x 7-12.5μm.

Specimen examined: On leaf-litter of *C. congesta and soil*; Verna, Goa; 18/9/1997; Miriam, J.; GUFCC No.546 (Live culture); 131 (Herb. slide); Isolated by particle plating.

Curvularia richardiae Alcorn, 1971. Trans. Br. mycol. Soc. 56:155-157 (Fig. 48)

Colonies on MEA dark brown, irregular, 6.5cm diam. in 7 days, reverse brown. Mycelium moderate, interspersed with brown spores, with thin walled, smooth, septate, moderate brown, 1.5-2 μm wide hyphae. Conidiophores mononematous, straight to flexuous, nodose, moderately brown, smooth, septate, 23-24 x 4-5μm. Conidia solitary, simple, deeply curved, 3-euseptate, central two cells darker brown, smooth with scar within, 15.6-25 x 6.6-12.5μm.

Specimen examined: On leaf-litter of *C. congesta*; Verna, Goa; 22/1/1998; Miriam, J.; GUFCC No.545 (Live culture); 143 (Herb. slide); Isolated by particle plating

Curvularia senegalensis (Speg.) Subram. 1956. J. Indian Bot. Soc. 35: 466 (Fig. 49)

Colonies on MEA very dark green with patches in between, granular, irregular, few darker cicular concentric bands only towards the periphery, radial, 6.5cm diam. in 7 days, reverse light green. *Mycelium* thin, white, interspersed with dark brown spores, with thin walled, smooth, septate, hyaline, 2-4.5 μm wide hyphae. *Conidiophores* mononematous, straight to flexuous, nodose, light brown, brown at the tip, smooth, septate, 50-410 x 3-5 μm. *Conidia* solitary, simple, curved at centre, 3-4-pseudoseptate, brown, smooth with scar outside, 26.3-27.5 x 10-11.3μm.

Specimen examined: On leaf-litter of *C. congesta* and *F. benghalensis*; Verna, and GU campus, Goa; 11/9/1997; Miriam, J.; GUFCC No. 546 (Live culture); 144 (Herb. slide); Isolated by particle plating.

Curvularia trifolii (Kauffman) Boedijn, 1933. Bull. Jard bot. Buitenz. 13: 128

(Fig. 50)

Colonies on MEA, dirty brown with patches in between, granular, regular, concentric circles, reverse brown, 5.9cm diam. in 7 days. Mycelium moderate, interspersed with brown spore masses, with thin walled, smooth, septate, hyaline, 1.5-2.0μm wide hyphae. Conidiophores mononematous, straight to flexuous, nodose, light, smooth, septate, 179-180 x 2-4μm. Conidia solitary, simple, curved, 3-euseptate, central two cells darker with central septa with a very dark band, smooth with scar, 15.6-19 x 6.6-12.5μm.

Specimen examined: On leaf-litter of *C. congesta*; Verna, Goa; 18/9/1997; Miriam, J.; GUFCC No.547 (Live culture); 145 (Herb. slide); Isolated by particle plating and moist chamber incubation.

Colonies on MEA, very dark brown, almost black, regular, concentric circles, reverse of the colony dark brown, 5.9cm diam. in 7 days. Mycelium moderate, interspersed with brown spores, with thin walled, smooth, septate, hyaline, 1.5-2.0μm wide hyphae. Conidiophores mononematous, appear in bunches, aggregated, straight to flexuous, nodose, dark brown at the base, decreasing in color intensity towards the apex, smooth, septate, 150-180 x 3-5μm. Conidia solitary, simple, curved, 3-euseptate, central two cells moderately brown, smooth with scar outside, 15-20 x 7-10μm.

Specimen examined: On leaf-litter of *C. congesta* and *soil*; Verna, Goa; 12/5/1997; Miriam, J.; GUFCC No.548 (Live culture); 146 (Herb. slide); Isolated by particle plating

Cylindrocarpon ianthothele Wollenw. (Ref: Matsushima, 1975. Icones Fungurum A

Matsushima Lectorum: 45)

(Fig. 52)

Colonies on MEA, effuse, brown to reddish yellow, regular, 6.6 cm diam in 7 days, reverse brown. *Mycelium* thin, loose, white, interspersed by light greyish droplets, with thin, smooth, septate, branched, hyaline, 1.5 μm wide hyphae. *Conidiophores* mononematous, straight to flexuous, branched, hyaline, septate, smooth, 3.7-37.5 x 1.5 μm. *Conidiogenous cells* phialidic, integrated, terminal, determinate, cylindrical. *Conidia* simple, elongated, curved, with rounded ends, 4-septate, hyaline, smooth, 15-21.5 x 2.5-2.6 μm.

Specimen examined: On decaying leaf litter and fresh leaf of *C. congesta* and soil; Verna, and GU Campus, Taleigao, Goa; 17/10/1998; Miriam, J.; GUFCC No. 549 (live culture); 147 (Herb. slide). Isolated by particle plating and moist chamber incubation.

Colonies on MEA circular, with wavy margin, white to light brown, slimy, 5 cm diam on 7 days, reverse of the colony light brown. *Mycelium* thin, with hyaline, smooth, thin-walled, septate, branched, 1.5 μm wide hyphae. *Conidiophores* mononematous, flexuous, erect, smooth, branched, hyaline, thick-walled, 50-240 x 3-10 μm, terminating in a loosely branched fertile head and a single sterile erect, smooth, hyaline hypha with a swollen pointed tip 160-173 x 1.0-1.5 μm. *Conidiogenous cells* monophialidic, cylindrical, slightly tapering to the tip, discrete, determinate. hyaline, 10-13 x 2-2.6 μm. *Conidia* cylindrical, straight, with rounded ends, 1-septate, smooth, hyaline, 13-17 x 1.5-2 μm.

Specimen examined: On decaying leaf litter of *C. congesta* and on soil, GU campus, Taleigao, Goa; 7/9/1998, Miriam, J.; GUFCC No. 550 (live culture); 148(Herb. slide). Isolated by particle plating from leaf litter and moist chamber incubation.

Cylindrocladium parvum Anderson (Ref: Boedijn and Reitsma, 1950. Reinwardtia 1:54) (Fig. 54)

Colonies on MEA circular, with wavy margin, white, cottony, slimy, 4.8 cm diam. on 7 days; reverse of the colony light brown. *Mycelium* thin, white, with smooth, thin-walled, septate, branched, hyaline, 1.5 μm wide hyphae often thickened to form light brown, smooth chlamydospores. *Conidiophores* mononematous, occassionally arising in clusters, smooth, branched, erect, white, moderately thickwalled, up to 65 μm long, 6-7 μm wide, terminating in a branched fertile head and a single sterile, erect, smooth, hyaline, up to 128 long, 1.2-1.5 μm wide stipe with a

swollen biconic tip. *Conidiogenous cells* phialidic, discrete, penicillately arranged, cylindrical, slightly tapering to the tip, hyaline, $6.5-7.8 \times 2-2.6 \mu m$. *Conidia* slimy, cylindrical, rounded at both ends, 1-septate, smooth, hyaline, $2.5-3.7 \times 1.5 \mu m$.

Specimen examined: On fresh leaf of *F. benghalensis*; Verna, Goa; 16/41998; Miriam, J.; GUFCC No. 551 (Live culture); 149 (Herb. slide); Isolated by particle plating

Dichtyochaeta assamica (Agnihothrudu) Hughes et Kendrick, 1968. N.Z.Jrl.Bot,6: 334-335 (Fig. 55)

Colonies effuse, moderately dark brown, hairy. Mycelium immersed, setae not observed. Conidiophores mononematous, unbranched, straight to flexuous, dark brown lighter towards the apex, smooth, broad at the basal foot cell, 50-180 μm long, 3-6 μm wide. Conidiogenous cells mono- to polyphialidic, integrated, terminal, cylindrical with conspicuous flared collarettes. Conidia semi-endogenous, simple, cylindrical, falcate, rounded at ends, aseptate, colourless, smooth, with up to 10 μm long fine seluta at each end, 11-16 x 2-2.6 μm, aggregated in slimy groups.

Specimen examined: On leaf-litter of *C. congesta*; Verna, Goa; 21/9/1997; Miriam, J.; GUFCC No. 150 (Herb. slide); Isolated by moist chamber incubation.

Dicyma carisseae Miriam et Bhat sp. nov. (Fig. 56)

Colonies effuse, dark brown, black. Mycelium partly superficial, smooth, with septate, branched, light brown, 1.25 μm wide hyphae. Conidiophores mononematous, erect, straight, smooth, septate, branched, dark brown, thick-walled, becoming paler and thin-walled apically, 300-875 μm long, up to 3-5 μm thick. Conidiogenous cells polyblastic, intergrated, terminal, determinate, sometimes remaining apically hyaline

and sterile. Conidia solitary, dry, simple, obovid to spherical, spinulose, hyaline to light brown, $3-5 \times 2-4 \mu m$.

Holotype: On leaf litter of *C.congesta* GU campus, Taleigao, Goa; 27/9/1997, Miriam, J.; GUFCC No. 552 (Live culture); 151 (Herb. slide); Isolated by particle plating.

The monotypic genus *Dicyma* Boulanger, typified by *D. ampullifera* Boulanger, is the conidial state of *Ascotricha chartarum* Berk. (Carmichael et al. 1980; Ellis, 1971). *Dicyma carisseae* differs from the type species with its extensively long and sparcely branched conidiophores and comparatively smaller conidia.

Doratomyces indicus Miriam et Bhat anam.-sp.nov.

(Fig. 181)

Conidial fungi, hyphomycetes. *Colonies* effuse, medium to golden brown, hairy. *Mycelium* partly immersed, dense, cord-like, with branched, septate, pale to medium brown hyphae 2-3 µm wide. *Conidiophores* synnematous, conspicuous, erect, septate, branched, with 6-10 filaments compacted into distinct tufts and 15- 22 similar tufts interwoven in below half and splayed apart in above half giving appearance of branched synnema, up to 1000 µm long and up to 1500 µm in splayed apart above half region; each tuft of conidiophore 15-25 µm wide, narrowing towards the tip. *Conidiogenous cells* monoblastic, discrete, terminal or intercalary, ampuliiform, straight or curved, thick-walled, smooth, pale brown, 7-10 x 3-5µm. *Conidia* solitary, dry, oblong to obovate, rounded at the apex, truncate at the base, aseptate, thick-walled, colourless to pale brown, smooth, 7-12 x 2-5 µm.

With its synnematous conidiophores, discrete, ampulliform, curved and monoblastic conidiogenous cells and aseptate conidia, the taxon described above clearly belongs to the genus *Doratomyces* Corda, typified by *D. stemonites* (Pers. ex

Fr.) Morton & Smith (Ellis, 1971). So far 8 species have been described in the genus (Hawksworth et al, 1995). Of these, *D. microsporus* (Sacc.) Morton & Smith and *D. purpureofuscus* (Fr.) Morton & Smith show some similarity with their ampuliform conidiogenous cells and ovoid conidia but none of the hitherto described species of the genus exhibit interwoven and branching tufts of conidiophores in the synnema, as seen in *D. indicus*. Further, the conidia in *D. indicus* are not catenate where as these are in chains in all other species.

Holotype: From dead and decaying twigs of Ficus benghalensis L.; Verna, Goa, 12/11/1997, Miriam, J.; GUFCC No. 274 (Herb. slide); Isolated by moist chamber incubation..

Dreschlera tripogonis A.S. Patil et V.G. Rao, 1972. Trans. Br. mycol. Soc. 59: 339-341 (Fig. 57)

Colonies on MEA effuse, dark brown to black, regular, velvety, 5.2 cm diam in 7 days, reverse black. *Mycelium* thin, light brown spreading sparsely over the colony, interspersed with tiny brownish spores, with thin, smooth, septate, branched, light brown, 2.8-4.4 μm wide hyphae. *Conidiophores* mononematous, straight to flexuous, unbranched, smooth, septate, light brown, bearing numerous scars at the terminal part, 28-126 x 1.7-4 μm. *Conidiogenous cells* polytretic, integrated, terminal, sympodial, occassionally swollen, with cicatrized pores. *Conidia* solitary, acropleurogenous, simple, slightly curved, elongate, ellipsoidal, 3-6-pseudoseptate, smooth, light brown, 12.5-40 x 4.7-8.7 μm.

Specimen examined: On leaf-litter of *C. congesta*; Verna, Goa; 23/5/1998; Miriam, J.; GUFCC No.553(Live culture); 152 (Herb. slide); Isolated by particle plating.

Epicoccum purpurascens Ehreb. ex Schlecht., 1824. Synop.Pl.Crypt.: 136 (Fig. 58)

Colonies on MEA effuse, granular, slimy, very light, in concentric circles, 4.6 cm diam in 7 days with regular margin, light orangish brown, reverse dotted pale orange. *Mycelium* thin, raised, traversing entire colony in irregular chimps along with spores, brown, with smooth, thin-walled, septate, branched, pale yellow, 1-15 μm wide hyphae. *Sporodochia* pulvinate, black, randomly dispersed all over colony. *Conidiophores* mononematous, densely packed into clusters, branched, short, straight to flexuous, colourless, smooth. *Conidiogenous cells* monoblastic, integrated, terminal, determinate, cylindrical, 2-10 x 1.7-4.3 μm. *Conidia* solitary, dry, acrogenous, sub-spherical or pyriform, dark golden brown, muriform, with septa obscured by the rough opaque conidial wall, 14-30 μm diam.

<u>Specimen examined</u>: On leaf-litter of *C. congesta*; Verna, Goa; 28/10/1997; Miriam, J.; GUFCC No.554 (Live culture); 153 (Herb. slide); Isolated by particle plating.

Exosporium bryophylli T.S. Ramakrishnan, 1957. Proc. Indian Acad. Sci., B.46: 153-154 (Fig. 59)

Colonies effuse, hairy, dark brown black, 2.8 cm diam in 7 days, reverse of colony dark brown. *Mycelium* pale brown, with thin, smooth, septate, branched hyphae. *Conidiophores* mononematous, straight to flexuous, unbranched, moderately brown, smooth, up to 300 μm long and 5 μm wide. *Conidiogenous cells* polytretic, integrated, terminal, later becoming intercalary, sympodial, cylindrical to clavate, with dark cicatrized scars. *Conidia* solitary, acropleurogenous, simple, obclavate, slightly curved towards the apex, pale brown, smooth, 6-7-pseudoseptate, generally with a thick scar at the base, 20-50 x 11-15 μm.

Specimen examined: On leaf-litter of *C. congesta*; Verna, Goa; 22/7/1997; Miriam, J.; GUFCC No. 555 (Live culture); 154 (Herb. slide); Isolated by particle plating.

Fulvia fulva (Cooke) Ciferri, 1954. Atti Ist. bot. Univ. Lab. crittogam. Pavia, Ser. 5, 10: 245-246 (Fig. 60)

Colonies on MEA olive green, regular, circular, granular, with faint concentric zonations, edge off-white, 0.6 cm diam in 7 days, reverse green-grey with off-white edge. *Mycelium* thin, white, interspersed by slimy black, dark green to black masses of spores with thin, smooth, septate, branched, 1.5-2.8 μm wide hyphae. *Conidiophores* mononematous, straight to flexuous, geniculate, brown, smooth, septate, unbranched, 10.4-25.6 x 1.7-2.4 μm. *Conidiogenous cells* polyblastic, integrated, terminal, sympodial, slightly cicatrized. *Conidia* catenate, simple, elongate, cylindrical with truncate ends, slightly broader at the centre, 0-septate, smooth, light brown, 8.7-15.2 x 1.5-2.2 μm. *Ramoconidia* when present 10-11 x 2.4-2.6 μm, variable in shape with scars at both ends.

Specimen examined: On leaf-litter of *C. congesta*; Verna, Goa; 17/6/1997; Miriam, J.; GUFCC No. 556 (Live culture); 155 (Herb. slide); Isolated by particle plating.

Fusarium culmorum (W.G.Smith) Sacc. (Ref: Booth, 1971. The Genus Fusarium, 236)

(Fig. 61)

Colonies on MEA effuse, white, irregular, 2.6cm diam. in 7 days, granular, slimy, shiny droplets, reverse of the colony, cream. Mycelium moderate, white with thin-walled, smooth, septate, branched, 1-1.2μm wide hyphae. Conidiophore mononematous, straight to flexuous, branched profusely, septate, hyaline, smooth, 15-30 x 1.0-1.5μm. Conidiogenous cells phialidic, integrated, terminal, intercalary,

cylindrical, ampuliform. *Conidia* two types, microconidia catenate, 0-septate, ellipsoidal, hyaline, smooth, 4-5.2 x 1.7-2.2μm; macroconidia solitary 5-septate, elongate with beaked ends, hyaline, smooth, 16-21 x 1.7-2.2μm

Specimen examined: On leaf litter of *C. congesta* and soil, Verna, Goa; 21/10/1997; Miriam, J.; GUFCC No. 557(Live culture); 156 (Herb. slide); Isolated by particle plating and moist chamber incubation.

Fusarium decemcellulare Brick (Ref: Gilman, G.C.1957. A Manual of Soil Fungi: 362)

(Fig. 62)

Colonies on MEA effuse, bright pink, irregular, 1.8cm diam. in 7 days, granular, slimy, shiny droplets, reverse of the colony, bright pink with cream periphery. Mycelium moderate, white with thin-walled, smooth, septate, branched, 0.9-1.3μm wide hyphae. Conidiophore mononematous, straight to flexuous, branched profusely, septate, hyaline, smooth, 44-52 x 1-1.3μm. Conidiogenous cells phialidic, integrated, terminal, intercalary, cylindrical, ampuliform. Conidia two types: microconidia catenate, 0-septate, ellipsoidal, hyaline, smooth, 2.6-5 x 1.74μm; macroconidia solitary, 3-6-septate, elongate with beaked ends, hyaline, smooth, 51-63 x 5 μm

<u>Specimen examined</u>: On fresh leaf and leaf-litter of *C. congesta and F. benghalensis*; and soil, Verna, Goa; 23/10/1997; Miriam, J.; GUFCC No. 558 (Live culture); 157 (Herb. slide); Isolated by particle plating and moist chamber incubation.

Fusarium phragmitis Arnaud, 1953. Bull. Soc. mycol. Fr. 69: 298-301 (Fig. 63)

Colonies on MEA effuse, white, irregular, 2.6cm diam. in 7 days, granular, slimy, shiny droplets, reverse of the colony, cream. Mycelium moderate, white with thin-walled, smooth, septate, branched, 0.8-1.2µm wide hyphae. Conidiophore

mononematous, straight to flexuous, branched profusely, septate, hyaline, smooth, 12-18x 1.0-2μm. *Conidiogenous cells* phialidic, integrated, terminal, intercalary, cylindrical, ampuliform. *Conidia* two types, microconidia catenate, 0-septate, ellipsoidal, hyaline, smooth, 4.0-5.2 x 1.7-2.2μm; macroconidia catenate, 3-5-septate, elongate with beaked ends, hyaline, smooth, 16-21.3 x 1.7-2.2μm

Specimen examined: On decaying leaf litter and fresh leaf of *C. congesta* and soil; Verna, Goa; 12/4/1997; Miriam, J.; GUFCC No.559 (Live culture); 158 (Herb. slide); Isolated by particle plating and moist chamber incubation.

Fusarium semitectum Berk. et Rav., (Ref: Gilman, G.C.1957. A Manual of Soil Fungi: 362) (Fig. 64)

Colony on MEA effuse, white, irregular,1.2cm diam. in 7 days, granular, slimy, shiny droplets, reverse of the colony, cream. *Mycelium* moderate, white with thin-walled, smooth, septate, branched, 1.0-1.1μm wide hyphae. *Conidiophore* mononematous, straight to flexuous, branched profusely, septate, hyaline, smooth, 30-52 x 1.0-1.3μm. *Conidiogenous cells* phialidic, integrated, terminal, intercalary, cylindrical, ampuliform. *Conidia* two types, microconidia catenate, 0-septate, ellipsoidal, hyaline, smooth, 8.3-11.3 x 2.6-3.04μm; macroconidia catenate, 3-5-septate, elongate with beaked ends, hyaline, smooth, 17.4-28 x 2.2-3.5μm

Specimen examined: On decaying leaf litter and fresh leaf of *C. congesta* and soil; Verna, Goa Goa; 12/9/1997; Miriam, J.; GUFCC No. 560 (Live culture); 159 (Herb. slide); Isolated by particle plating and moist chamber incubation.

Fusarium solani (Mart.) Applel et Wollenw. (Ref: Gilman, G.C.1957. A Manual of Soil Fungi: 362) (Fig. 65)

Colony on MEA effuse, white, irregular, 2.4.0cm diam. in 7 days, granular, slimy, shiny droplets, reverse of the colony, cream. Mycelium moderate, white with

thin-walled, smooth, septate, branched, 1-1.2μm wide hyphae. *Conidiophore* mononematous, straight to flexuous, branched profusely, septate, hyaline, smooth, 12-28 x 1.3-1.7μm. *Conidiogenous cells* phialidic, integrated, terminal, intercalary, cylindrical, ampuliform. *Conidia* two types, microconidia catenate, 0-septate, ellipsoidal, hyaline, smooth, 4-8.3 x 2.2-3.5μm; macroconidia catenate, 3-5-septate, elongate with beaked ends, hyaline, smooth, 19-24 x 2.6μm.

Specimen examined: On soil; Verna, Goa; 13/10/1997; Miriam, J.; GUFCC No. 561(Live culture); 160 (Herb. slide); Isolated by particle plating.

Fusarium tabacinum (v.Beyma) W. Gams (Ref. Matsushima, 1975. Icones
Fungorum A Matsushima lectorum, 71) (Fig.66)

Colonies on MEA effuse, white, irregular, 3.0cm diam. in 7 days, granular, slimy, shiny droplets, reverse of the colony, cream. Mycelium moderate, white with thin-walled, smooth, septate, branched, 1-1.2μm wide hyphae. Conidiophore mononematous, straight to flexuous, branched profusely, septate, hyaline, smooth, 24-32 x 1.1-1.5μm. Conidiogenous cells phialidic, integrated, terminal, intercalary, cylindrical, ampuliform. Conidia two types, microconidia catenate, 0-septate, ellipsoidal, hyaline, smooth, 5-6 x 2-2.6μm; macroconidia catenate, 9-17-septate, elongate with beaked ends, hyaline, smooth, 35-44.4 x 3-4μm

Specimen examined: On soil; Verna, Goa; 12/3/1998; Miriam, J.; GUFCC No. 562 (Live culture); 161 (Herb. slide); Isolated by particle plating.

Gliomastix murorum (Corda) Hughes, 1958. Can. J. Bot. 36: 769. (Fig. 67)

Colonies on MEA olive green to light brown, with slightly convex surface, velvety, deep in agar, with circular margin, 1.5 to 2 cm diam after 7 days. Mycelium white to moderate grey, often compacting into 6-14 um thick dark brown strands,

with septate, smooth hyphae 1.25 μ m wide. Conidiophores mononematous, erect to slightly flexuous, unbranched, hyaline, up to 125 um long, 2-5-septate, 2-2.5 um wide. Conidiogenous cells monophialidic, integrated, terminal, determinate, smooth, sometimes percurrently proliferating, faintly darker near the apex, 2.5-125 x 1.25-2.5 μ m, with a distinct and flared collarette 1.8-3.7 μ m wide. *Conidia* slimy, ellipsoidal to cylindrical, rounded at the apex, narrow and truncate at the base, moderately brown, smooth, 0-septate, 2.5-6.5 x 2.5-3 um.

Specimen examined: On leaf-litter of *C. congesta*; Verna, Goa; 18/11/1997; Miriam, J.; GUFCC No. 563(Live culture); 162(Herb. slide); Isolated by particle plating.

Gliomastix uniseptata Miriam et Bhat sp. nov.

(Fig. 68)

Colonies on MEA green, regular, 1.3 cm diam in 7 days, reverse brownish green. Mycelium greyish green, cord-like, with thin, smooth, septate, branched, 1.5-1.7 μm wide hyphae. Conidiophores mononematous, arising from a mass of hyphae, unbranched, straight to flexuous, light brown, smooth, septate, faintly darker at the apex. Conidiogenous cells monophialidic, integrated, terminal, determinate, sometimes percurrent, 18-72 μm long, 2.2-4 μm wide above the base; collarette 1-2 μm wide. Conidia endogenous, simple, hyaline, smooth, 0-1-septate, ellipsoidal, with pointed ends, 4.7-7 x 1-2 μm.

Holotype: On leaf litter of *C. congesta* and leaf litter and fresh leaves of *Ficus benghalensis*; GU campus, Taleigao, Goa; 27/9/1997, Miriam, J.; GUFCC No. 564 (Live culture); 163 (Herb. slide); Isolated by particle plating and moist chamber incubation.

This is the only species in the genus *Gliomastix* Gueguen, typified by *G. murorum* (Corda) Hughes, with septate conidia (Ellis, 1971, 1975; Matsushima, 1975).

Gonatobotryum bimorphospora Miriam et Bhat sp. nov., 2000. Cryptogam. mycol. 9: 23-27. (Fig. 69)

Colonies in malt extract agar effuse, moderately fast growing effuse, dark brown, with irregular margin, with rough surface, faintly concentric, attaining 4-4.5 cm diam after 7 days. *Mycelium* partly immersed, partly superficial, often compacted into root-like aggregates towards the periphery of the colony, with septate, repeatedly branched, smooth-walled, pale brown, hyphae 4.5-10 μm wide. *Conidiophores* distinct, mononematous, flexuous to erect, brown, septate, unbranched, generally smooth, 175-550 μm long, 6.2-10 μm wide, percurrently regenerating, nodose and distinctly echinulate in the terminal and intercalary conidiogenous ampullae 13.5-45 x 15-22 μm. *Conidiogenous cells* polyblastic, integrated, terminal or intercalary, spherical to subspherical, ampullae with slightly raised and truncate conidiogenous loci. *Conidia* catenate, 0-septate, smooth, pale to moderately brown, cicatrized at the base, of two types. First formed conidia arise directly on conidiogenous cells, elongate-ellipsoidal to elongate-obclavate, with 1-3 apical conidiogenous loci, 7.5-11.5 x 3-4.5 μm; later formed conidia arise on first-formed conidia, ellipsoidal, basipetal, 3-6.2 x 2.5-3.5 μm.

Holotype: Dried agar culture mat of the fungus isolated from fresh leaves of *Carissa congesta*, Taleigao Plateau, 22 Nov 1998, Miriam J., Herb. No. GUFCC-0398.

The genus *Gonatobotryum*, with *G. fuscum* (Sacc.) Sacc. as type species, is characterised by mononematous conidiophores producing integrated, terminal or intercalary, percurrent and polyblastic conidiogenous cells and catenate conidia (Kendrick, Cole and Bhatt, 1968; Ellis, 1971; Manjal and Gill,1968). *G. bimorphospora* differs from the hitherto described species in the genus, namely *G.*

fuscum, G. apiculatum (Peck) Hughes (Hughes, 1953) and G. indicum Manjal and Gill, with its two distinct types of conidia and endophytic habitat. The earlier described species produced only one type of conidia and were isolated from habitats other than endophytic.

Colonies effuse, brown, velvety. Mycelium partly immersed. Conidiophores mononematous, unbranched, straight to flexuous, dark brown at the base, light brown to hyaline at the tip, septate, smooth, 40-180 x 6.5-10 μm. Conidiogenous cells polyblastic, integrated, terminal, determinate, sympodial, denticulate, with dark scars on conidial secession. Conidia solitary, dry, simple, helicoid, hyaline, smooth, multiseptate, coiled in and rounded at the tip, narrow and truncate at the base, 3-6 μm wide, with full spore 17-20 μm in diam.

Specimen examined: On leaf litter of *C.congesta* and *F. benghalensis*; GU campus, Taleigao, Goa; 21/11/1998, Miriam, J.; GUFCC No. 565(Live culture); 164 (Herb. slide); Isolated by particle plating.

Helicomyces roseus Link (Ref: Matsushima, 1975. Icones Fungorum a Matushima Lectorum: 81) (Fig. 71)

Colonies effuse, light brown. Mycelium partly immersed. Conidiophores mononematous, branched, straight to flexuous, light brown at the base, hyaline at the tip, septate, smooth, 65-78 x 2.2-3μm. Conidiogenous cells polyblastic, integrated, terminal, determinate, denticulate, with dark scars on conidial secession. Conidia solitary, dry, simple, helicoid, hyaline, smooth, multiseptate, coiled in and rounded at the tip, narrow and truncate at the base, 2.6-3.5 μm wide, with full spore 17-20 μm in diam.

Specimen examined: On leaf-litter of *C. congesta*; Verna, Goa; 17/12/1997; Miriam, J.; GUFCC No. 566 (Live culture); 165 (Herb. slide); Isolated by particle plating and moist chamber incubation.

Helicosporium guianensis Linder 1929. Ann. M. Bot. Gdn. 16:280 (Fig. 72)

Colonies effuse, light brown. Mycelium partly superficial. Conidiophores mononematous, branched, straight to flexuous, very light brown, septate, smooth, 65-280 x 3-4 μ m. Conidiogenous cells monoblastic, discrete, cylindrical, determinate, denticulate, 1.5-2.5 x 1-1.5 μ m. Conidia solitary, dry, simple, hyaline, multiseptate, helicoid, 2-3 times coiled in one plane; each filament 1.5 μ m wide; conidia 14-17 μ m diam.

<u>Specimen examined</u>: On leaf litter of *C.congesta and F. benghalensis* GU campus, Taleigao, Goa; 23/11/1997, Miriam, J.; GUFCC No.166(Herb. slide); Isolated by moist chamber incubation.

Helicosporium lumbricoides Sacc. (Ref.: Linder 1929. Ann. M.. Bot. Gdn. 16:282)

(Fig. 73)

Colonies effuse, hairy, velvety, greyish brown. Mycelium partly superficial, partly immersed. Conidiophores mononematous, unbranched, straight to flexuous, brown, slightly darker at the base, smooth, septate, 336-416 μm long, 2.5 μm wide at the tip, 5.0 μm wide at the base. Conidiogenous cells polyblastic, discrete, terminal or intercalary, determinate, lobed, denticulate, denticles cylindrical, 6.5-7.5 x 2.5 μm. Conidia solitary, dry, simple, helicoid, hyaline, smooth, multiseptate, borne on cyclic pegs, 1-2 times coiled, rounded at apex, truncate at base, each filament 1.25 - 1.8 μm wide; conidia 14-26 μm diam.

Specimen examined: On leaf litter of *C.congesta and F. benghalensis*; GU campus, Taleigao, Goa; 15/9/1997, Miriam, J.; GUFCC No. 567 (Live culture); 167(Herb. slide); Isolated by particle plating.

Helminthosporium dalbergiae M.B. Ellis, 1961. Mycol Pap. 82: 2-21. (Fig. 74)

Colonies effuse, hairy, dark brown. Mycelium partly superficial, partly immersed. Conidiophores mononematous, smooth, septate, unbranched, dark brown, thick-walled, straight to flexuous, with pores at the apex and laterally below the septa, up to 1100 μm long, 8-10 μm wide in the middle, 5-6 μm wide above half. Conidiogenous cells polytretic, integrated, cylindrical, terminal or intercalary determinate. Conidia solitary, developing laterally in verticles, obclavate to obpyriform, with an elongate tip, with a prominent scar at the base, 5-6-pseudoseptate, smooth, light brown, 62-94 μm long, 8-15 μm wide in the middle, 3-4 μm wide at the tip.

Specimen examined: On leaf litter of *C.congesta*; GU campus, Taleigao, Goa; 16/12/1997, Miriam, J.; GUFCC No. 568 (Live culture); 168(Herb. slide);. Isolated by particle plating and moist chamber incubation.

Helminthosporium mauritianum Cooke (Ref: Ellis, 1971. Dematiaceous Hyphomycetes, p. 212-213) (Fig. 75)

Colonies effuse, hairy, dark brown. Mycelium smooth, with septate, branched, light brown, thick-walled, 1.25 μm wide hyphae. Conidiophores mononematous, straight or flexuous, smooth, septate, unbranched, dark brown, thick-walled, with pores at the apex and laterally below the terminal 3-4 septa, 300-875 μm long, up to 65 μm wide at the base and tapering to 12 μm wide at the tip. Conidiogenous cells polytretic, intergrated, terminal or intercalary, determinate, cylindrical. Conidia solitary, dry, developing laterally often in verticels, growth of conidiophore ceases with conidia at the tip, obclavate with prominent scar at the base, 5-pseudoseptate,

smooth, light brown, 96-112 μm long, 16-22 μm wide at the base, tapering to 8-12 μm wide tip.

Specimen examined: On leaf litter of *C.congesta*; GU campus, Taleigao, Goa; 23/11/1997, Miriam, J.; GUFCC No.169 (Herb. slide);. Isolated by moist chamber incubation.

Helminthosporium microsorum Sacc. (Ref.: Ellis, 1971. Dematiaceous Hyphomycetes: p. 393) (Fig. 76)

Colonies effuse, hairy, dark brown, velvety. Mycelium partly immersed, partly superficial. Conidiophores mononematous, smooth, septate, unbranched, dark brown, thick-walled, straight to flexuous, cylindrical, with pores at the apex and laterally below the septa, up to 300 μm long, 6.5 μm wide in the middle and below half, 4 μm wide at the tip. Conidiogenous cells polytretic, integrated, terminal, later becoming intercalary, determinate, cylindrical. Conidia solitary, acropleurogenous, obclavate, with a prominent ciccatrized scar at the base, 4-7-pseudoseptate, smooth, brown, 30-40 μm long, 6-7 μm wide in the middle, 2-4 μm wide at the tip.

<u>Specimen examined</u>: On leaf litter of *C.congesta* GU campus, Taleigao, Goa; 17/9/1997, Miriam, J.; GUFCC No. 569 (Live culture); 170 (Herb. slide);. Isolated by particle plating and moist chamber incubation.

Heteroconium solaninum (Sacc. et Syd.) M.B. Ellis, 1971. More Dematiaceous
Hyphomycetes: 65-66 (Fig. 77)

Colonies on MEA, effuse, brown, 2.1 cm diam in 7 days, reverse light brown. Mycelium light brown, with smooth, thin-walled, septate, branched, pale brown, 1.7-3.4 µm wide hyphae. Conidiophores mononematous, straight to flexuous, unbranched, light brown, smooth, 7.5-93.6 x 1-10 µm. Conidiogenous cells polyblastic, integrated, terminal, sympodial, with prominent cicatrized scars.

Ramoconidia elongated, almost cylindrical, smooth, 1-3-septate, light brown, with cicatrized scars, $11.5\text{-}15.6 \times 3.7\text{-}5 \ \mu\text{m}$. Conidia catenate, acropleurogenous, simple, elongated, wide in the middle, narrow and truncate at both ends, smooth, 3-septate, light brown, $10.6\text{-}15 \times 2.5\text{-}3.5 \ \mu\text{m}$.

Specimen examined: On leaf litter of *C.congesta*; GU campus, Taleigao, Goa; 22/7/1997, Miriam, J.; GUFCC No. 570 (Live culture); 171 (Herb. slide);. Isolated by particle plating.

Humicola brevis (Gilman et Abbott) Gilman (Ref: Gilman, 1957. A Manual of Soil Fungi, 325) (Fig. 78)

Colonies on MEA, effuse, very pale brown, regular, reverse very light brown. Mycelium thin, white, more concentrated in centre with thin smooth, septate, hyaline, branched, 1.5 μm wide hyphae. Conidiophores mononematous, unbranched, straight to flexuous, cylindrical sometimes flask-shaped with a narrow base, slightly broad in the middle, narrow at the tip, moderately brown, smooth, septate, 14-60 x 3-4.6 μm. Conidiogenous cells monoblastic, integrated, terminal, determinate, cylindrical. Conidia solitary, dry, simple, spherical, pale to moderately brown, faintly verrucose, 0-septate, 9.4 μm diam.

Specimen examined: On leaf litter of *C. congesta*; GU campus, Taleigao, Goa; 27/9/1997, Miriam, J.; GUFCC No. 571(Live culture); 172 (Herb. slide);. Isolated by particle plating.

Humicola grisea Traaen, 1914. Nyt. Mag. Naturvid., 52: 34 (Fig. 79)

Colonies effuse, cottony, white, irregular, 1.5 diam in 7 days, reverse off-white. Mycelium moderate white, with thin smooth, septate, branched 1-2.2 µm wide hyphae. Conidiophores mononematous, unbranched, elongate, cylindrical, sometimes swollen, straight to flexuous, hyaline, smooth, 1-13 x 1-4 µm. Conidiogenous cells

monoblastic, integrated, terminal, determinate, cylindrical. *Conidia* solitary, dry, simple, spherical to obovate, golden brown, smooth, 0-septate, 8.5-15.6 µm diam.

Specimen examined: On leaf litter of *C. congesta*; GU campus, Taleigao, Goa; 27/9/1997, Miriam, J.; GUFCC No. 572 (Live culture); 173 (Herb. slide); Isolated by Particle plating.

Humicola verrucosa Miriam et Bhat sp. nov.

(Fig. 80)

Colonies effuse, cottony, golden brown. Mycelium superficial, 1.7-2.6 μm wide hyphae. Conidiophores mononematous, branched, basipetal, straight to flexuous, hyaline, smooth, 7- 30 x 1-2 μm. Conidiogenous cells monoblastic, integrated, terminal, determinate, cylindrical. Conidia solitary, dry, simple, spherical, pale to mid-golden brown, wavy at the margin, aseptate, 8.7-15.6 μm diam.

Holotype: On decaying leaf litter of *C. congesta*, and soil; Verna, and GU Campus, Taleigao, Goa; 17/10/1998; Miriam, J.; GUFCC No. 573 (live culture); 174 (Herb. slide). Isolated by particle plating.

Hyaloscolecobasidium Miriam et Bhat gen nov.

Colonies effuse, reddish brown, Mycelium thin, with smooth, branched, hyaline, septate hyphae. Conidiophores mononematous, straight, hyaline, thin-walled, septate, branched. Conidiogenous cells polyblastic, integrated, terminal or intercalary, denticulate. Conidia solitary, simple, fusiform, elongate cylindrical with rounded ends, hyaline, septate, smooth.

Type sp. Hyaloscolecobasidium indicum Miriam et Bhat sp. nov.

The fungus is compared with genera such as *Scolecobasidium* Abbott and *Cercosporula* Arnaud (Carmichael et al. 1980) in which the conidiophores and conidia are dematiaceous, although all these produce blastic type conidia. The conidia and conidiophores in *Scolecobasidium* are verrucose where as in

Hyaloscolecobasidium, these are smooth. The denticulate conidiogenous loci in Hyaloscolecobasidium are terminal and at one plane whereas in other related genera the conidiogenous loci are spread over the conidiogenous cells and conidia are echinulate.

Hyaloscolecobasidium indicum Miriam et Bhat sp. nov. (Fig. 81)

Colonies on MEA circular, reddish brown, margin roots irregular, with 3 concentric zones, central circle showing mycelium in an anticlockwise direction, 3 cm diam. in 7 days, pores seen as white tiny stars distributed all over the colony centrally dark green with off-white peripherral region. *Mycelium* thin, white to light brown, with smooth, branched, hyaline, thin-walled, septate, 1-1.5 μm wide hyphae. *Conidiophores* mononematous, smooth, straight, hyaline, thin-walled, septate, branched, 1-25 x 1-1.5 μm. *Conidiogenous cells* polyblastic, integrated, terminal or intercalary, denticulate; denticles cylindrical, sometimes arising directly from the hyphae. Conidia solitary, simple, fusiform, elongate cylindrical with rounded ends, hyaline, 2-septate, smooth, 13.5-35 x 1-2 μm.

Holotype: On decaying leaf litter of *C. congesta* and *F. benghalensis* and soil; Verna, and GU Campus, Taleigao, Goa; 17/10/1998; Miriam, J.; GUFCC No. 574 (Live culture); 175 (Herb. slide). Isolated by particle plating and moist chamber incubation.

Hyphodiscosia jaipurensis Lodha et Reddy, 1974. Trans. Br. mycol. Soc. 62: 418-421 (Fig. 82)

Colonies effuse, pale brown, velvety. Mycelium mostly immersed. Conidiophores mononematous, erect, straight to flexuous, often geniculate, pale brown, unbranched, 2-4-septate, smooth, terminating in a wedge-shaped tip, 30-50

 μ m long, 4-5.5 μ m wide. *Conidiogenous cells* polyblastic, sympodial, integrated, terminal. *Conidia* solitary, cylindrical, with rounded tip and truncate base, slightly curved, 1-septate, colourless, smooth, with a fine seluta at each end, 17.4 - 24 x 3.4-4.7 μ m; setulae up to 10 μ m long.

Specimen examined: On leaf-litter of *C. congesta*; Verna, Goa; 18/9/1997; Miriam, J.; GUFCC No.575 (Live culture); 176 (Herb. slide); Isolated by moist chamber incubation.

Idriella fertilis (Pirozynski et Hodges) Matsushima, 1975. Icones Microfungorum

Matsushima Lectorum: p.86 (Fig.83)

Colonies on MEA circular, slimy, granular, irregular, dark green, 3 cm diam in 7 days; reverse dark green at the centre. *Mycelium* thin, white, with smooth, thinwalled septate, branched, light brown, 1.5 μm wide hyphae; hyphae often thickened to form light brown smooth, 1-3-septate, 18.7-27 x 12-16 μm chlamydospores. *Conidiophores* mononematous, smooth, unbranched, straight to flexuous, light brown, 18-40 μm long, 2-2.5 μm wide at the base, 1.25 μm wide at the tip. *Conidiogenous cells* polyblastic, denticulate, integrated, terminal. *Conidia* dry, solitary, simple, falcate with pointed ends, smooth, hyaline, 0-septate, 7.5 x 1.25 μm.

Specimen examined: On decaying leaf litter and fresh leaf of *C. congesta*, and soil; Verna, and GU Campus, Taleigao, Goa; 17/10/1998; Miriam, J.; GUFCC No. 576 (live culture); 177 (Herb. slide). Isolated by particle plating and moist chamber incubation.

Idriella lunata Nelson et Wilhelm, 1956. Mycologia 48: 550 (Fig. 84)

Colonies on MEA circular, with wavy margin, dark green at the centre with decreasing colour intensity towards the periphery, with granular surface, 3.5 cm diam. in 7 days; reverse of the colony dark green at the centre, extended towards the

periphery with creamish white concentric rings. *Mycelium* thin, with smooth, thinwalled, septate, branched, light brown, 1.5 μm wide hyphae; hyphae often thickened to form dark brown, smooth, 1-3- septate, 19-27 x 12-16 μm chlamydospores with reduced cell lumen. *Conidiophores* mononematous, smooth, unbranched, straight to flexuous, light brown, thick-walled, 18-44 μm long, 3-4 μm wide at the base, tapering to 1.5 μm neck and 4-5 μm wide tip. *Conidiogenous cells* polyblastic, integrated, denticulate, terminal. *Conidia* dry, solitary, falcate, smooth, with pointed ends, hyaline, 0-septate, 8.5-18 x 1-3 μm.

Specimen examined: On decaying leaf litter and fresh leaf of *C. congesta* and soil; Verna, and GU Campus, Taleigao, Goa; 17/10/1998; Miriam, J.; GUFCC No. 577(live culture); 178(Herb. slide). Isolated by particle plating and moist chamber incubation.

Idriella mucoidea Miriam et Bhat sp. nov.

(Fig. 85)

Colonies on MEA light brown, regular, with numerous concentric and occassional radial zonations, 1.3 cm diam in 7 days; reverse light brown. *Mycelium* thin, white, loose and prominent at the periphery, with smooth, thin-walled, septate, branched, light brown, 1.5 μm wide hyphae; hyphae often thickened to form spherical, dark brown, smooth, 15-26 μm diam. chlamydospores. *Conidiophores* mononematous, smooth, unbranched, straight to flexuous, pale brown, 7.5-25 x 2.5 μm; conidiophore wall covered by a thick, granular, slimy layer. *Conidiogenous cells* polyblastic, integrated, sympodial, terminal, denticulate; denticles, minute, conspicuous. *Conidia* solitary, simple, falcate, with pointed ends, 0-septate, hyaline, smooth, 7.5-20 x 1.5-2.5 μm.

Holotype: On decaying leaf litter and fresh leaf of *C. congesta* and soil; Verna, and GU Campus, Taleigao, Goa; 17/10/1998; Miriam, J.; GUFCC No. 578 (live culture); 179(Herb. slide). Isolated by particle plating and moist chamber incubation.

The genus *Idriella* Nelson et S. Wilhelm, typified by *I. lunata* Nelson et S. Wilhelm, is characterised by spherical, thick-walled, dark brown chlamydospores, mononematous conidiophores, polyblastic conidiogenous cells and lunar shaped, hyaline, dry conidia. So far 22 species have been recognised in the genus based on the size and shape of the conidiogenous apparatus and conidia (Hawksworth et al., 1995). None of the so far described species are known to possess slimy conidiophores and conidia (Ellis, 1971).

Idriella multiseptata Miriam et Bhat sp. nov.

(Fig. 86)

Colonies on MEA, green, with concentric zonation, 1.1 cm diam in 7 days, reverse light green. *Mycelium* thin, greyish white, with smooth, thin-walled, septate, branched, light brown, 1.25 μm wide hyphae. *Conidiophores* mononematous, smooth, branched, straight to flexuous, brown, 3-8-septate, pale brown towards the tip, 20-85 x 3-4 μm; Chlamydospores catenate, thgick-walled, light brown, smooth, 8-10 um diam. *Conidiogenous cells* polyblastic, terminal, later becoming intercalary, wedge-shaped, light brown, denticulate, 10-20 x 2.5-7.5 μm. *Conidia* solitary, simple, fusiform, broad in the middle and above, pointed at both ends, hyaline, smooth, 3-10-septate, 25-40 μm long and 2.5 μm wide at the middle, 1.5 μm wide at the pointed tip.

Holotype: On decaying leaf litter of *C. congesta* and soil; Verna, Goa; 17/10/1998; Miriam, J.; GUFCC No. 579(live culture); 180(Herb. slide). Isolated by particle plating.

The genus *Idriella* Nelson et S. Wilhelm, typified by *I. lunata* Nelson et S. Wilhelm, is characterised by spherical, thick-walled, dark brown chlamydospores, mononematous conidiophores, polyblastic conidiogenous cells and lunar shaped,

hyaline, dry conidia. So far 22 species have been recognised in the genus based on the size and shape of the conidiogenous apparatus and conidia (Hawksworth et al., 1995). Of these, *I. ramosa* Matsushima is known to possess 1-2-septate conidia. The present taxon with its multiseptate conidia is recognised as distinct amonst the species of *Idriella*.

Idriella ramosa Matsushima, 1971. Microfungi of the Solomon Islands and Papua-New Guinea, 31. (Fig. 87)

Colonies on MEA, greyish green, irregular, with concentric zonations, 3 cm diam. in 7 days; reverse centrally dark green and remaining off-white. *Mycelium* moderate, light brown, loose, receeding towards the periphery, olive green, with smooth, thin-walled septate, branched light brown 2.5 μm wide hyphae. *Conidiophores* mononematous, straight to flexuous, light golden brown, becoming paler towards the tip, septate, branched, smooth, thick-walled, 45-74 μm long, 4.3-7.8 μm wide at the base, 2-2.5 μm wide at the tip. *Conidiogenous cells* polyblastic, integrated to discrete, sympodial, terminal, denticulate. *Conidia* solitary, simple, lunate, with pointed ends, 0-1-septate, hyaline, smooth, 4-10 x 1.5-2 μm.

Specimen examined: On decaying leaf litter and fresh leaf of *C. congesta* and soil; Verna, and GU Campus, Taleigao, Goa; 17/10/1998; Miriam, J.; GUFCC No. 580 (live culture); 181 (Herb. slide). Isolated by particle plating.

Kumbhamaya Miriam et Bhat anam. -gen. nov. 2000. Cryptogam. mycol. 9: 23-27.

Conidial fungi, hyphomycetes. Colonies effuse, medium to dark brown. Mycelium dense, with branched, thickly septate, medium to dark brown hyphae. Conidiophores mononematous, indistinct, septate. Conidiogenous cells monophalidic, integrated, terminal or intercalary, kettle- to pitcher-like, often with an elongated base, straight to curved, thick-walled, smooth, medium to dark brown, with a distinct and flared collarette, constricted and narrow at the neck. Conidia solitary, fusiform, curved to sigmoid, pointed at both ends, sharply beaked at the tip, occasionally recurved at the base, hyaline, thick-walled, smooth, 1-3-septate, aggregate in slimy mass.

Type species: Kumbhamaya indica Miriam et Bhat sp. nov.

Several genera of mononematous hyphomycetes with conspicuous phialides bearing flared collarette and conidia in slimy heads are known (Carmichael et al. 1980; Ellis, 1971, 1976; Matsushima, 1987) These include Bahusutrabeeja Subram.and Bhat (1977), Craspedodidymum Holubova-Jacova (1972), Dischloridium Sutton (1977), Nawawia Marvanova (1980), Obeliospora Kuthub.and Nawawi (1990) and Phialophora Medlar and Phialomyces Misra and Talbot (Carmichael et al. 1980). In the genus Bahusutrabeeja, the branched conidiophores terminating in apically inflated phialides with small funnel-shaped collarettes, produce hyaline, globose to pear-shaped, non-septate, setulate conidia. In Craspedodidymum, the apically inflated phialides with large funnel-shaped collarette, produce brown, non-septate conidia. In Dischloridium phialides bear no conspicuous collarette and conidia are non-septate. The conidia are turbinate and setulate in Nawawia although the phialides possess flared collarette. The monophialidic conidiophores with conspicuously flared collarette in Obeliospora develop along with setae and produce setulate and globose conidia. In Phialophora phialides are small and conidia inconspicuous. In Phialomyces, although the phialides are typical with flared collarette, the conidia not only globose but also are in chains. Kumbhamaya differs from all these genera in that the kettle or pitcher-shaped phialides bearing conspicuous and flared collarette produce fusiform, curved, septate, hyaline conidia which are pointed at both ends.

Kumbhamaya indica Miriam et Bhat sp. nov. 2000. Cryptogam. mycol. 9: 23-27.

(Fig. 88)

Colonies on malt extract agar effuse, with moderately dense aerial mycelium, irregular at margin, medium brown at the centre, pale brown towards the periphery, 2-3.5 cm diam. in 7 days; reverse of the colony dark brown. Mycelium dense, with branched, with thick septa, medium to moderately dark brown hypha 2.2-7.5 μ m wide. Conidiophores mononematous, indistinct, septate, 2.2-2.5 um diam. Conidiogenous cells monophalidic, integrated, terminal or intercalary, vase-like to wormiform, often with an elongate base, straight to curved, thick-walled, smooth, medium to dark brown, 12-50 μ m long and 2.5-4.5 um wide at the base, 5.5- 8.5 μ m wide in the middle, with a distinct and flared collarette 2-3.5 x 1.5-2.5 um, constricted and narrow at the neck region. Conidia solitary, fusiform, curved to sigmoid, pointed at both ends, sharply beaked at the tip, occasionally recurved at the base, hyaline, thick-walled, smooth, mostly 3-septate, rarely 1-septate, 25-40 x 3.5-5.5 μ m, aggregating in slimy mass at the apex of the phialide.

Holotype: Dried culture grown on MEA medium and isolated from tip of young leaf of *Carissa*, 14.11.1997. Verna, Goa, Miriam, J. Slide No.GUFCC-; *Paratype:* Dried culture of the fungus grown on MEA medium and isolated from mature leaf of *Bambusa*, 14.11.1997, Mollem, Goa, Maria D'Souza, Herb. No. GUFCC-0238.

(Fig. 89)

Colonies effuse, white, shining, with irregular wavy margin, with black spore masses spread over, powdery, 1-1.5 cm diam. in 7 days, reverse of the colony dark brown. *Mycelium* thin, with hyaline, smooth, thin-walled, septate, branched, up to 1.5 μm wide hyphae. *Conidiophores* mononemaotous, simple, grey, darker at the tip and decreasing in colour intensity towards the base, faintly verrucose at the tip and decreases in intensity towards base, 24-65 x 1.5-2.6 μm, terminating in 4-6 phialides at its tip. *Conidiogenous cells* monophialidic, integrated, terminal. usually 6, clavate to pyriform, dark grey, 5.6-11 x 2-2.6 μm. *Conidia* catenate, spherical or flattened dorsiventrally, grey to black, verrucose, 3.5-5.2 μm diam.

Specimen examined: On decaying leaf litter of *C. congesta* and on soil; Verna, Goa; 15/5/1997; Miriam, J.; GUFCC No. 581 (Live culture); 182 (Herb. slide). Isolated by particle plating and moist chamber incubation..

Monodictys nigra Matsushima, 1975. Icones Fungorum A Matsushima Lectorum: 98

(Fig. 90)

Colonies effuse, blackish brown. Mycelium mostly superficial, with thin, septate, hyaline, pale brown, smooth, 2.2-2.6 μm wide hyphae. Conidiophores mononematous, unbranched, straight to flexuous, hyaline, to light brown, smooth, cells sometimes swollen, 5.6-10.8 x 2.6- 8.2 μm. Conidiogenous cells monoblastic, integrated, terminal, determinate, cylindrical, subspherical. Conidia solitary, dry, acrogenous, simple, oblong, subspherical to ellipsoidal or irregular, dark brown to black, smooth, muriform, basal cells often inflated and paler, 23-41 x 8.7-32 μm.

<u>Specimen examined</u>: On leaf-litter of *C. congesta*; Verna, Goa; 13/6/1997; Miriam, J.; GUFCC No. 582 (Live culture); 183 (Herb. slide); Isolated by particle plating and moist chamber incubation.

Monodictys lepraria (Berk.) M.B.Ellis, 1976. More dematiaceous Hyphomycetes: 44

(Fig. 91)

Colonies effuse, black. Mycelium superficial, with thin hyaline to pale yellow, septate, smooth, 2-4 μm wide hyphae. Conidiophores mononematous, unbranched, straight to flexuous, hyaline, to pale brown, smooth, with oblique septa, 22-30 x 3-5 μm. Conidia solitary, dry, acrogenous, simple, oblong, ellipsoidal, irregular, muriform, with wavy margin, 34-55 x 17-40 μm.

Specimen examined: On leaf-litter of *C. congesta*; Verna, Goa; 18/7/1997; Miriam, J.; GUFCC No. 583 (Live culture); 184 (Herb. slide); Isolated by particle plating.

Monodictys fluctuata (Tandon et Bilgrami) M.B. Ellis, 1976. More dematiaceous Hyphomycetes: 44 (Fig. 92)

Colonies on MEA, brown, effuse, granular. Mycelium moderate, pale brown, with thin septate, smooth 2.4-4 µm wide hyphae. Conidiophores mononematous, unbranched, straight to flexuous, golden brown, smooth, cells sometimes swollen and larger than the others, 7.4-38 x 5.6-8 µm. Conidia solitary, dry, acrogenous, simple, oblong to subspherical, ellipsoidal, with wavy irregular outerwall, brown to dark brown, muriform, basal cells inflated, paler and thinner walled than the other cells.

Specimen examined: On leaf-litter of *C. congesta*; Verna, Goa; 12/11/1997; Miriam, J.; GUFCC No.584 (Live culture); 185 (Herb. slide); Isolated by particle plating.

Monodictys glauca (Cooke et Harkn.) Hughes, 1958. Can.J. Bot. 36: 785 (Fig. 93)

Colonie on MEA, olive green, regular, granular, faint concentric circles, darker at the centre, decreasing in color intensity towards the periphery, 4 cm diam. in

7 days; reverse greenish brown. *Mycelium* thin, light brown, very loosely interspersed, with dark brown to black slimy long spores spread over the colony, with thin-walled, hyaline to pale brown, septate, smooth, 2-3.3 μm wide hyphae. *Conidiophores* mononematous, unbranched, straight to flexuous, hyaline to pale brown, smooth, 4.3-12.6 x 2.2-4.7 μm. *Conidia* solitary, dry, simple, subspherical, twisted, brown to dark brown, smooth, muriform, basal cells sometimes inflated, paler and thinner walled than the other cells 10-20 x 11.7-20 μm.

Specimen examined: On leaf-litter of *C. congesta*; Verna, Goa; 21/9/1998; Miriam, J.; GUFCC No. 585 (Live culture); 186 (Herb. slide); Isolated by particle plating.

Monodictys cultura Miriam et Bhat sp. nov.

(Fig. 94)

Colonies on MEA light brown, 2.8 cm diam in 7 days; reverse light brown. Mycelium with thin-walled, smooth, septate, branched, 1-2 μm wide hyphae. Conidiophores mononematous, branched, straight to flexuous, hyaline, smooth, 10-57 x 1.5-3 μm. Conidia solitary, dry, ellipsoidal, sub-spherical, irregular, sometimes twisted, light brown to moderately brown, smooth, muriform, basal cell paler, 14-26 x 8-16.5 μm.

Holotype: On leaf-litter of *C. congesta*; Verna, Goa; 13/10/1997; Miriam, J.; GUFCC No.586 (Live culture); 187 (Herb. slide); Isolated by particle plating.

Several species of the genus *Monodictys* Hughes, typified by *M. putredinis* (Wallr.) Hughes, are known in the literature (Ellis, 1971, 1976; Mastushima, 1971, 1976). *M. cultura* is unique in the genus with its pale brown, muriform, twisted conidia.

Colonies on MEA, pale olive green, granular, with concentric circles, slimy, 1 cm diam. in 7 days; reverse olive green. *Mycelium* thin, traversing flatly, with thinwalled septate, branched, hyaline,1-1.7 μm wide hyphae. *Conidiophores* mononematous, straight to flexuous, hyaline, smooth, 1.5-3 x 1-1.5 μm. *Conidia* solitary, simple, sub-spherical to spherical sometimes twisted, pale to golden brown, smooth, muriform, basal cell inflated and slightly darker than other cells, 8.3-14.5 x 6.5-13.5 μm.

Specimen examined: On leaf-litter of *C. congesta*; Verna, Goa; 22/5/1998; Miriam, J.; GUFCC No.587 (Live culture); 188 (Herb. slide); Isolated by particle plating.

Moorella ficusensis Miriam et Bhat sp. nov.

(Fig. 96)

Colonies effuse, brown. Mycelium mostly immersed. Conidiophores mononematous, branched, erect, straight to flexuous, thick-walled, septate, verrucose, dark brown, 70-102 x 3.4-5.7 μm. Conidiogenous cells polyblastic, discrete, terminal or intercalary, flask shaped, wider and darker at the base, hyaline and denticulate at the tip, 6.5-17 x 3-4.3 μm; denticles cylindrical, up to 2.5 long. Conidia solitary, dry, helicoid, with one coil, hyaline, smooth, multiseptate, rounded and coiled in at the tip, narrow and truncate at the base, 15-23 x 4-5 μm wide.

Holotype: On decaying leaf litter of *F. benghalensis*; Verna, and GU Campus, Taleigao, Goa; 17/10/1998; Miriam, J.; GUFCC No.189 (Herb. slide). Isolated by moist chamber incubation.

The monotypic genus *Moorella* Rao et Rao, typified by *M. speciosa* Rao et Rao, is characterised by conidiophores with verticillate branching, polyblastic conidiogeous cells and helicoid conidia developing on conspicuous denticles. The new species recognised here differs from the type species by flask-shaped

conidiogenous cells developing randomly on the conidiophores. The conidia in both species are similar.

Mucor javanicus Wehmer (Ref: Gilman, 1957. A Manual of Soil Fungi, 33) (Fig. 97)

Colonies on MEA slow growing, with irregular margin, dusty green, 1.5 cm diam in 7 days, reverse of the colony with dusty green roots. *Mycelium* thin, with nonseptate, verrucose, hyaline, up to 1.5 μm wide hyphae. *Sporangiophores* solitary, erect, branched, hyaline, slightly verrucose, 55-65 x 1.5-2 μm. *Sporangia* solitary, spherical, hyaline, verrucose, sometimes with dome-shaped columella, dehiscing vertically, 6-10 μm diam. *Sporangiospores* numerous, hyaline, smooth, reniform, 2.6-4.3 x 1-1.7 μm.

Specimen examined: On soil; Verna, Goa; 14/11/1997; Miriam, J.; GUFCC No. 588 (Live culture); 190 (Herb. slide). Isolated by particle plating.

Mucor silvaticus Hagem (Ref: Gilman, 1957. A Manual of Soil Fungi, 33) (Fig. 98)

Colonies on MEA white, irregular, 2.3 cm diam in 7 days; reverse white. Mycelium thin, with non-septate, smooth, hyaline, 4 μm wide hyphae. Sporangiophores solitary, sometimes radiately branching at the apex, hyaline, smooth, up to 120 μm long, 2.3-4.3 μm wide. Sporangia spherical, hyaline, smooth, dehiscing at the apex, 7-10 μm diam. Spores cylindrical to ellipsoidal, with truncate ends, hyaline, smooth, 3.5-4.4 x 1.5- 2.2 μm.

Specimen examined: On leaf-litter of *C. congesta*; Verna, Goa; 13/2/1998; Miriam, J.; GUFCC No.589 (Live culture);191 (Herb. slide); Isolated by particle plating.

Mucor flavus Bainier (Ref: Gilman, 1957. A Manual of Soil Fungi, 33) (Fig. 99)

Colonies on MEA pinkish brown, regular, 2.8 cm diam in 7 days; reverse light brown. Mycelium thin, moderately brown, with non-septate, smooth, 5 μm wide hyphae. Sporangiophores solitary, unbranched, light brown, smooth, up to 180 μm long, 4-8 μm wide. Sporangia elongate-spherical, light pinkish brown, smooth, 30-32 μm diam.; columella dome-shaped, pinkish. Spores ellipsoidal, ovate, smooth, light pink, 9-13 x 4-7 μm.

Specimen examined: On leaf-litter of *C. congesta*; Verna, Goa; 18/8/1998; Miriam, J.; GUFCC No.590 (Live culture); 192 (Herb. slide); Isolated by particle plating and moist chamber incubation.

Mycovellosiella perfoliata (P. Henn.) Rangel ex Trotter, 1931. In Syll.Fung. 25: 942 (Fig. 100)

Colonies on MEA effuse, olivaceous to dark brown, 2 cm diam in 7 days, reverse brown. *Mycelium* brown, with thin, smooth, septate, branched, pale brown, 1.5 μm wide hyphae. *Conidiophores* mononematous, straight to flexuous, arising in sparse groups, unbranched, moderately dark brown, up to 70 μm long, 3-5 μm wide, smooth septate bearing numerous scars. *Conidiogenous cells* polyblastic, integrated, terminal and intercallary, sympodial, cicatrized scars usually prominent. Ramoconidia cylindrical, elongate, smooth, not septate, moderately brown, variable in shape, with cicatrized scars, 12.6-22.2 x 4.8-5.6 μm. *Conidia* solitary, acropleurogenous, simple, elongated, rounded at the tip, triovalate at the base, constricted in the middle, 1-septate, smooth, moderately brown, 12.6-16.5 x 3.4-4.3 μm.

Specimen examined: On leaf-litter of *F. benghalensis*; Verna, Goa; 10/8/1998; Miriam, J.; GUFCC No.591 (Live culture); 193 (slide); Isolated by particle plating.

Neocercosporella Miriam et Bhat Gen nov.

Colonies irregular, with reddish patches, dark brown. Mycelium moderate, white. Conidiophores smooth, branched, straight, septate, very pale brown. Conidiogenous cells mono- to polyblastic, integrated, determinate, intercalary, cylindrical to club shaped. Conidia solitary, simple, elongated, vermiform, curved, smooth, hyaline, thick-walled, septate.

Type species: Neocercosporella indica Miriam et Bhat sp. nov.

The closely related genus *Cercosporella* Sacc. has conidiophores with long, denticulate, conidiogenous cells although the conidia in both genera are similar (Carmichael et al. 1980). The knob-like conidiogenous loci are very characteristic of the newly recognised taxon *Neocercosporella*.

Neocercosporella indica Miriam et Bhat sp. nov.

(Fig. 101)

Colonies on MEA, irregular, shows some irregular reddish patches, dark brown, media 1.1 cm diam in 7 days, reverse of the colony dark brown. *Mycelium* moderate, white with smooth, thin walled, septate, branched, hyaline, 1.0 μm wide hyphae, 1.739 - 3.26 μm wide. *Conidiophores* smooth, branched, straight, septate, very pale brown. *Conidiogenous cells* monoblastic, integrated, determinate, cylindrical to club shaped, 0.8695 - 9.130 x 1.304 - 3.478 μm. *Conidia* solitary, simple, elongate but curved at the centre, smooth, hyaline, 3-septate, 36.0869 - 65.6521 x 2.1739 - 2.6086 μm.

Specimen examined: On leaf-litter of *C. congesta*; Verna, Goa; 15/9/1997; Miriam, J.; GUFCC No.592 (Live culture); 194 (Herb. slide); Isolated by particle plating.

Colonies on MEA effuse, with regular margin, light green, with central 1.2 cm diam. slightly darker greyish green, with faintly concentric zones, 2.0 cm diam. in 7 days, with scattered sporodochia in groups of 2-3. Mycelium thin, with smooth, thinwalled, septate, branched, hyaline, 1.5 μm wide hyphae; stroma golden brown. Setae simple, long, straight to inwardly curved, broader at the base, pointed to rounded at the apex, septate, very dark brown, smooth, up to 170 μm long, 3-12.5 μm wide at the base, 1.5-3 μm at the tip. Conidiophores sporodchial, compactly arranged, smooth, septate, 1-1.5 μm wide. Conidiogenous cells monophialidic, vase-like, pyriform to vermiform, straight to curved, integrated, terminal, thick-walled, smooth, golden brown, 21-57μm long, 2.5μm at the base, 1.5μm at the tip, with an indistinct collarette, constricted at the narrow neck region. Conidia solitary, curved, sickle shaped, pointed at both ends, hyaline, thin-walled, smooth, 12.5-16 x 2-3 μm, with one 2.5-3.7 μm long setula each on either end.

f.

Specimen examined: On leaf litter of *F. benghalensis*; Verna, Goa; 13/5/1997; Miriam, J.; GUFCC No.593 (Live culture);195 (Herb. slide); Isolated by particle plating.

Nigrospora endophytica Miriam et Bhat sp. nov. (Fig. 103)

Colonies on MEA white, later becoming cream-coloured, regular, granular, 3.5 cm diam in 7 days; reverse white. Mycelium moderate, white, interspersed with black spores, with thin-walled, hyaline, gutulate, septate, smooth, 1.5-2.5 µm wide hyphae. Conidiophores mononematous, branched, straight to flexuous, hyaline, smooth, 4-6 µm thick. Conidiogenous cells monoblastic, discrete, determinate, ampulliform, subspherical, hyaline, gutulate. Conidia solitary, oval to spherical,

compressed dorsiventrally, black, smooth, 0-septate, 14-20 μ m, with a conspicuous groove along the long axis.

Holotype: On fresh leaf, leaf litter and soil of *C. congesta*; Verna, Goa; 23/11/1998; Miriam, J.; GUFCC No.594 (Live culture); 196 (Herb. slide); Isolated by particle plating.

None of the so far described species of the genus *Nigrospora* Zimmermann, typified by *N. panici* Zimm, have grooved conidia. In all species, the conidia are smooth.

Nigrospora sphaerica (Sacc.) Mason, 1927. Trans. Br. Mycol. Soc., 12:158 (Fig. 104)

Colonies on MEA, white becoming greenish, regular, granular, 5.3 cm diam in 7 days; reverse greenish. *Mycelium* moderate, appears as waves, white, very loosely interspersed with black slimy spores spread over the colony in bunches, with thinwalled, hyaline to pale brown, septate, smooth, 1.5-2μm wide hyphae. *Conidiophores* mononematous, branched, straight to flexuous, hyaline to pale brown, smooth, 4.3-4.8 μm thick. *Conidiogenous cells* monoblastic, discrete, solitary, determinate, ampulliform, subspherical, colourless. *Conidia* solitary, spherical, ellipsoidal, compressed dorsiventrally, black, shiny, smooth, 0-septate, 14-20 μm.

Specimen examined: On leaf-litter of *C. congesta* and soil; Verna, Goa; 17/2/1997; Miriam, J.; GUFCC No.595 (Live culture); 197 (Herb. slide); Isolated by particle plating.

Nodulisporium honiaraense Matsushima 1971. Microfungi of the Solomon Islands & Papua-New Guinea, p.40. (Fig. 105)

Colonies on MEA, brown, irregular, 1.2 cm diam in 7 days; reverse brownish grey. Mycelium moderate, with thin-walled, smooth, septate, branched, hyaline, 1.5 µm wide hyphae. Conidiophores mononematous, branched, brown, slightly

verrucose, decreasing in colour intensity towards apex, erect, straight to flexuous, 125-200 μm long, 2.6-4 μm wide; each branch is pencillalety branched towards the apex; terminal branches 12-16 x 2-3 μm. *Conidiogenous cells* polyblastic, integrated, terminal, becoming intercalary or discrete, cylindrical to clavate, denticulate; denticles conspicuous, short, fragile. *Conidia* solitary, acropleurogenous, simple, elliptical, obovoid, 0-septate, hyaline, faintly smooth, 2.5-3.7 x 1.2-2.5 μm.

Specimen examined: On fresh leaf and leaf-litter of *F. benghalensis*; Verna, Goa; 10/8/1998; Miriam,J.; GUFCC No.596 (Live culture); 198 (slide); Isolated by particle plating and moist chamber incubation..

Paecilomyces Bainier

Colony effuse, light brown, visible after 7 days, granular. Mycelium moderate, light brown, with thin-walled, smooth, septate, branched hyphae. Conidiophores mononematous, straight to flexuous, branched, septate, light brown, smooth. Conidiogenous cells phialidic, integrated, terminal, intercalary, cylindrical. Conidia catenate, aseptate, ellipsoidal with rounded apex and truncate base, coloured.

The three isolates recovered during the study are recognised as separate species below

Paecilomyces	Colony	Conidiophore	Conidia	Habita	t/Locality/	
sp.	diam. in	in μm.	in μm.	Substrate/GUFCC		
	cm			No.		
1	1.8, pale	7.8-25 x 1.3-1.7	3.9-4.8 x 2.24-2.8,	Leaf	199	106
	brown		ellipsoidal, brown	litter		
2	1.1, pale	3.7-74 x 1.5-2.5	spherical, brown,	Air	200	107
	brown		verrucose, 2.5-3.13			
3	1.3,	8.7-25 x 1.3-1.74	ellipsoidal, hyaline,	Soil	201	108
	white		1.3-2.6 x			
			2.2-3.5			

Paracylindrocladia Miriam et Bhat Gen. et sp. nov.

Colonies on MEA golden brown, effuse. Mycelium pale brown, with thin-walled, septate, branched, hyphae. Conidiophores mononematous, branched, with a septate, smooth, sterile stipe terminating in a bulbose tip. Conidiogenous cells polyblastic, integrated, determinate, terminal. Conidia solitary, ellipsoidal, simple.

Type species: Paracylindrocladia indica

Presence of a sterile stipe along with the conidiogenous cells is a common feature in genera such as *Cylindrocladium* Morgan, Gyrothrix Corda, *Circinotrichum* Nees and *Zygosporium* Mont. (Ellis, 1971). In *Gyrothrix* and *Circinotrichum, the* stipe is setiferous and distinct from the conidiogenous cells whereas in *Cylindrocladium* and *Zygosporium* the stipe arises from the base of conidiogenous cells. In other words, one of the conidiogenous cells becomes stiff, extended, bulbose at the tip and remains sterile. The genus *Cylindrocladium* is moniliaceous and phialidic whereas *Paracylindrocladia* is dematiaceous and blastic. The genus *Zygosporium* distinguishes itself from the new taxon by its conspicuous curved and swollen vesicle.

Paracylindrocladia indica Miriam et Bhat Gen. et sp. nov. (Fig. 179)

Colonies on MEA golden brown, effuse, wavy, 2.6 cm diam in 7 days, reverse light brown. Mycelium moderate, brown with thin-walled, septate, branched, 1.5-2.5 μ m wide hyphae. Conidiophores mononematous, branched, with a septate, smooth, 70-100 x 3-4 μ m sterile stipe, dark brown at the base, hyaline at the tip, terminating in

a bulbose tip up to 12 μ m diam. *Conidiogenous cells* polyblastic, integrated or discrete, determinate, terminal, cylindrical, denticulate. *Conidia* solitary, ellipsoidal to rounded, simple, aseptate, smooth, pale brown, 4-6 x 3-4 μ m.

Holotype: On leaf-litter of *C. congesta*; Verna, Goa; 22/11/1997; Miriam, J.; GUFCC No.599 (Live culture); 202 (Herb. slide); Isolated by particle plating.

Parahumicola Miriam et Bhat Gen. et sp. nov.

Colonies on MEA dark brown, effuse, regular, brown. Mycelium moderate with thin-walled, light brown, smooth, septate, branched. Conidiophores mononematous, erect, straight to flexuous, branched, sometimes swollen, at random points, light brown, septate, smooth. Conidiogenous cells terminal, intercalary, integrated, blastic, cylindrical. Conidia catenate, dark brown, thick-walled, smooth, variable in shape.

Type species: Parahumicola endophytica

The closely related genera such as *Humicola* Traaen and *Chlamydomyces*Bainier are characterized by thick-walled, dark, aseptate, blastoconidia and simple condiogenous cells. The new genus and species described here is very much similar to these two genera but differ in that the conidia are highly variable in size and shape. Besides it is an endophytic fungus.

Parahumicola Miriam et Bhat Gen. et sp. nov.

(Fig. 172)

Colonies on MEA dark brown, effuse, regular, 1.3 cm diam. in 7 days, reverse dark brown. *Mycelium* moderate with thin-walled, light brown, smooth, septate, branched, 1.7-2.6 µm wide hyphae. *Conidiophores* mononematous, erect, straight to flexuous, branched, sometimes swollen, light brown, septate, smooth, sometimes the

base of the conidiophore covered very densely by slime, $10-45 \times 1.5-6 \mu m$. Conidiogenous cells terminal, intercalary, integrated, blastic, cylindrical. Conidia solitary or catenate, dark brown, thick-walled, smooth, variable in shape, spherical, oblong, elongate, $11-33 \times 5-8-13.5 \mu m$.

Holotype: On internal leaf tissues of *C. congesta*; Verna, Goa; 12/12/1998; Miriam, J.; GUFCC No.600 (Live culture); 203 (Herb. slide); Isolated by particle plating.

Periconiella musae M.B. Ellis, 1967. Mycol. Pap. 117: 5-7 (Fig. 109)

Colonies on MEA effuse, flat, with irregular margin, light grey, 1 cm diam. in 7 days; reverse very light brown. *Mycelium* thin, loosely arranged, raised with the conidiophores and spores, with smooth, thin-walled, septate, branched, pale brown, 1-2 μm wide hyphae. *Conidiophores* mononematous, smooth, unbranched, straight, with a single septum at the base, brown, 21-46.5 μm long, 2-3.5 μm wide. *Conidiogenous cells* polyblastic, integrated, terminal, determinate, with cicatrized thick circular scars. *Conidia* solitary, simple, elliptical, slightly narrower at the base, rounded at the tip, 5-10 x 2-4 μm.

Specimen examined: On leaf-litter of *F. benghalensis*; Verna, Goa; 10/8/1998; Miriam, J.; GUFCC No.601(Live culture); 204 (slide); Isolated by particle plating.

Periconiella smilacis M.B. Ellis, 1967. (Ref: Ellis, 1971. Dematiaceous

Hyphomycetes, p. 299-301) (Fig. 110)

Colonies effuse, hairy, light browni. Mycelium partly immersed, thin, faintly verrucose, septate, branched, light brown, 1.5-2 µm wide. Conidiophores

mononematous, faintly verruculose, septate, branched, erect, straight to flexuous, hyaline, 18-137 x 2-4 μm. *Conidiogenous cells* polyblastic, integrated, terminal, denticulate at the tip. *Conidia* solitary, obclavate to elliptical, rounded at the apex, truncate or pointed at the base, 0-septate, smooth, hyaline, 2-2.5 x 2.5 μm.

Specimen examined: On leaf litter of *F.benghalensis*; Verna, Goa; 15/7/1997; Miriam, J.; GUFCC No.205 (Herb. specimen/slide); Isolated by Moist chamber incubation method.

Penicillium Link (Ref: Carmichael et al., 1980; Ellis, 1971)

Type species: P. expansum Link. Three species of the genus *Penicillium* are distinguished here based on colony morphology and microscopic characters.

Colonies effuse, coloured, dry, powdery, growth visible in 7 days, Conidiophores mononematous, straight to flexuous, branched, septate. Conidiogenous cells phialidic, penicillus, discrete, terminal, cylindrical, ampullate. Conidia catenate, coloured, spherical.

Penicillium	Colony	Conidiophore	Ampullae	Conidia	Habitat/Locality/	Fig. No.
spp.	diam. in cm	in μm.	in μm.	in μm.	Substrate	
	light green, 5.5, irregular	branched, usually three at the tip, 1.5-2.2 x 11-110	4-6, 6-10 x 2-2.5	1.5-2.5, green spherical	soil	111
2	blue to grey,	12.5-100 x 1.5	6-7.5 x 1.5	1.5-2.5, hyaline	Soil	112
3	light green,	64-245 x 1.5-2.5	12.5-16.5 x 1.5-2.5	3-4 x 2-3, elliposidal, hyaline	Air	113

Periconia byssoides Pers. ex. Merat, 1821, Nouv. Fl. Environs Paris, Ed.2, 1: 18-19

(Fig. 114)

Conidiophores 60-90μm long, 2.5-3μm at base, 5.2μm immediately below head, subhyaline apical cell, 6.5-5.2μm., smooth, dark brown, septate, thick-walled. Conidiogenous cells, polyblastic, integrated, terminal. Conidia spherical, dark brown, verrucose, 3.5-5.2μm diam.

Specimen examined: On leaf-litter of *C. congesta*; Verna, Goa; 11/7/1997; Miriam, J.; GUFCC No.605 (Live culture); 209 (Herb. slide); Isolated by particle plating.

Periconia lateralis Ellis and Everh., 1886, J. Mycol., 2:104 (Fig. 115)

Colonies effuse, hairy, dark brown to black. Mycelium immersed. Conidiophores often curved, dark brown, 180-200 μm long, 4-5μm at the base, 4.5μm just above the basal swelling, tapering to 1.5 μm at apex. Conidiogenous cells polyblastic, integrated, intercalary, borne directly on the stipe and on unilateral branches which sometimes have sterile setiform apices. Conidia verrucose, spherical, dark brown, 6-8μm diam.

Specimen examined: On leaf-litter of *C. congesta*; Verna, Goa; 23/6/1997; Miriam, J.; GUFCC No.210 (Herb. slide); Isolated by moist chamber incubation.

Periconia saraswatipurensis Bilgrami, 1963. Sci & Cult., 29: 48 (Fig. 116)

Colony effuse, dark brown, granular, slimy, regular, 1.2cm diam in 7 days, reverse of colony brown. *Mycelium* moderate, brown, with thin-walled, smooth, septate, branched 2.6-3.4μm wide hyphae. *Conidiophores* 57-130μm long, 1.3-2.2μm at base, dark brown, septate, thick-walled, verrucose. *Conidiogenous cells*,

polyblastic, *integrated*, terminal, intercalary. *Conidia* spherical,dark brown, verrucose, 8-12µm diam.

Specimen examined: On leaf-litter of *C. congesta*; Verna, Goa; 24/12/1998; Miriam, J.; GUFCC No.606 (Live culture); 211(Herb. slide); Isolated by particle plating.

Phaeotrichoconis crotalariae (Salam et Rao) Subram. 1956. Proc. Indian Acad. Sci. Sect.B. 44: 2 (Fig. 117)

Colonies on MEA, grey brown, irregular, 1.9cm diam in 7 days. reverse light green brown, *Mycelium* with smooth, white, thin-walled, septate, branched, hyaline, 1.7-2.2 μm wide hyphae. *Conidiophores* mononematous, straight to flexuous, unbranched, sometimes geniculate, brown, smooth, 72-109 x 3.04-6.0 μm wide. *Conidiogenous cells* polytretic, integrated, terminal, intercalary, sympodial, cicatrized, scars large, black, cylindrical. *Conidia* solitary, dry, elongated, obclavate, transversely septate, golden brown, thick-walled, smooth, large brown scars at the base, with a long narrow hyaline beak at the tip, 37.4-68 μm long, 5-6μm at the broadest part, and beak 13-45 x 0.43-0.9μm.

Specimen examined: On leaf-litter of *C. congesta*; Verna, Goa; 20/11/1997; Miriam, J.; GUFCC No.607 (Live culture); 212 (Herb. slide); Isolated by particle plating.

Phialocephala carrisseae Miriam et Bhat sp. nov. (Fig. 118)

Colonies effuse, cottony, dark brown. Conidiophores mononematous, smooth, septate, unbranched at the base, thickwalled, very dark brown, straight, with a complex head made up of several series of branches at the terminal region that decrease in colour intensity towards the tip, the ultimate ones bearing the conidiogenous cells surrounded by 5-6 sterile hairs; basal stipe region 100-450 µm

long and 9.5-15.5 μ m wide at the base, tapering to 6-10 μ m wide at the tip. Conidiogenous cells monophialidic, cylindrical, penicillately arranged, hyaline, smooth. 8.5-10 x 2-2.5 μ m; sterile hairs, hyaline, smooth, 310-337 x 12.5 μ m. Conidia slimy, ellipsoidal to oval, with rounded ends, hyaline, smooth, 0-septate, 3.5-5 x 2.5 μ m.

Holotype: On leaf-litter of *C. congesta* Verna, Goa; 28/6/97; Miriam, J.; GUFCC No.213 (herb. specimen/slide); isolated from moist chamber incubation.

The fungus differs from all the known species of the genus *Phialocephala* Kendrick (Carmichael et al. 1980), typified by *P. dimorphospora* Kendrick, with some of its conidiogenous metulae terminating into sterile extensions.

Phialocephala nephrospora Miriam et Bhat sp. nov.

(Fig. 119)

Colonies on MEA effuse, light brown to pink, granular, 1.8 cm diam. in 7 days, reverse light pink brown. *Mycelium* thin, with thin-walled, hyaline, septate, branched, smooth, 1.5 um diam. hyphae. *Conidiophores* mononematous, straight to flexuous, branched, hyaline, upto 82 μm long, 2-3.58 μm wide. *Conidiogenous cells* phialidic, terminal, discrete, ampulliform, hyaline, smooth, 8.7-25 x 1.5-2 μm. *Conidia* solitary, hyaline, smooth, reniform, light pinkish brown, 7-8.3 x 2.6-4 μm.

Holotype: On leaf litter of C. congesta; Taleigao, Goa; 23/11/91997; Miriam, J GUFCC No.608 (Live culture); 214 (Herb. slide); Isolated by particle plating and moist chamber incubation method.

Phialocephala Kendrick, typified by P. dimorphospora Kendrick, has 15 species so far known in the genus (Hawksworth et al., 1995). In all these, the conidia in general are cylindrical to ovate and P. nephrospora is distinct with its reniform conidia.

Phialocephala xalepensis Maggi et Persiani, 1984. Mycologia 20: 253 (Fig. 120)

Colonies effuse, dark brown. Conidiophores mononematous, dark brown, smooth, septate, unbranched at the base, thickwalled, erect, with a complex head made up of several series of branches, the ultimate ones bearing the conidiogenous cells; basal stipe region 185-500 x 3-10 μm. Conidiogenous cells monophialidic, cylindrical, discrete, penicillately arranged, determinate, light brown, smooth, 4-9 x 1.5 μm. Conidia ellipsoidal to oval, with rounded ends, light brown, smooth, aggregate in slimy heads, 0-septate, 2.5-3 x 1.5-2 μm.

Specimen examined: On leaf-litter of *C. congesta* Verna, Goa; 14/7/98; Miriam, J.; GUFCC No.609 (Live culture); 215 (Herb. slide); Isolated by particle plating.

Phialomyces microsporus Miriam et Bhat sp. nov.

(Fig. 121)

Colonies on MEA, effuse, greenish to grey at the centre, remaining part cream-coloured, with concentric zonations, which appear like clouds, 0.2 cm diam. in 7 days, reverse central green with off-white/cream edge. *Mycelium* thin, white, interspersed by dark brown bunches of conidia with thin walled, hyaline, smooth, branched, septate, 2-2.6 μm wide hyphae. *Conidiophores* mononematous, staright to flexuous, branched, smooth, pale brown, cylindrical, sometimes bulging at its tip, 2.6-6.5 x 1.5-3 μm. *Conidiogenous cells* phialidic, integrated, light brown, smooth. *Conidia* catenate, ellipsoidal to ovate, smooth, aseptate, in masses green to brown, 2.2-6 x 1.7-3.5 μm.

Holotype: On leaf litter of *C. congesta*; Taleigao, Goa; 13/11/91997; Miriam, J.; GUFCC No.610 (Live culture); 216 (Herb. slide); Isolated by particle plating and moist chamber incubation method.

The monotypic genus *Phialomyces* Misra et Talbot, typified by *P. macrosporus* Misra et Talbot, has catenate, verrucose, limoniform conidia which are

22-27 x 16-20 μm in the type species. The conidia in *P. microsporus* are smooth, ovate and 2.2-6 x 1.7-3.5 μm .

Pithomyces chartarum (Berk. et Curt.)M.B. Ellis, 1960. Mycol Pap. 76:13. (Fig. 122)

Colonies effuse, cottony, circular, with smooth margin, with granular tiny grape-like bunches on the surface, light to dark brown, 3-3.5 cm diam. in 7days; reverse of the colony light yellow brown at centre. *Mycelium* moderate, olivaceous brown, interspersed with black clusters of raised spore masses, with smooth, thinwalled, septate, branched, hyaline, 1.5 μm wide hyphae. *Conidiophores* mononematous, straight to flexous, smooth, unbranched, light brown, up to 12 um long, 2-2.5 um wide. *Conidiogenous cells* monoblastic, integrated, cylindrical, terminal or intercalary, determinate, denticulate. *Conidia* dry, solitary, simple, clavate, elongated, with rounded ends, obovoid, echinulate, dark brown, 3-4 transverse septate, with 1-2 oblique septa, 10.5-20 x 7-9 μm, with upper part of the denticle remain attached to the base of the conidium.

Specimen examined: On latex of fresh leaf and on leaf litter of *F. benghalensis* and from air and soil, Verna, and from air and soil, GU Campus, Taleigao, Goa; 26/12/98; Miriam,J.; GUFCC No.611 (Live culture on MEA); 217 (Herb. specimen/slide); isolated by particle plating.

Pithomyces graminicola R.Y. Roy et Rai, 1968. Trans. Br. mycol. Soc. 51: 154-155 (Fig. 123)

Colonies on MEA effuse, with wavy margin, light brown, with dark brown to black spores in scattered masses, sparsely granular, 4.2cm diam in 7days; reverse of the colony dark brown with occasional thin black strands. *Mycelium* moderate, light brown, profuse at centre of colony, thinning out towards the periphery, smooth, with

thin-walled, septate, branched, hyaline, 1.5 μm wide hyphae. *Conidiophores* mononematous, straight to flexous, light brown, smooth, unbranched, septate, up to 20 μm long. *Conidiogenous cells* monoblastic, integrated, terminal or intercalary, determinate, cylindrical, denticulate, denticles cylindrical, sometimes up to 1 μm long. *Conidia* solitary, dry, simple, elliptical, clavate, obovoid, faintly echinulate, dark brown, 2-3-transverse septate, sometimes with one oblique septa, 9-18 x 4.5-12 μm, part of the denticle often remain attached to the base of the conidium.

Specimen examined: On leaf-litter of *C. congesta*, GU Campus, Taleigao, Goa; 12/7/98, Miriam, J.; GUFCC No.612 (culture on MEA); 218 (slide); Isolated by particle plating.

Polyshema clavulata M.B. Ellis, 1975. More Dematiaceous Hyphomycetes: 370-371.

(Fig. 124)

Colonies on MEA, brown yellow, regular, granular, 1.2cm diam. in 7 days, reverse light brown. Mycelium thin, with thin-walled, septate, branched, hyaline, 0.9-1.3 μm wide hyphae. Conidiophores mononematous, straight to flexuous, unbranched, brown, smooth, septate, 0.4-26 x 0.7-0.9 μm wide. Conidiogenous cells blastic, integrated, terminal, cylindrical.Conidia solitary, dry, elongate, ellipsoidal, 4-5 septate, basal cell hyaline, verrucose, brown, 12-20 x 4-5.2μm.

Specimen examined: On leaf-litter of *C. congesta*; Verna, Goa; 14/6/1997; Miriam, J.; GUFCC No.613 (Live culture); 219 (Herb. slide); Isolated by particle plating.

Pseudobotrytis terrestris (Timonin) Subram., 1956. J. Indian Bot. Sci. 35:86.

(Fig. 125)

Colonies circular, with irregular margin, granular, faintly concentric, dusty brown, 2.5 cm diam. in 7 days; reverse of the colony light brown. Mycelium very thin,

with smooth, thin-walled, septate, branched, moderately dark brown, 1.5-2.5 μm wide hyphae. *Conidiophores* mononematous, straight or flexuous, erect, smooth, septate, unbranched, dark brown, medium brown to hyaline at the tip, 100-210 μm long and 3-4 μm wide at the base, terminating in verticillately branched fertile apex. *Conidiogenous cells* polyblastic, discrete, hyaline, smooth, rounded at the base, tapering to 2.5 μm wide, minutely denticulate tip, developing in whorls on 12.5-17.5 x 2.5 μm branches. *Conidia* dry, oval, rounded at the tip, slightly pointed at the base, moderately brown, smooth, 5-7 x 2.5-3 μm.

Specimen examined: On leaf litter of *C. congesta* and in soil, Verna, Goa; 29/7/98, Miriam, J.; GUFCC No.614 (Live culture on MEA), 220 (Herb. specimen/slide); Isolated by particle plating.

Ramichloridium fasciculatum Vasant Rao et de Hoog, 1986. Stud. Mycol. 28: 39-41 (Fig. 126)

Colonies on MEA effuse, cottony, with irregular margin, light grey to light brown, reverse dark green with creamish margin, 0.5 cm diam. in 7 days. Mycelium moderate, slightly raised, with smooth, thin-walled, septate, branched, light brown, 1.5 μm wide hyphae. Conidiophores mononematous, sometimes in fascicles, straight to flexuous, unbranched, 2-3-septate, smooth, 35-105 μm long, 2-4 μm wide at the base, 1-2 um wide above half. Conidiogenous cells polyblastic, with thin cicatrized scars. Conidia ellipsoidal, smooth to slightly verrucose, hyaline to pale brown, 3-8 x 2.5-3.5 μm.

Specimen examined: On leaf litter of F. bengalensis, Verna, Goa; 14/9/1997; Miriam, J.; GUFCC No. 615 (Live culture); 221 (slide); Isolated by particle plating.

Rhinocladiella cellaris (Pers. ex S.F. Gray) M.B. Ellis, 1971. Dematiaceous
Hyphomycetes: 247-248 (Fig. 127)

Colonies on MEA, effuse, brown, velvety, regular, granular, 2.4cm diam in 7 days, reverse of colony brown. *Mycelium* moderate, brown, with thin hyaline, smooth, branched, septate, 1.2-20 wide hyphae. *Conidiophores* mononematous, unbranched, straight to flexuous, usually geniculateor curved at the tip, light brown, smooth, septate, 3-54 x 0.9-2.2μm. *Conidiogenous cells* polyblastic, integrated, terminal. *Conidia* solitary, arranged cyclically, ellipsoidal with rounded tip and pointed base, smooth, light brown, 3-8.3 x 0.9-2.2μm.

Specimen examined: On leaf-litter of *C. congesta*; Verna, Goa; 19/12/1998; Miriam, J.; GUFCC No.616 (Live culture); 222 (Herb. slide); Isolated by particle plating.

Septonema secedens Corda 1837. (Ref: Ellis, 1971. Dematiaceous Hyphomycetes)

(Fig. 183)

Colonies on MEA effuse, dark brown, velvety, with mycelium in waves interspersed by dark brown spores, 2.1cm diam. in 7 days. *Mycelium* moderate, light brown, with thin-walled, smooth, septate, 1.5-2.5 μm wide hyphae. *Conidiophores* mononematous, straight to flexuous, brown, smooth, septate, sometimes swollen at the septa, 250-350 x 4-6 μm. *Conidiogenous cells* polyblastic, integrated, terminal, intercalary, arising in a whorl. *Conidia* catenate, cylindrical with rounded ends, 3-4-septate, brown, verrucose, 15-41 x 0.45-0.6 μm.

Specimen examined: On leaf litter of *F. benghalensis* Verna, Goa; 12/12/98, Miriam, J.; GUFCC No.658 (Live culture on MEA), 275 (Herb. specimen/slide); Isolated by particle plating.

Conidial fungi, hyphomycetes. *Colonies* effuse, dark brown, hairy. *Mycelium* partly immersed, composed of sparce, branched, septate, pale brown hyphae 2-2.5 μm wide. *Conidiophores* synnematous, distinct, erect, flexuous, septate, branched, smooth, 2-3.5 μm wide; synnema erect, compacted and dark brown in below half, splayed apart and pale brown in above half, upto 260 μm long, 10-15 μm wide in below half, up to 50 μm in above half. *Conidiogenous cells* polyblastic, integrated, terminal or intercalary, sympodial, cylindrical, thick-walled, smooth, pale brown, with inconspicuous denticles. *Conidia* solitary, dry, ovoid, rounded at both ends, colourless to pale brown, aseptate, smooth, 4-5 x 2-3 μm.

With its compactly arranged, synnematous conidiophores, polyblastic, sympodial and integrated conidiogenous cells and solitary, dry conidia, the fungus described above can be accommodated in the genus *Sclerographium* Berkeley (Ellis, 1971). The type species, *S. aterrimum* Berk. Was described on leaves of *Indigofera* from India. The only other species, *S. phyllanthicola* Deighton was described on *Phyllanthus* from Siera Leone. Both these produce phragmoseptate and verrucose conidia whereas in *S. goanensis* has aseptate and smooth conidia.

Hollotype: From dead and decaying twigs of *Ficus benghalensis* L.; Verna, Goa; 14/11/1997; Miriam, J.; GUFCC No. 273 (Herb. slide); Isolated by moist chamber incubation.

Scolecobasidium constrictum Abbott, 1927. Mycologia 19: 29-31 (Fig. 128)

Colonies effuse, flat, radialy concentric, with wavy margin, forming a soft cushion like mass in the centre, dark brown, 2.0 cm diam. in 7 days reverse dark brown in the centre. *Mycelium* moderate, dense at center, thinner towards the periphery, with smooth, septate, branched, light brown, 1-2 µm wide hyphae.

Conidiophores mononematous, smooth, unbranched, straight to flexous, 4-9 μ m long, 1.5-4 μ m wide. Conidiogenous cells polyblastic, sympodial, cylindrical, discrete, terminal and intercalary, denticulate; denticles cylindrical, up to 2 μ m long. Conidia solitary, dry, elongate, tapering at the base, rounded at the tip, 1-septate, simple, slightly verrucose, slightly constricted at the septum, golden brown, 8.7-11.7 x 2.6-4.5 μ m.

On fresh leaf of *C. congesta*; Taleigao, Goa; 25/3/1997; Miriam, J.; GUFCC No. 617 (Live culture); 223 (Herb. slide); Isolated by particle plating.

Scolecobasidium humicola Barron et Busch, 1962, Can. J. Bot., 40:83 (Fig. 129)

Colonies effuse, flat, radialy concentric, with wavy margin, forming a soft cushion like mass in the centre, dark brown, 0.7-1 cm diam. in 7 days reverse dark brown in the centre. *Mycelium* moderate, dense at center, thinner towards the periphery, with smooth, septate, branched, light brown, 1-2 μm wide hyphae. *Conidiophores* mononematous, smooth, unbranched, straight to flexous, 13.4-19.6 μm long, 1.3-2 μm wide. *Conidiogenous cells* polyblastic, sympodial, cylindrical, discrete, terminal and intercalary, denticulate; denticles cylindrical, up to 2 μm long. *Conidia* solitary, dry, elongate, tapering at the base, rounded at the tip, 1-septate, simple, verrucose, slightly constricted at the centre, golden brown, 5.6-9.6 x 1.7-3 μm.

Specimen examined: On fresh leaf of *C. congesta*.; Taleigao, Goa; 15/5/1997; Miriam, J.; GUFCC No.618 (Live culture); 224 (Herb. slide); Isolated by particle plating.

Colonies on MEA effuse, flat, circular, with smooth margin, very dark brown, slightly raised with concentric circles a little before the periphery, 0.5-1 cm diam. in 7 days; reverse dark brown. *Mycelium* moderate, with smooth, thin-walled, septate, branched, light brown, 1-1.5 μm wide hyphae. *Conidiophores* mononematous, smooth, branched, straight to flexuous, medium brown, 2-13 x 1.5-2.6 μm. *Conidiogenous cells* polyblastic, sympodial, cylindrical, terminal, sometimes discrete, denticulate; denticles cylindrical, up to 2 μm long. *Conidia* solitary, dry, cuneiform to turbinate, 1-septate, slightly constricted at the septum, verrucose, sometimes part of the denticle remains attached to the base, golden brown, 4.3-7.8 x 2.6 μm.

Holotype: On fresh leaf of *F. benghalensis*; Taleigao, Goa; 23/6/1997; Miriam, J.; GUFCC No.619 (Live culture); 225 (Herb. slide); Isolated by particle plating.

Of the many species so far described in the genus *Scolecobasidium* Abbott, typified by *S. terreum* Abbott, only the type species shows some similarity with *S. triangularis* which has Y of T-shaped conidia. The conidia of *S. triangularis* are cuneiform or turbinate and quite distinct from those of *S. terreum*.

Scolecobasidium variabile Barron et Busch, 1962, Can. J. Bot., 40:83-84 (Fig. 131)

Colonies effuse, flat, radialy concentric, with wavy margin, forming a soft cushion like mass in the centre, dark brown, 2.5 cm diam. in 7 days reverse dark brown in the centre. *Mycelium* moderate, dense at center, thinner towards the periphery, with smooth, septate, branched, light brown, 1-2 µm wide hyphae. *Conidiophores* mononematous, smooth, unbranched, straight to flexous, 13.4-19.6 µm long, 1.3-2 µm wide. *Conidiogenous cells* polyblastic, sympodial, cylindrical, discrete, terminal and intercalary, denticulate; denticles cylindrical, up to 2 µm long.

Conidia solitary, dry, elongate, tapering at the base, rounded at the tip, 1-septate, simple, smooth, slightly constricted at the centre, golden brown, 5.6-9.6 x 1.7-3 µm.

Specimen examined: On fresh leaf of *C. congesta*.; Taleigao, Goa; 18/5/1997; Miriam, J.; GUFCC No.620 (Live culture); 226 (Herb. slide); Isolated by particle plating.

Scolecobasidium verrucosum Roy, Dwivedi et Misra, 1962. Lloydia, 25: 164-166 (Fig. 132)

Colonies effuse, flat, radially concentric, with wavy margin, forming a soft cushion like mass in the centre, dark brown, 2.4 cm diam. in 7 days reverse dark brown in the centre. *Mycelium* moderate, dense at center, thinner towards the periphery, with smooth, septate, branched, light brown, 1.3-2.2 μm wide hyphae. *Conidiophores* mononematous, smooth, unbranched, straight to flexous, 4.4-24.8 x 1.3-2.6 μm. *Conidiogenous cells* polyblastic, sympodial, cylindrical, discrete, terminal and intercalary, denticulate; denticles cylindrical, up to 2 μm long. *Conidia* solitary, dry, elongate, tapering at the base, rounded at the tip, 1-septate, simple, highly verrucose, slightly constricted at the centre, golden brown, 4.8-7.5 x 2.6 μm.

Specimen examined: On fresh leaf of *C. congesta.*; Taleigao, Goa; 23/61997; Miriam, J.; GUFCC No.621 (Live culture); 227 (Herb. slide); Isolated by particle plating and moist chamber incubation.

Scolecobasidium longisporum Matsushima, 1975. Icones Fungorum A Matsushima
Lecturum: 127

(Fig. 133)

Colonies effuse, flat, radialy concentric, with wavy margin, forming a soft cushion like mass in the centre, dark brown, 2.0 cm diam. in 7 days reverse dark brown in the centre. Mycelium moderate, dense at center, thinner towards the

periphery, with smooth, septate, branched, light brown, 1.3-1.74 μm wide hyphae. *Conidiophores* mononematous, smooth, unbranched, straight to flexous, 2.3-20.0 x 1.3-2.2 μm. *Conidiogenous cells* polyblastic, sympodial, cylindrical, discrete, terminal and intercalary, denticulate; denticles cylindrical, up to 2 μm long. *Conidia* solitary, dry, elongate, tapering at the base, rounded at the tip, 1-septate, simple, faintly verrucose, slightly constricted at the centre, hyaline, 9.13-15.7 x 1.3-1.74 μm.

Specimen examined: On fresh leaf and decaying litter of *C. congesta*.; Taleigao, Goa; 21/5/1997; Miriam, J.; GUFCC No. 622 (Live culture); 228 (Herb. slide); Isolated by particle plating and moist chamber incubation.

Scytalidium lignicola Pesante, 1957. Annali Sper. agr., N.S., 11: 241-245 (Fig. 134)

Colonies on MEA effuse, creamish to off-white, irregular, 0.6cm diam. in 7 days, reverse off-white. *Mycelium* moderate, white, as loose shambles all over arranged in an almost radial manner, with thin-walled, septate, hyaline, 1.5-2 μm wide hyphae. *Conidiophores* mononematous, straight to flexuous, unbranched, smooth, hyaline, 1.5-10 x 1.5-2.5 μm. *Conidiogenous cells* monoblastic, terminal, integrated, cylindrical. *Conidia* sometimes solitary, smooth, hyaline, 0-septate, 2-2.5 μm diam, sometimes catenate, cylindrical, smooth, hyaline, aseptate, 2.2-4 x 1.5-2 μm.

Specimen examined: On leaf-litter of *C. congesta*; Verna, Goa; 12/11/1997; Miriam, J.; GUFCC No.623 (Live culture); 229 (Herb. slide); Isolated by particle plating.

Colonies on MEA brown, regular, with irregular margin, velvety, 2.3 cm diam. in 7 days, reverse brown. *Mycelium* moderate brown, with thin-walled, hyaline, smooth, septate, branched, 1.5-2.6 μm wide hyphae. *Conidiophores* mononematous, flexuous, geniculate, septate, unbranched, smooth, brown, 5-70 x 2-3.5 μm. *Conidiogenous cells* polytretic, terminal or intercalry, integrated, with unthickened pores. *Conidia* solitary, obclavate, rounded at the tip, pointed at the base, 1-septate, with a constriction at the septum, pale brown, smooth, 5.2-8.5 x 2.5-3.5 μm.

Holotype: On leaf-litter of *C. congesta*; Verna, Goa; 24/8/1998; Miriam, J.; GUFCC No.624 (Live culture); 230 (Herb. slide); Isolated by particle plating.

Spadicoides bina (Corda) Hughes, 1958. Can.J. Bot., 36: 806 (Fig. 136)
Colonies effuse, dark brown, hairy. Mycelium partly superficial, partly immersed,
2.6086μm wide hyphae, smooth, brown, septate, branched. Conidiophores thick
walled, smooth, septate, brown, mononematous, straight to flexuous, unbranched,
25.21-53.91 x 2.18μm. Conidiogenous cells polytretic, terminal, intercalary,
integrated, cylindrical. Conidia catenate, 0-septate, ellipsoidal, brown, smooth, 3.911.3 x 2.17 -2.8 μm.

Specimen examined: On leaf-litter of *C. congesta*; Verna, Goa; 21/12/1997; Miriam, J.; GUFCC No.231(Herb. slide); Isolated by moist chamber incubation.

Spadicoides Hughes and Diplococcium Grove are closely related and have recently been revised genera (Ellis, 1971, 1976). S. indicus is unique in the genus Spadicoides with its curved conidiophores and pale brown conidia.

Speiropsis pedatospora Tubaki, 1958. J. Hattori bot. Lab. 20: 171-173 (Fig. 137)

Colonies effuse, hairy, dark brown. Mycelium immersed. Conidiophores mononematous, straight to flexuous, dark brown, smooth, thick-walled, septate, with broad holdfast/basal cell. Conidiogenous cells polyblastric, cylindrical to branched, discrete, terminal, intercalary, hyaline, smooth, 1-16 x 2.5 μm. Conidia solitary, multiseptate cells joined by triradiate, filamentous, made of numerous cells joined together, smooth, 60-85 μm long in the longest arm, 28-32 μm long at the shortest arm and 1.5-2.25 μm wide.

Specimen examined: On leaf-litter of C. congesta; Verna, Goa; 22/7/1997; Miriam, J.; GUFCC No.232 (Herb. slide); Isolated by particle plating.

Sporidesmium adscendens Berk. (Ref: M.B.Ellis, 1958. Mycol.Pap. 70: 32) (Fig. 138)

Colony effuse, olive brown to grey, fuscous, hairy. Mycelium partly immersed. Conidiophores mononematous, caespitose, unbranched, straight to flexuous, moderate to dark brown, 9-40 x 5-6 μm. Conidiogenous cells monoblastic, integrated, terminal, determinate, cylindrical, percurrent. Conidia solitary, dry, acrogenous, simple, obclavate to obpyriform, drawn out very narrow and long at the tip, dark brown, smooth, transversely 24-30-septate, 143-192μm long, 12-20 μm wide at its broadest part and 3-6 μm wide at the base.

Specimen examined: On decaying leaf litter of *C. congesta*; Verna, Goa; 20/4/1998; Miriam, J.; GUFCC No.625 (Live culture); 233(Herb. slide); Isolated by particle plating and moist chamber incubation.

Sporidesmium altum (Preuss) M.B.Ellis, 1958. Mycol.Pap. 70: 46-48 (Fig. 139)

Colonies effuse, olive brown to grey, fuscous, hairy. Mycelium partly immersed. Conidiophores mononematous, caespitose, unbranched, straight to flexuous, moderate to dark brown, 28-37 x 4-6 μ m. Conidiogenous cells monoblastic, integrated, terminal, determinate, cylindrical to doliform. Conidia solitary, dry, acrogenous, simple, obclavate to obpyriform, dark brown, hyaline at the tip, smooth, transversely 4-7-septate, 31-44 μ m long, 10-14 μ m wide at its broadest part and 1.5-2.5 μ m wide at the base.

Specimen examined: On decaying leaf litter of *C. congesta;* Verna, Goa; 20/4/1998; Miriam, J.; GUFCC No.234 (Herb. specimen/slide). Isolated by moist chamber incubation.

Sporidesmium eupatoriicola M.B. Ellis, 1958. Mycol. Pap. 70: 67 (Fig. 140)

Colonies effuse, olive brown to grey, fuscous, hairy. Mycelium partly immersed. Conidiophores mononematous, caespitose, unbranched, straight to flexuous, moderate to dark brown, 92-180μm long, 15-17μm wide at the base, tapering to 4-8μm at the tip. Conidiogenous cells monoblastic, integrated, terminal, determinate, cylindrical to doliform. Conidia solitary, dry, acrogenous, simple, obclavate to obpyriform, dark brown, smooth, transversely 10-26 septate, 98-236 μm long, 17-26 μm wide at its broadest part and 8-9 μm wide at the base.

Specimen examined: On decaying leaf litter of *C. carandas;* Verna, Goa; 23/5/1998; Miriam, J.; GUFCC No 235 (Herb. specimen/slide). Isolated by moist chamber incubation.

Colonies on MEA effuse, circular, regular, with smooth margin, creamish white, with black, slimy, shining spore masses, 1.3-1.5 cm diam. in 7 days, with faint concentric circles; reverse of the colony creamish white. *Mycelium* thin, with hyaline, 1.5-2 μm wide, smooth, septate, branched, thin-walled hyphae usually grouped together in bunches or fascicles from which the conidiophores arise. *Conidiophores* mononematous, simple, unbranched, darker grey at the tip and with decreasing colour intensity towards the base, smooth, nonseptate, 30-36 x 3-5 μm, bearing at its apex a crown of phialides. *Phialides* clavate, smooth, light grey, 6.5-8.3 x 3.5-4 μm. *Conidia* broadly ellipsoidal to subspherical, light grey to dark blackish brown, verrucose, 1-septate, 6.5-10 x 3-5.5 μm, aggregated in large, slimy, black, shining heads.

Specimen examined: On latex of fresh leaf of F. benghalensis, GU Campus, Taleigao, Goa, 26/12/98; Miriam, J.; GUFCC No.626(Live culture); 236 (Herb. slide). Isolated by particle plating.

Stachybotrys nephrospora Hansf., 1943. Proc. Linn. Soc. Lond. 1942-43: 44-45

(Fig. 142)

Colonies effuse, circular, white with black spore masses. Mycelium partly immersed, partly superficial. Conidiophores mononematous, simple, erect, unbranched, greyish at tip, lighter at base, smooth, septate, 30-36 x 3-5 μm, bearing at its apex a crown of phialides. Phialides clavate, smooth, hyaline, 6.5-8.3 x 3.5-4 μm. Conidia aggregated in large slimy black glistening heads, individually broadly ellipsoidal, reniform, black, verrucose, 6.5-10 x 3-5.5 μm.

Specimen examined: On leaf-litter of *C. congesta*; Verna, Goa; 22/7/1997; Miriam, J.; GUFCC No.237 (Herb. slide); Isolated by moist chamber incubation.

Stachylidium bicolor Link ex S.F. Gray (Ref: M.B.Ellis, 1971. Dematiaceous Hyphomycetes, 538). (Fig. 143)

Colonies on MEA,effuse, green brown, regular, velvety, 2.2cm diam. in 7 days, reverse of the colony brown. *Mycelium* moderate, white, wavy, with thinwalled, hyaline, septate,1.5-2.0 μm wide hyphae. *Conidiophores* mononematous, straight to flexuous, smooth, lower part sterile, light brown, upper part and branches hyaline with verticillately arranged phialides. 74-83 x 2.2-4.3μm. *Conidiogenous cells* monophialidic, discrete, arranged in verticils, determinate, cylindrical, rounded at the apex with a minute opening, smooth,4.3-14.3 x 2.2-2.6μm. *Conidia*, in slimy masses, simple, elliptical, elongate, smooth, hyaline,5-9.1 x 3-4μm

Specimen examined: On leaf-litter of *C. congesta*; Verna, Goa; 25/10/1997; Miriam, J.; GUFCC No. 238 (Herb. slide); Isolated by moist chamber incubation.

Sympodiella gracilispora Matsushima, 1975. Icones Microfungorum a Matsushima Lactorum, 151. (Fig. 144)

Colonies effuse, olivaceous brown, velvety. Mycelium partly superficial, with thin, septate, pale brown, smooth, 2-2.5 μm wide hyphae. Conidiophores mononematous, branched, straight, dark brown, smooth, up to 100 μm long, 10-15 μm wide at the base, 4-6 μm wide above. Conidiogenous cells polyblastic, sympodial, integrated, terminal, indeterminate, elongated. Conidia solitary, often in

straight, false-chains, dry, simple, cylindrical, with truncate ends, hyaline, smooth, 0-1-septate, $10-12 \times 2-3 \mu m$.

<u>Specimen examined</u>: On decaying leaf litter of *C. carandas;* Verna, Goa; 20/4/1998; Miriam, J.; GUFCC No.627 (Live culture); 239 (Herb. slide). Isolated by particle plating and moist chamber incubation.

Sympodiella multiseptata Tubaki, 1971. Trans. mycol. Soc. Japan, 12:24. (Fig. 145)

Colonies effuse, olivaceous brown, velvety. Mycelium partly superficial, with thin, septate, pale brown, smooth, 2-2.5 μm wide hyphae. Conidiophores mononematous, branched, straight, dark brown, smooth, septate, up to 220 μm long, 10-15 μm wide at the base, 4-8 μm wide above. Conidiogenous cells polyblastic, sympodial, integrated, terminal or intercalary, indeterminate. Conidia solitary, often in straight, branched or unbranched false-chains, simple, cylindrical, with truncate ends, hyaline, smooth, 2-3-septate, 14-30 x 2-3.5 μm.

Specimen examined: On decaying leaf litter of *F. benghalensis*; Verna, Goa; 23/8/1998; Miriam, J.; GUFCC No. 240 (Herb. specimen/slide). Isolated by moist chamber incubation.

Tetraposporium sp. (M.B. Ellis, 1971. Dematiaceous Hyphomycetes: 54) (Fig. 146)

Colonies on MEA, slightly irregular, grey, velutinous, diam in 7 days, reverse of the colony dark green. *Mycelium* moderate, hyal, with thin-walled, septate, smooth, 1-1.5 μm wide hyphae. *Conidiophores* smooth, branched, straight, very pale brown, 2.17 - 2.60 μm wide. *Conidiogenous cells* monoblastic, integrated, determinate, cylindrical, left behind as cylindrical denticles, 2.17 - 3.04 x 2.17 - 4.78 μm. *Conidia* solitary, simple, tetraradiate, light brown darker towards the centre and lighter on the arms.

Specimen examined: On leaf-litter of *F. benghalensis*; Verna, Goa; 13/9/1997; Miriam, J.; GUFCC No.628 (Live culture); 241(Herb. slide); Isolated by particle plating.

Trichobotrys effusa (Berk. & Br.) Petch, 1924. Ann. R. bot. Gdns. Peradeniya, 9: 169

(Fig. 147)

Colonies effuse, dark, olivaceous brown, velvety. Mycelium mostly superficial, with thick, septate, dark brown, verrucose, 2.5-3 μm wide hyphae. Conidiophores mononematous, branched, straight to flexuous, dark brown, verrucose, up to 1 mm long, 4-6 μm wide. Conidiogenous cells polyblastic, integrated, terminal or discrete, determinate, ellipsoidal to subspherical. Conidia solitary, often in false chains, dry, acrogenous, simple, spherical, dark brown to black, verrucose, 0-septate, 3-6 μm diam.

Specimen examined: On decaying leaves of *F. benghalensis*, GU Campus, Taleigao, Goa, 26/12/98; Miriam, J.; GUFCC No.629 (Live culture); 242 (Herb. slide); Isolated by particle plating and moist chamber incubation.

Trichoderma Persoon (Ref: Rifai, 1969. The genus Trichoderma. CMI Papers 11:1-56)

Type species: T. viride Pers.

Colonies effuse, white, irregular, visible as distinct colonies after 7 days. Mycelium thin, white to green, with thin, hyaline, smooth, branched, septate hyphae. Conidiophores mononematous, straight to flexuous, branched, septate. Conidiogenous cells phialidic, integrated, terminal, discrete, cylindrical. Conidia solitary, hyaline, spherical, smooth, ofter in slimy aggregates or false chains.

Trichoderma	Colony	Conidiophores	Conidia	Substrate	Fig. No
species	(Diam in cm)	(diam in µm)	(diam in µm)		
1-	0.6, irregular	12-25 x 1.5-2.5	3.4-4 x 2.5, slimy	on leaf	
		branched	in chains	litter of C.	148
				congesta	
2	1.2, regular	12-25 x 1.5-2.5	3.4-4 x 2.5, slimy	on leaf	
		much branched	in aggregates	litter of C.	149
				congesta	

Trichothecium roseum Link, 1809. Mag. Ges. Naturf. Freunde, Berlin 3: 18.

(Fig. 162)

Colonies effuse, white, cottony, tenuous. Mycelium partly immersed. Conidiophores mononematous, straight, unbranched, white, smooth, septate, up to 130 μ m long and 2.8-3.5 μ m wide. Conidiogenous cells polyblastic, integrated, terminal, cylindrical. Conidia solitary, simple, elongated, rounded and broad at the tip, truncate and narrow at base, slight curved at the tip, 1-septate, colourless, smooth, 15-20 x 3-8 μ m, aggregated in a slimy chains or masses.

Specimen examined: On leaf-litter of *C. congesta*; Verna, Goa; 22/9/1997; Miriam, J.; GUFCC No.245 (Herb. slide); Isolated by moist chamber incubation.

Trichocladium cylindroclavatum Matsushima, 1975. Icones Fungorum A

Matsushima Lectorum, p. 155. (Fig. 150)

Colony on MEA circular, punctate, granular, white, with regular margin, 0.25-0.5 cm diam. in 7 days,. Mycelium thin, smooth, with hyaline, septate, branched, thin-walled, 1.25 µm wide hyphae. Conidiophores mononematous, loosely branched, straight to flexuous, hyaline, 1.5-2 µm wide. Conidiogenous cells monoblastic, cylindrical, integrated, terminal or intercalary, determinate. Conidia solitary, dry, acrogenous, simple, clavate, cylindrical, rounded at the apex, smooth, thick-walled,

with 1-6 septa, moderately brown, sometimes differently coloured, basal cells usually hyaline to pale brown, 8.5-30 μm long, up to 5 μm wide at the broadest part.

Specimen examined: On decaying leaf litter of *F. benghalensis*; Verna, Goa; 29/10/1997; Miriam, J.; GUFCC No. 632 (Live culture); 246 (Herb. slide). Isolated by particle plating.

Trichocladium opacum (Corda) Hughes, 1952. Trans. Br. mycol. Soc., 35: 154-157 (Fig.151)

Colonies on MEA, light green, irregular, 1 cm diam in 7 days. Mycelium white, reverse light green, with smooth, thin-walled, septate, branched, hyaline, 2-2.2 μm wide hyphae. Conidiophores mononematous, straight to flexuous, branched, hyaline, smooth, up to 25 μm long, 1.7-3.3 μm wide. Conidiogenous cells monoblastic, integrated, intercalary, determinate, cylindrical. Conidia solitary, elongated, with rounded tip and truncate base, ellipsoidal, smooth, moderately brown, 1-6-septate, 10- 45 x 2.6-7.4 μm, sometimes part of conidiogenous cells remain attached at the base of the conidium.

Specimen examined: On decaying leaf litter and fresh leaf of *C. congesta* and soil; Verna, and GU Campus, Taleigao, Goa; 17/10/1998; Miriam, J.; GUFCC No. 633 (live culture); 247 (Herb. slide). Isolated by particle plating.

Tritriachium album Limber, 1940. Mycologia 32: 47 (Fig. 152)

Colonies on MEA effuse, faintly granular, white, cottony, circular, with irregular margin, 2 cm diam. in 7 days. *Mycelium* moderate, mostly superficial, white, with smooth, thin-walled, septate, branched, hyaline, 0.5-1.0µm wide hyphae. *Conidiophores* mononematous, smooth, unbranched, straight to flexuous, hyaline, 46-55µm long, 1-1.5 µm wide at the base, 1 µm wide above. *Conidiogenous cells*

monoblastic, sympodial, integrated, terminal or intercalary, determinate. *Conidia* solitary, dry, spherical, smooth, hyaline, 0-septate, 1-1.5 µm diam.

Specimen examined: On decaying leaf litter of *C. congesta*; Verna, and GU Campus, Taleigao, Goa; 27/7/1998; Miriam, J.; GUFCC No. 634 (live culture); 248 (Herb. slide). Isolated by particle plating and moist chamber incubation.

Tritirachium oryzae (Vincens) de Hoog 1972. Stud. Mycol. 1: 22 (Fig. 153)

Colonies on MEA effuse, very light pinkish, granular, powdery, dry, 1.8cm in 7 days, reverse creamish. Mycelium moderate, interspersed with light pinkish spore masses with thin-walled, hyaline, smooth, septate, 1.2-2 μm wide hyphae. Conidiophores mononematous, with 2 or more branches arising in verticils at regular intervals, smooth, hyaline, 300-370 x 3-4 μm. Conidiogenous cells polyblastic, sympodial, integrated, terminal. Conidia solitary, ovate, ellipsoidal with rounded apex and pointed base, smooth, hyaline, 3-4.3 x 2.2-3μm.

Specimen examined: On leaf litter of *F. benghalensis*; Verna, Goa; 11/4/98; Miriam, J.; GUFCC No. 635 (Live culture); 249 (Herb. slide); Isolated by particle plating.

The genus *Tritirachium* Limber, typified by *T. dependens* and characterised by sympodially developing conidiophores and minute conidia, has several species so far known (Carmichael et al., 1980; Hawksworth et al. 1995).

Torula herbarum (Pers.) Link ex S.F. Gray, 1821. Nat. Arr. Br. Pl., 1: 557 (Fig. 154)

Colonies effuse to puncate, black, velvety. Mycelium partly immersed hyphae septate, smooth, branched, hyaline, thin-walled, 1.5 µm wide. Conidiophores erect, flexuous, hyaline to pale brown, branched, smooth, 7-10 x 5-7 µm. Conidiogenous cells polyblastic, cupulate, verrucose, spherical, 5-7 µm diam. Conidia straight or

curved, cylindrical, rounded at both ends, verrucose, 5-7-septate, constricted at the septa, thick-walled, very dark brown, 20-30 x 5-10 µm.

Specimen examined: On decaying leaf litter of *F. benghalensis*; GU Campus, Taleigao, Goa; 20/3/1998; Miriam, J.; GUFCC No.636 (Live culture); 250 (Herb. slide). Isolated by particle plating and moist chamber incubation.

Vanakrippa parva Bhat et Kendrick, 1993. Mycotaxon 49: 77-79. (Fig. 155)

Colonies effuse, dark brown to black, velvety. Mycelium partly immersed hyphae, septate, smooth, branched, hyaline, thin-walled, 1.5 μm wide. Conidiophores in groups pulvinate, erect, flexuous, hyaline, septate, branched, smooth, arranged compactly forming a sporodochium, 30-78 x 3.2-6.4 μm. Conidiogenous cells holoblastic, geniculate, curved, hyaline. Conidia ellipsoidal, ovoid, rounded at the tip and flat at base, very dark brown, smooth, non-septate, thick-walled, 12.5-18.7 x 7.8-12.5 μm.

On decaying leaf litter of *F. benghalensis*; GU Campus, Taleigao, Goa; 19/10/1997; Miriam, J.; GUFCC No.251(Herb. slide). Isolated by moist chamber incubation.

Vermispora obclavata Vasant Rao et de Hoog, 1986. Stud. Mycol. 28: 53-54

(Fig. 156)

Colonies on MEA effuse, dark brown, granular, 1 cm diam. in 7 days, slightly irregular at the cord-like margin, with faint concentric zonations, reverse brown at the centre. *Mycelium* thin, white, with thin, smooth, septate, branched, hyphae 1.3-1.7 µm wide interspersed tiny clumps of spore masses. *Conidiophores* mononematous, straight to flexuous, branched, smooth, hyaline, up to 60 µm long,1-2 µm wide. *Conidiogenous cells* polyblastic, terminal, denticulate, globose to subglobose,

hyaline. *Conidia* elongated, club-shaped, broadly rounded at the tip, gradually tapering to a narrow truncate base, 3-4-septate, smooth, hyaline, 22.6-28.6 x 2.2-2.8 µm.

Specimen examined: On leaf litter of *F. benghalensis*; Verna, Goa; 9/4/98; Miriam, J.; GUFCC No. 637 (Live culture); 252 (Herb. slide); Isolated by particle plating.

Vernonea botryosa Cif. et Montemartini 1957. Atti. Ist.Bot.Univ.Lab.crittogam., Pavia, Ser.5,15: 68. (Fig. 157)

Colonies on MEA, effuse, brown regular, reverse of the colony brown. 2.0cm diam. in 7 days. Mycelium thin, brown, interspersed with dark brown spores, with thin walled, smooth, septate, hyaline, 1.7-2 μm wide hyphae. Conidiophores mononematous, straight to flexuous, branched, brown, smooth, septate, 74-130 x 1.3-1.7μm. Conidiogenous cells polyblastic, integrated, teminal, cylindrical, cicatrised. Conidia solitary, simple, ovate, ellipsoidal with rounded tip and pointed base, 0-septate, hyaline, smooth, 3.5-5.2 x 1-1.8 μm.

Specimen examined: On leaf-litter of *C. congesta*; Verna, Goa; 18/11/1997; Miriam, J.; GUFCC No.638 (Live culture); 253(Herb. slide); Isolated by particle plating.

Volutella roseola Cooke (Ref: Gilman, 1957. A Manual of Soil Fungi, 356) (Fig. 158)

Colonies on MEA white with granular yellow slimy masses of spores randomly distributed, regular, 1.5cm diam. in 7 days, reverse off-white. *Mycelium* thin, white, thin-walled, branched, septate, smooth, 1.5cm wide hyphae. Sporodochia solitary. Setae numerous, unbranched, erect, hyaline, 220-460 x 2.2-2.5 µm wide at the base. *Conidiophores* mononematous, straight to flexuous, branched, septate,

hyaline, smooth, 22-25 x 1.5 μ m. *Conidiogenous cells* terminal, discrete, phialidic, ampulliform, hyaline, smooth, 6.96-7.4 x 1.3 μ m. *Conidia* solitary, individually appearing hyaline when in a group yellow, cylindrical with truncate ends, smooth, 0-septate, 5.7-7.4 x 1 μ m.

Specimen examined: On leaf-litter of *C. congesta*; Verna, Goa; 26/8/1997; Miriam, J.; GUFCC No.639 (Live culture); 254(Herb. slide); Isolated by particle plating.

Wiesneriomyces javanicus Koorders, 1907. Bot. Untersuch., 246-247. (Fig. 159)

Colonies on MEA effuse, smooth, with slightly irregular margin, dark olive green to dark brown, with concentric circular zones, granular, 4.5 cm diam. in 7 days, reverse green. *Mycelium* white grey, loose across the colony, with thin-walled, smooth, dark brown, septate, up to 4 μm wide hyphae and dark brown chlamydospores. *Sporodochia* usually in groups of 5-6, scattered beneath the aerial mycelium, with dark brown stromatic base, pale brown to hyaline conidiophores and golden yellow slimy mass of conidia encircled by curved dark brown setae; young sporodochia with white or green and the mature ones with yellow conidial masses. *Setae* stiff, long, inwardly curved, subulate, swollen at the base, pointed at the tip, septate, brown, smooth, up to 550 μm long. *Conidiophores* closely arranged, branched at the apex, straight or flexuous, hyaline, smooth, 35-50 x 2-3 μm. *Conidiogenous cells* polyblastic, discrete, determinate, clavate to cylindrical, usually with 2-3 terminal denticles on which chains of conidia are borne, 8-12 x 3-4 μm. *Conidia* slimy, in chains, individually colourless, smooth, 0-septate; conidia at each end of the chain tapered and intermediate ones cylindrical, 10-12 x 3-4.5 μm.

On decaying leaf litter of *C. congesta*, *F. benghalensis a*nd on fresh leaf of *C. congesta* and on soil; Verna, and GU Campus, Taleigao, Goa; 11/7/1998; Miriam, J.; GUFCC No.640 (Live culture); 255 (Herb. slide). Isolated by particle plating and moist chamber incubation.

Zygosporium gibbum (Sacc.) Hughes 1958. Can. J. Bot. 36: 825 (Fig. 160)

Colonies on MEA effuse, compact, greyish brown, circular, 1.3 cm diam in 7 days, reverse white. *Mycelium* partly immersed, hyphae septate, smooth, branched, hyaline, thin-walled, 1.5 μm wide. *Conidiophores* mononematous, flexuous, pale brown, smooth, septate, branched, 30-70 x 3.2-6.5 μm, with a hyaline, sterile, hyphae terminating with a spherical knob. *Conidiogenous cells* holoblastic, discrete, 2-4, curved, ampuliiform, hyaline, perched on a black, curved, vesicle. *Conidia* spherical, smooth, thick-walled, hyaline, 6-9 μm diam.

Specimen examined: On decaying leaf litter of *F. benghalensis*; GU Campus, Taleigao, Goa; 14/5/1998; Miriam, J.; GUFCC No.641 (Live culture); 256 (Herb. slide). Isolated by particle plating and moist chamber incubation.

Zygosporium masonii (Sacc.) Hughes 1951. Mycol. Pap. 44:15-16 (Fig. 161)

Colonies on MEA effuse, compact, greyish brown, circular, 2.0 cm diam in 7 days, reverse white. *Mycelium* partly immersed, hyphae septate, smooth, branched, hyaline, thin-walled, 1.5 μm wide. *Conidiophores* mononematous, with chains of upto 6integrated vesicles and a hyaline, sterile apical region which terminates in a small knob, flexuous, brown, smooth, upto 100μm long and 1.5-2.0 μm wide. *Conidiogenous cells* holoblastic, discrete, 2-4, curved, ampuliiform, hyaline, perched on a black, curved, vesicle. Vesicles dark brown, smooth, 7-12 x 4-5μm. *Conidia* ellipsoidal, smooth, hyaline, 6-8 x 3-4 μm.

6integrated vesicles and a hyaline, sterile apical region which terminates in a small knob, flexuous, brown, smooth, upto 100μm long and 1.5-2.0 μm wide. *Conidiogenous cells* holoblastic, discrete, 2-4, curved, ampuliiform, hyaline, perched on a black, curved, vesicle. Vesicles dark brown, smooth, 7-12 x 4-5μm. *Conidia* ellipsoidal, smooth, hyaline, 6-8 x 3-4 μm.

Specimen examined: On decaying leaf litter of *F. benghalensis*; GU Campus, Taleigao, Goa; 17/7/1998; Miriam, J.; GUFCC No.642 (Live culture); 257 (Herb. slide). Isolated by particle plating and moist chamber incubation.

Undetermined Hyphomycete No.1

(Fig. 163)

Colonies on MEA effuse, granular, with irregular margin, yellow, 1.8 cm diam. in 7 days; reverse pale yellow. Mycelium partly immersed, moderate, with branched, septate, pale yellow, up to 2 μm wide hyphae. Conidiophores synnematous, distinct, erect, straight to flexuous, septate, branched, with two types of conidiophores: (i) the basal three fourth of the synnema interwoven by sterile, branched, septate, verrucose, golden brown, 4.8-7.2 μm wide hyphae; (ii) the other branched, septate, smooth, hyaline, 25-110 x 1.7-2.2 μm hyphae bearing conidiogenous apparatus at the apex. Synnemata up to 4.7 mm long, up to 2 mm wide. Chlamydospores present. Conidiogenous cells monobalstic, integrated, terminal. Conidia solitary, oblong to obovate rounded at the apex, truncate and narrow at the base, aseptate, hyaline, smooth, 15-24 μm long, 1.5 μm wide at the base, 4.7-8.5 μm wide in the middle and apex.

Specimen examined: On leaf litter of *F.benghalensis*; Taleigao, Goa; 15/11/1997; Miriam, J.; GUFCC No.643 (Live culture); 258 (Herb. slide).; Isolated by particle plating and moist chamber incubation method.

Undetermined Hyphomycete No. 2

(Fig. 164)

Colonies on MEA appearing like stubs, with regular margin, pale yellow brown, 1.5 cm diam. in 7 days, reverse light brown, darker at the centre. *Mycelium* thin, with smooth, thick-walled, septate, branched, brown, 1.7-2.2 μm wide hyphae. *Conidiophores* synnematous, conspicuous, erect, septate, branched, compactly arranged in the basal half and splayed apart in the apical part, up to 240 μm long, 14 μm wide at the base and 130 μm wide in the splayed apical region. *Conidiogenous cells* phialidic, integrated, determinate, thick-walled, smooth, brown, septate, up to 240 μm long and 1.5-2.5 μm wide. *Conidia* solitary, elliptical, rounded at the apex, pointed at the base, aseptate, thick-walled, brown, smooth, 5.2-7.4 x 2.2-4 μm.

Specimen Examined: On soil.; Taleigao, Goa; 13/9/1997; Miriam, J.; GUFCC No.644 (Live culture); 259 (Herb. slide); Isolated by particle plating.

Undetermined Hyphomycete No. 3

(Fig. 165)

Colonies on MEA effuse, dry, granular, with regular margin, white, with faint concentric circles, 1.2 cm diam. in 7 days. *Mycelium* thin, white, with septate, branched, hyaline, 3-3.5 μm wide hyphae. *Conidiophores* smooth, profusely branched, straight to flexuous, hyaline, up to 87 μm long, 2.2-4 μm wide, broader at regions where branches arise. *Conidiogenous cells* monophialidic, integrated, terminal, determinate, ampullate, smooth, hyaline, 8.2-18 μm long, 1.7-2.6 μm wide at the broadest part, 1-1.5 μm wide at the neck. *Conidia* solitary, obovate, rounded at the apex, narrow at the point of attachment, hyaline, 4.7-7.4 x 3-4 μm.

Specimen examined: On leaf litter of *F. benghalensis* and soil; Taleigao, Goa; 23/7/97; Miriam, J.; GUFCC No.645 (Live culture); 260 (Herb. slide); Isolated by particle plating.

Undetermined Hyphomycete No. 4

(Fig. 166)

Colonies on MEA effuse, flat, 1 cm diam. in 7 days, circular, with wavy margin, white, reverse cream coloured. *Mycelium* moderate, with smooth, thin-walled, septate, branched, 1.7 μm wide hyphae. *Conidiophores* mononematous, branched, straight to flexuous, smooth, hyaline, 17-40 μm long, 1.7-2.2 μm wide. *Conidiogenous cells* polyblastic, integrated, terminal or intercalary, denticulate, with cylindrical denticles. *Conidia* solitary, dry, elliptical, rounded at the apex, tapering at the point of attachment, 0-septate, smooth, hyaline, 4.5-7.4 x 2.3-3 μm.

Specimen examined: On leaf litter of *C. congesta*; Taleigao, Goa; 22/10/98; Miriam, J.; GUFCC No.646 (Live culture); 261(Herb. slide); Isolated by particle plating.

Undetermined hyphomycete No. 5

(Fig. 167)

Colonies on MEA effuse, punctate, with irregular, prominently rooted margin, dark brown, 0.25cm diam. in 7 days, reverse dark brown. *Mycelium* thin, extended like a mat over the entire colony, with smooth, thin-walled, septate, branched, hyaline to pale brown, 1.5-2.0μm wide hyphae. *Conidiophores* mononematous, smooth, unbranched, 2-7.5 μm wide, hyaline to pale brown. *Conidiogenous cells* monoblastic, integrated, determinate. *Conidia* solitary, dry, simple, vermiform, elongated, with rounded ends, 0-5-septate, smooth, hyaline, 26-65 μm long, 2-5 μm wide at its broadest part.

Specimen examined: On leaf litter of *C. congesta*; Taleigao, Goa; 18/3/1997; Miriam, J.; GUFCC No.647 (Live culture); 262 (Herb. slide); Isolated by particle plating.

Undetermined hyphomycete No.6

(Fig. 168)

Colonies on MEA effuse, velvety, faintly concentric, light brown, with regular margin, 4 cm diam. in 7 days; reverse centrally brown decreasing in colour intensity towards the periphery and produces a reddish-maroon coloured pigment. *Mycelium* thin, with smooth, septate, branched, pale brown, 1.5-2.6 μm wide hyphae often forming thick-walled chlamydospores. *Conidiophores* mononematous, smooth, unbranched, straight to flexuous, reddish brown to golden brown, 18-40 μm long, 1.5-3.5μm wide. *Conidiogenous cells* monoblastic, integrated, terminal. *Conidia* solitary, simple, almost fusiform, with rounded apex, narrow and truncate at the base, 2-3-septate, smooth, reddish to golden brown, 30-47 μm long, 1-1.5 μm wide at the base, 3.5-5.5 μm wide at its broadest part.

Specimen examined: On leaf litter of *C. congesta*; Taleigao, Goa; 21/101997; Miriam, J.; GUFCC No.648 (Live culture) 263 (Herb. slide); Isolated by particle plating.

Undetermined Hyphomycete No. 7

(Fig. 169)

Colonies on MEA circular, with faint concentric zones, with randoml radiating darker lines, with regular smooth margin, olive green, velvety, 2.6 cm diam. in 7 days; reverse dark green with irregular radial bands. *Mycelium* moderate, with smooth, septate, branched, hyaline, 1.5-2 μm wide hyphae. *Conidiophores* mononematous, smooth, indistinct, covered with a mucilaginous mass, thin-walled, septate, with very thick, dark septa, branched, light brown, 1.3-2.2 μm wide. *Conidiogenous cells* polyphialidic, integrated, terminal to intercalary, determinate to percurrent, smooth, covered with slime, unbranched, flexuous, light brown, 6.5-30 μm long, 1.7-2.6 μm wide at the base, 1-1.5 μm at the tip, with a up to 3.5 μm wide collarette. *Conidia*

simple, ellipsoidal, pointed at the base, smooth, hyaline, $2.6-7 \times 1-2 \mu m$, aggregating in a slimy mass at the tip of the phialide.

Specimen examined: On leaf litter of *C. congesta*; Taleigao, Goa; 13/6/1998; Miriam, J.; GUFCC No.649 (Live culture); 264 (Herb. slide); Isolated by particle plating.

Undetermined hyphomycete No. 8

(Fig. 170)

Colony on MEA effuse, white-grey with pale orange pinkish slimy granular spore masses randomly distributed, irregular, 0.9cm diam. in 7 days, reverse of the colony centrally dark grey with thion cream/off-white margin. *Mycelium* moderate grey with thin-walled, hyaline, septate, branched, smooth, 1.5-2 um-wide hyphae, sometimes darker brown strands of hyphae are also observed. Setae unbranched, erect, thickwalled, brown, upto 60.87um long and 2.24-3.04μm. *Sporodochia* with numerous setae and profusely branched conidiophores. *Conidiophores* monnematous, straight to flexuous, branched, smooth, hyaline, arising at the base of seta, 2.6-2.8μm wide. *Conidiogenous cells* phialidic, terminal, discrete, cylindrical almost ampulliform, hyaline, 2-90 x 2-3 μm. *Conidia* solitary, in slimy masses, 0-septate, cylindrical with rounded ends, hyaline, smooth, 10-18 x 4-5 μm.

<u>Specimen examined</u>: On fresh leaf and leaf-litter of *C. congesta*; Verna, Goa; 25/9/1997; Miriam, J.; GUFCC No.650 (Live culture); 265 (Herb. slide); Isolated by particle plating and moist chamber incubation.

Undetermined Hyphomycete No. 9

(Fig. 171)

Colonies on MEA effuse, olivaceous to dark brown, stubs, 2.3 cm diam. in 7 days, reverse of the colony brown. Mycelium thin, brown with thin-walled, smooth, light brown, septate, 1.8-2 cm um-wide hyphae. Conidiophores erect, septate,

light brown, septate, 1.8-2 cm um-wide hyphae. *Conidiophores* erect, septate, branched, upto 300μm long, splayed apart in the upper 3/4th region. *Conidiogenous cells* polyblastic, discrete, terminal, intercalary, denticulate, denticles cylindrical, light brown, 2.6-4.3 x 1-2.4μm. Also interspersed are spherical dark brown, smooth, sterile structures 8.7-11.3 μm diam. *Conidia* solitary, 1-septate, ovate with round apex and pointed at the base, hyaline, smooth, 7.4-10 x 2.4-3.5 μm.

Specimen examined: On leaf-litter of *C. congesta*; Verna, Goa; 28/11/1997; Miriam, J.; GUFCC No.651 (Live culture); 266 (Herb. slide); Isolated by particle plating.

Undetermined Hyphomycete No. 10

(Fig. 173)

Colonies on MEA velutinous, granular, with thin concentric circles placed close to one another, circular with wavy margin, grey to light green, spore masses in tiny clumps dispersed through out colony, 0.5 cm diam. in 7 days, reverse dark olive green in the centre and decreasing towards the light green margin. *Mycelium* moderate, interspersed with spore masses forming a compact mat, with smooth, thin-walled, septate, branched, light brown, 1.3-2 μm wide hyphae. *Conidiophores* mononematous, smooth, branched, straight to flexuous, brown, 10-100 μm long, 1.5-3 μm wide at the base. *Conidiogenous cells* polyblastic, integrated, terminal or intercalary, denticulate, with cylindrical denticles. *Conidia* solitary, obclavate with a narrow stalk-lke base, pale brown to hyaline, 1-septate, 8-40 μm long, up to 1.5 μm wide at the narrow stalk, 2-3.5 μm wide at the broadest part, up to 1.5 μm wide at the tip.

Specimen examined: On leaf litter of *C. congesta*; Taleigao, Goa; 23/8/1998; Miriam, J.; GUFCC No.652 (Live culture); 267 (Herb. slide); Isolated by particle plating.

Colonies on MEA effuse, partly flat, partly perched, with a single concentric circle just before the periphery, with regular margin, dark brown, 1 cm diam. in 7 days; reverse dark brown. *Mycelium* moderate, with smooth, thin-walled, septate, branched, brown to golden brown, 1.5-2.2 μm wide hyphae. *Conidiophores* mononematous, 2.6-14 x 1-4 μm. *Conidiogenous cells* monoblastic, integrated, terminal, determinate, with a cylindrical denticle after the cessation of conidia. *Conidia* solitary, elongated, rounded at the apex, truncate at the base, 2-multiseptate, generally 3-septate, smooth, brown to golden brown, 10-16 x 2.6-4.5 μm.

Specimen examined: On leaf-litter of *C. congesta*; Verna, Goa; 18/9/1997; Miriam, J.; GUFCC No.659(Live culture); 276(Herb. slide); Isolated by particle plating.

Undetermined Hyphomycete No. 12

(Fig. 175)

Colonies on MEA effuse, faintly concentric, with regular, smooth margin, olive green in the central 3/4th of colony, rest creamish, 1.7-2 cm diam. in 7 days. Mycelium moderate, forming a soft loose mat, smooth, with two types of hyphae; (i) thin-walled, smooth, septate, branched, light brown, 1.3-2.6 μm wide and (ii) thick, septate, rough, brown, with foldings on either side of the wall giving a verrucose appearance, 3 μm wide. Conidiophores mononematous, irregularly branched, straight to flexuous, smooth, hyaline, sometimes swollen or spherical 10-12 x 4-6 μm. Conidiogenous cells monoblastic, integrated, terminal or intercalary, determinate, cylindrical. Conidia solitary, oblong, rounded at ends, broadly ellipsoidal to subspherical, spirally twisted or recurved, thick-walled, smooth, septate, pale olivaceous to dark brown, 65-75 x 10-56 μm.

Specimen examined: On leaf-litter of *F. benghalensis*; Verna, Goa; 13/6/1998; Miriam, J.; GUFCC No.653 (Live culture); 268 (Herb. slide); Isolated by particle plating.

Undetermined Hyphomycete No.13

(Fig.176)

Colonies on MEA felty, dry, powdery with spore masses, with irregular, root like margin that radiates from the center, ash brown, with a single concentric circle in the periphery, 1 cm diam. in 7days. *Mycelium* thin, white, with smooth, thin-walled, septate, branched, light brown, 2-4.5 μm wide hypae. *Conidiophores* mononematous, profusely branched, straight to flexuous, smooth, light to dusty brown, 10-87 μm long, 1-2.6 μm wide. *Conidiogenous cells* monophialidic, integrated, terminal, determinate, flask-shaped, broader at the base, tapering at the neck, smooth, light brown, 6-17 μm long, 1.7-3 μm at the broadest basal part, 1-1.5 μm wide at the neck. *Conidia* catenate, elliptical, elongate, almost barrel shaped, light to dusty brown, 1.7-3.5 x 1.7-2.4 μm.

Specimen examined: On leaf litter of F. benghalensis; Taleigao, Goa; 13/10/1997; Miriam, J.; GUFCC No.654 (Live culture); 269 (Herb. slide). Isolated by particle plating.

Undetermined Hyphomycete No.14

(Fig. 177)

Colonies effuse, sparse, with smooth margin, dusty brown, 1 cm diam. in 7 days, reverse grey brown. *Mycelium* in thin loose tufts, with smooth, thin-walled, septate, branched, light brown, 1-2 µm wide hyphae. *Conidiophores* mononematous, indistinct. *Conidiogenous cells* monophialidic, integrated, unbranched, terminal, vase-like or elongated, with a long neck, sometimes the base is narrow and elongated before it broadens in the middle, thin-walled, smooth, medium brown, 4-10 µm long,

1-1.5 μ m wide at the base, 2-3 μ m wide in the middle, 1.5 μ m wide at the neck region, with a collarette. *Conidia* catenate, spherical, smooth, brown, moderately thick-walled, the outer wall appears contiguous, 3-5 μ m diam.

Specimen examined: On soil; Verna, Goa; 19/9/1997; Miriam, J.; GUFCC No. 655 (Live culture); 270 (Herb. slide); Isolated by particle plating.

Undetermined Hyphomycete No. 15

(Fig. 178)

Colony on MEA circular, shining, sparsely granular, with smooth margin, medium brown with clusters of spore masses scattered all over, 1 cm diam. in 7 days, reverse dark brown. Mycelium moderate, forming a soft mass, with smooth, thinwalled, septate, branched, hyaline, 1-1.7 μm wide hyphae. Conidiophores mononematous, straight, smooth, septate, branched, very pale brown. Conidiogenous cells monoblastic, cylindrical, integrated, determinate, with 1.3-2 μm wide denticles. Conidia solitary, spherical, thick-walled, smooth, very dark brown to black, 2.6-4 μm diam.

Specimen examined: On leaf-litter of *C. congesta*; Verna, Goa; 22/10/1997; Miriam, J.; GUFCC No.656 (Live culture); 271(Herb. slide); Isolated by particle plating.

Undetermined Hyphomycete No. 16

(Fig. 180)

Colonies on MEA white, effuse, 2.2 cm diam in 7 days, reverse cream coloured. Mycelium moderate, white, with thin-walled, septate, branched, 1-2 µm wide hyphae. Conidiophores mononematous, branched, septate, smooth, hyaline, 30-80 x 2-4 µm terminating in a smooth, hyaline, 4-8 µm globose head bearing the conidiogenous cells directly or on several branches; branches 23-55 x 1-2 µm. Conidiogenous cells phialidic, discrete, determinate, terminal, hyaline, smooth, vase-

like, 4-55 x 1.5-4.5 μm . Conidia catenate, spherical, simple, hyaline, smooth, 1.8-2 μm diam..

Specimen examined: On leaf-litter of *C. congesta*; Verna, Goa; 18/9/1997; Miriam, J.; GUFCC No.657 (Live culture); 272(Herb. slide); Isolated by particle plating.

Figure No.

29

Table: 4.1.3 Legend to figures

Cirrenalia indica

Name of fungus

1 Acremoniula sarcinellae 2-6 Acremonium sp. 7 Acrodictys goanensis Acronidiellina indica 8 9 Alternaria alternata Arxiella terrestris 10 Aspergillus sp. 11-20 21 Bacillispora aquatica Beltrania rhombica 22 Beltraniella buloloensis 23 24 Botryosporium diffusum 25 Brachysporiella gayana 26 Cercospora celosiae 27 Cercospora carisseae 30 Chlamydomyces palmarum Circinotrichum maculiforme 28

Cladosporium	31-39
Corynespora smithii	40
Cunninghamella echinulata	41
Curvularia brachyspora	42
Curvularia clavata	43
Curvularia crepinii	44
Curvularia lunata	45
Curvularia ovoidea	46
Curvularia pallescens	47
Curvularia richardiae	48
Curvularia senegalensis	49
Curvularia trifolii	50
Curvularia tritici	51
Cylindrocarpon ianthothele	52
Cylindrocladium ilicicola	53
Cylindrocladium parvum	54
Dichtyochaeta assamica	55
Dicyma carrisseae	56
Doratomyces indicus	181
Dreschlera tripogonis	57
Epicoccum purpurascens	58
Exosporium bryophylli	59
Fulvia fulva	60
Fusarium culmorum	61

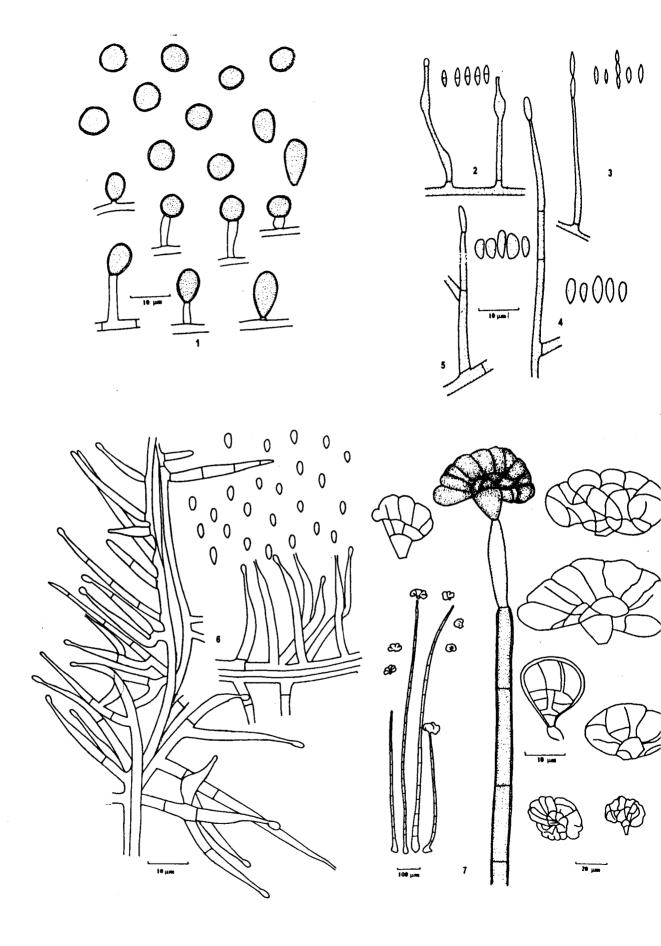
Fusarium decemcellulare	62
Fusarium phragmitis	63
Fusarium semitectum	64
Fusarium solani	65
Fusarium tabacinum	66
Gliomastix murorum	67
Gliomastix uniseptata	68
Gonatobotryum bimorphospora	69
Helicoma dennisii	70
Helicomyces roseus	71
Helicosporium guianensis	72
Helicosporium lumbricoides	73
Helminthosporium dalbergiae	74
Helminthosporium mauritianum	75
Helminthosporium microsorum	76
Heteroconium solaninum	77
Humicola brevis	78
Humicola grisea	79
Humicola verrucosa	80
Hyaloscolecobasidium indicum	81
Hyphodiscosia jaipurensis	82
Idriella fertilis	83
Idriella lunata	84
Idriella mucoidea	85

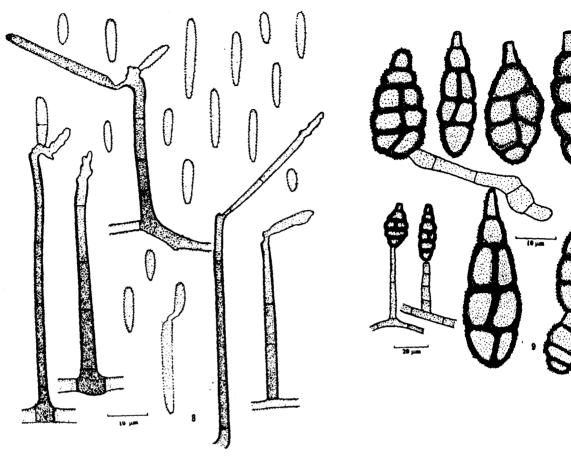
Idriella multiseptata	86
Idriella ramosa	87
Kumbhamaya indica	88
Memnoniella echinata	89
Monodictys nigra	90
Monodictys lepraria	91
Monodictys fluctuata	92
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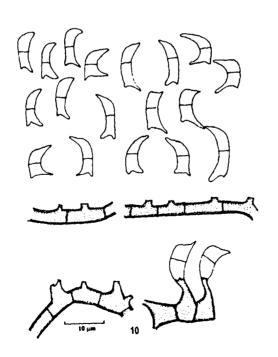
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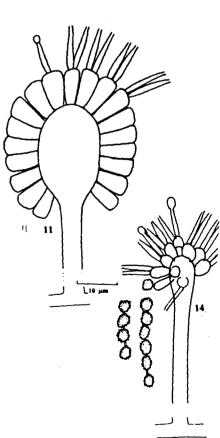
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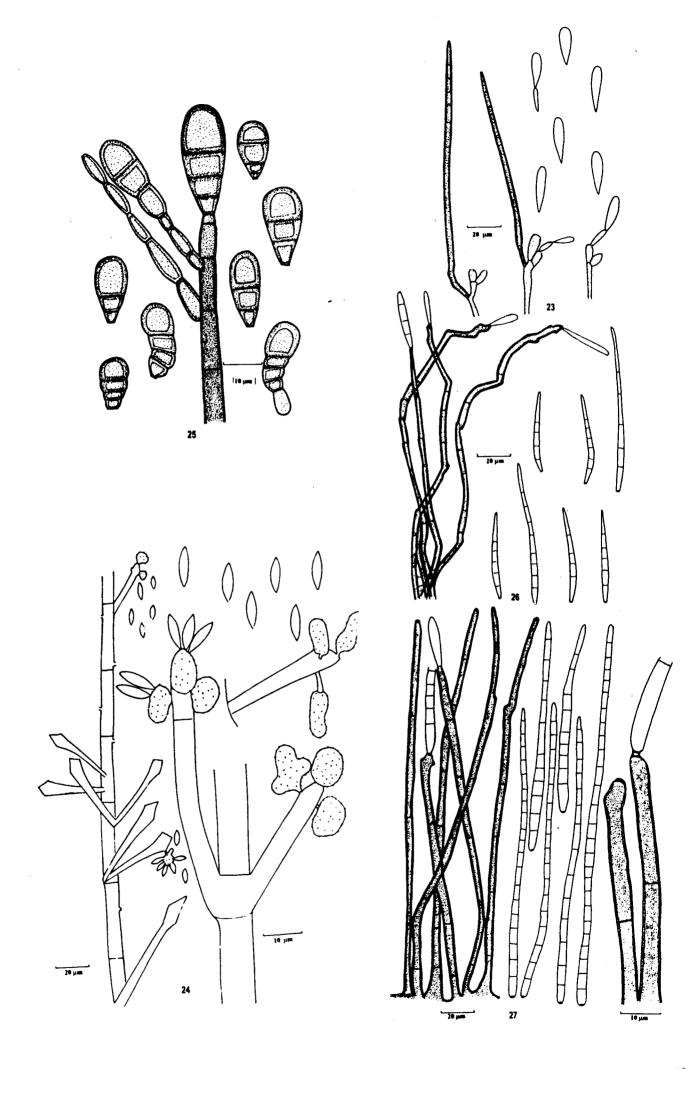




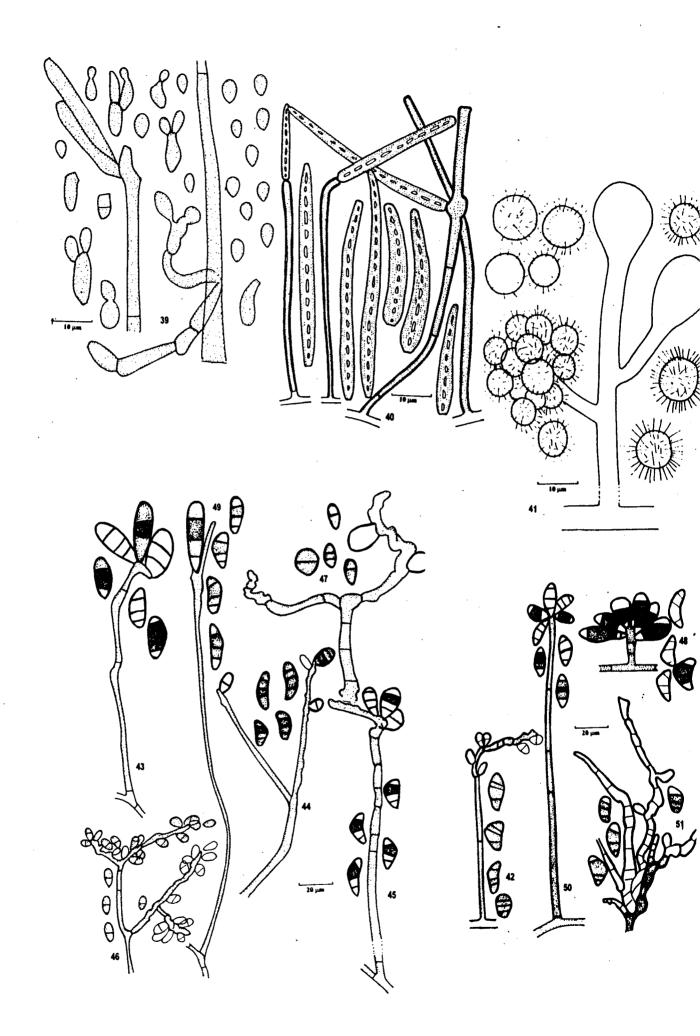


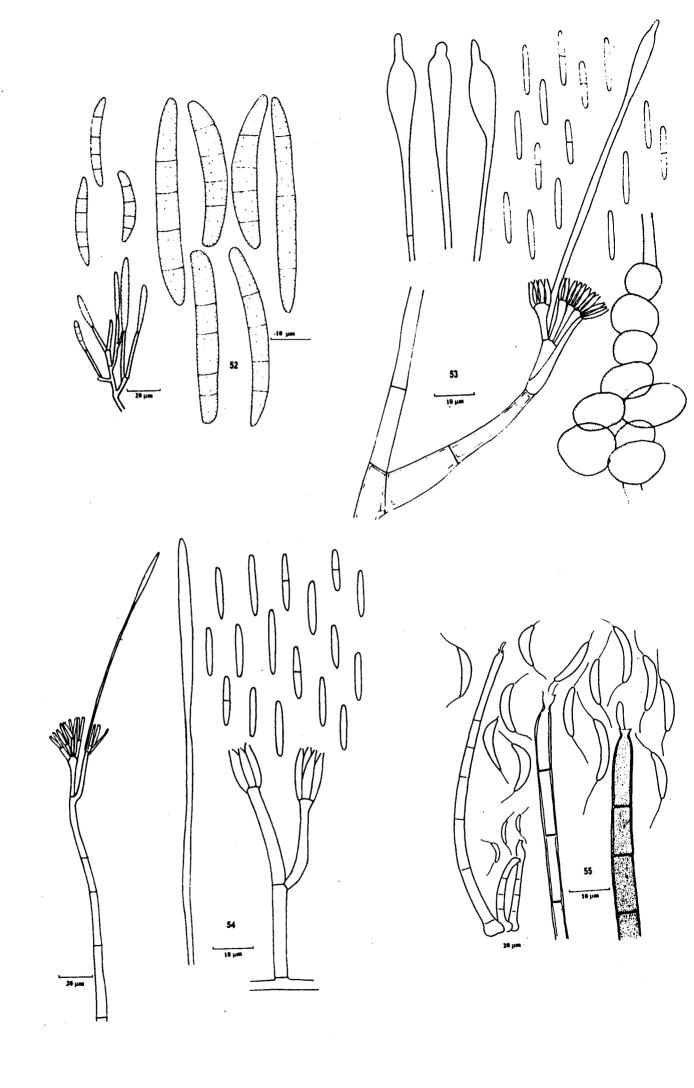


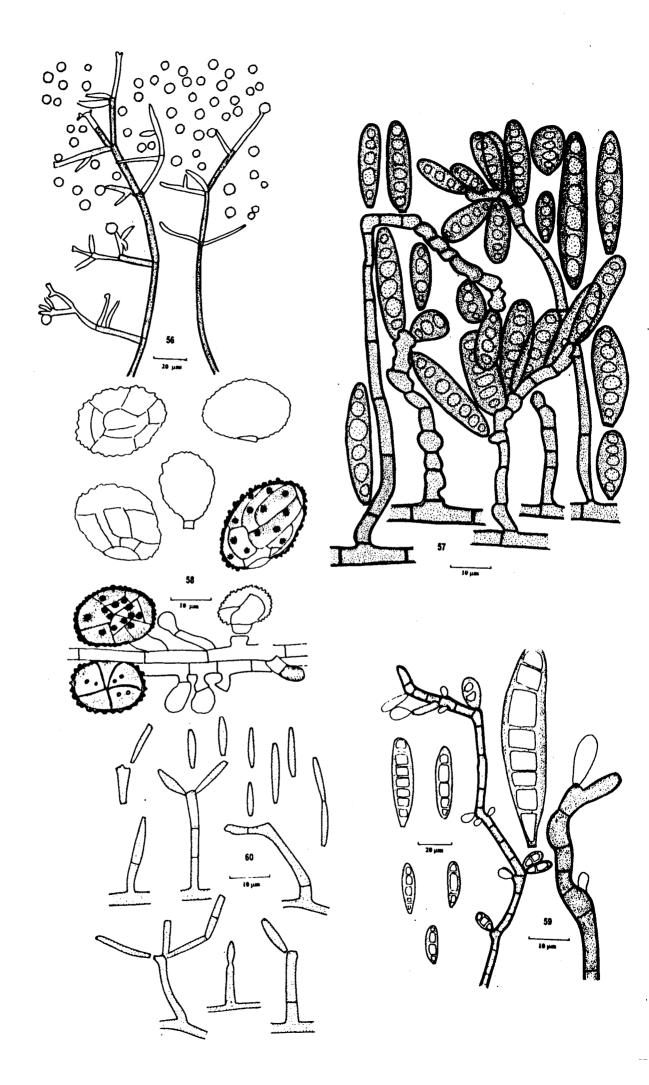


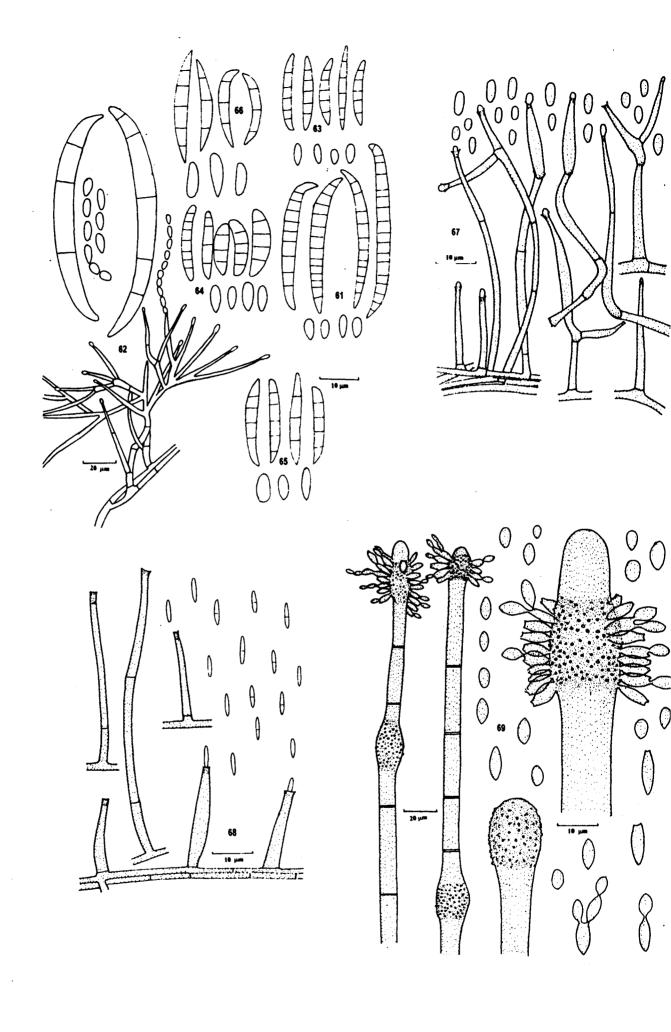


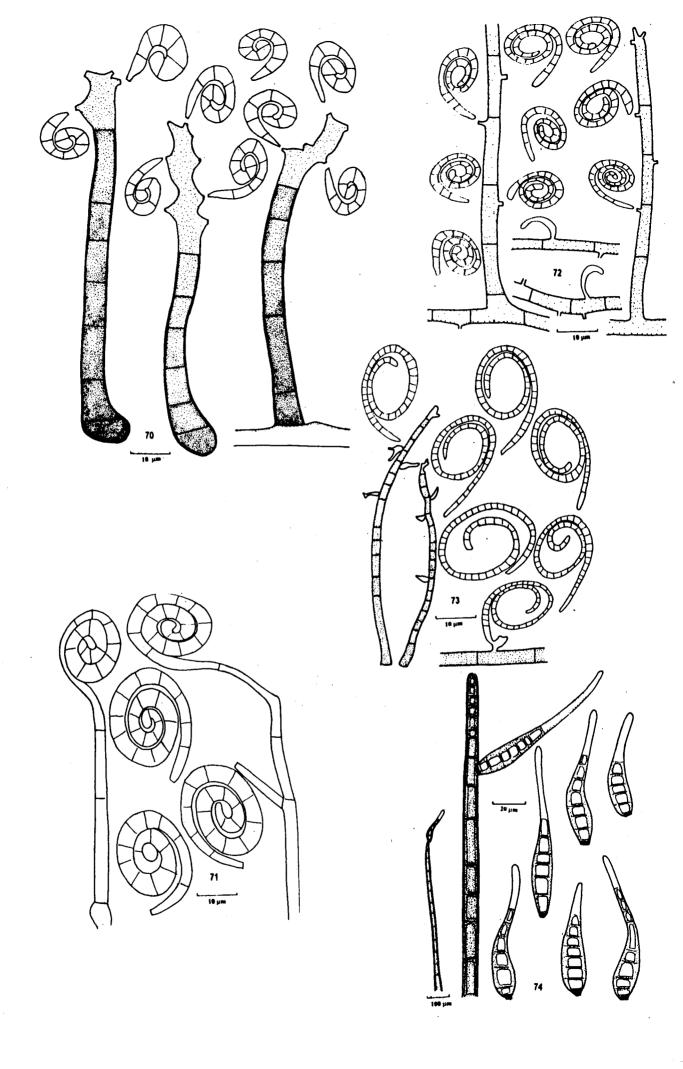


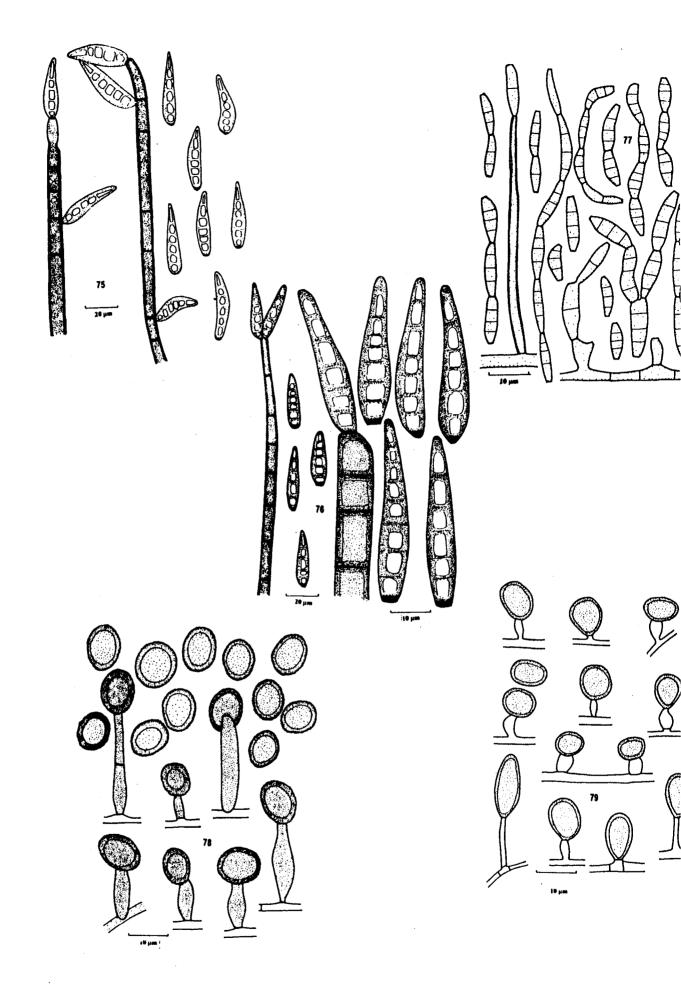


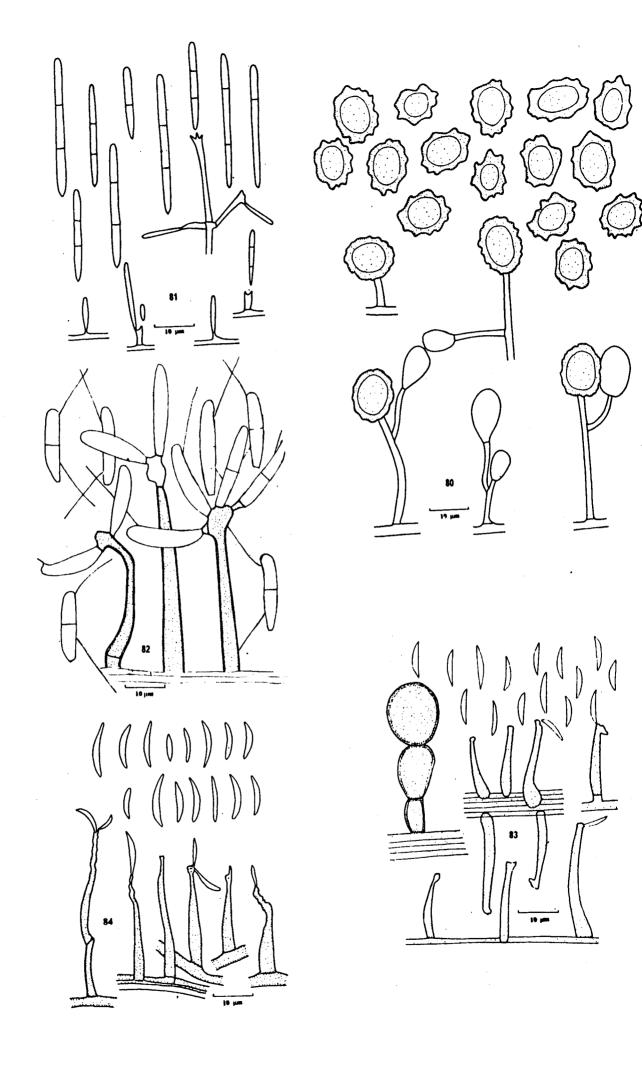


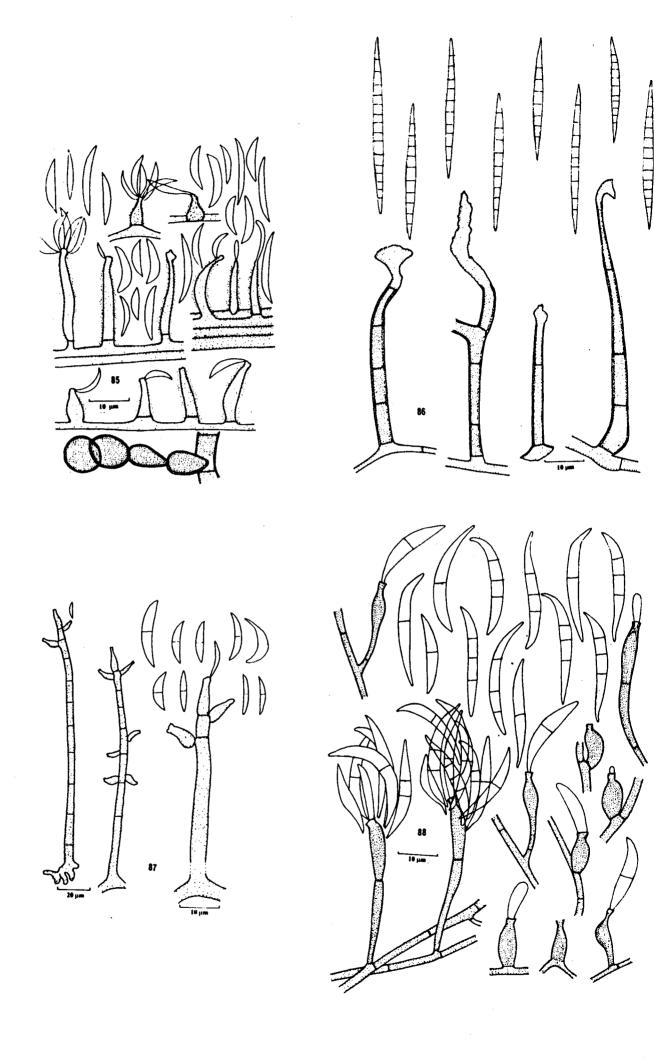


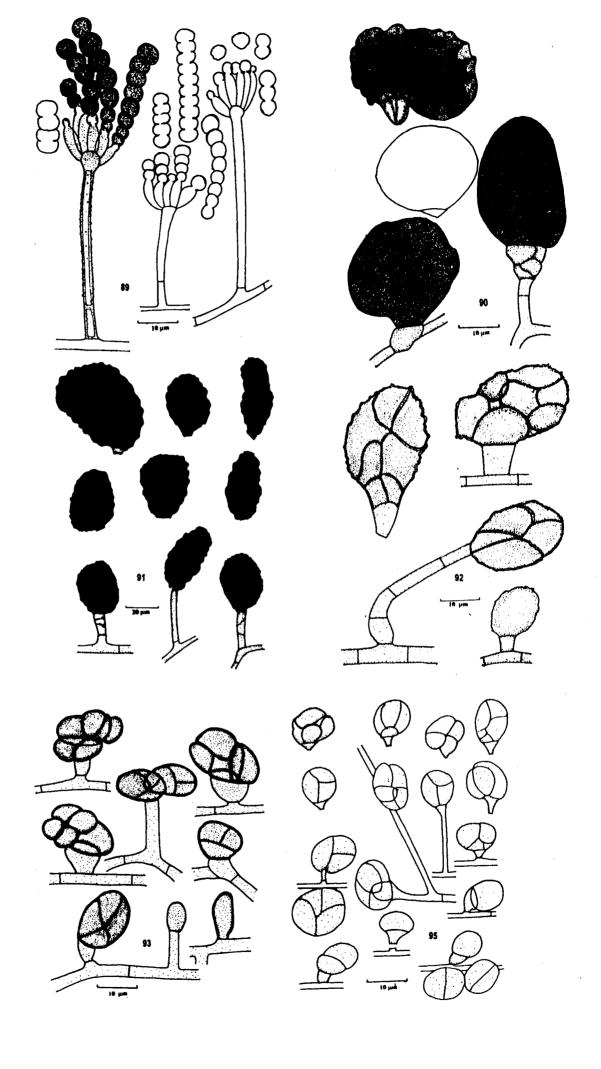


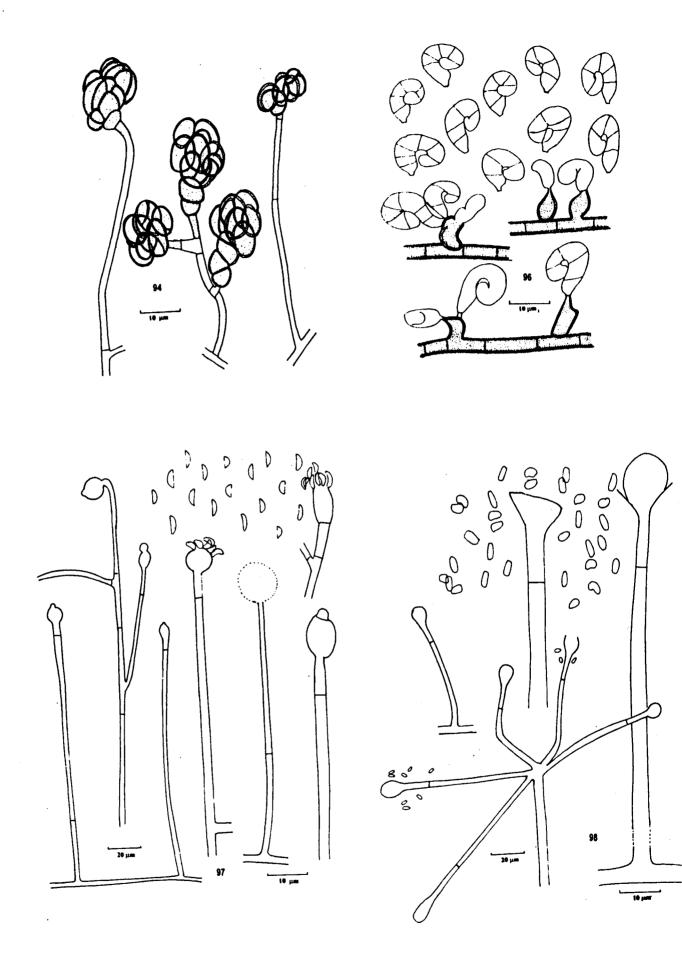


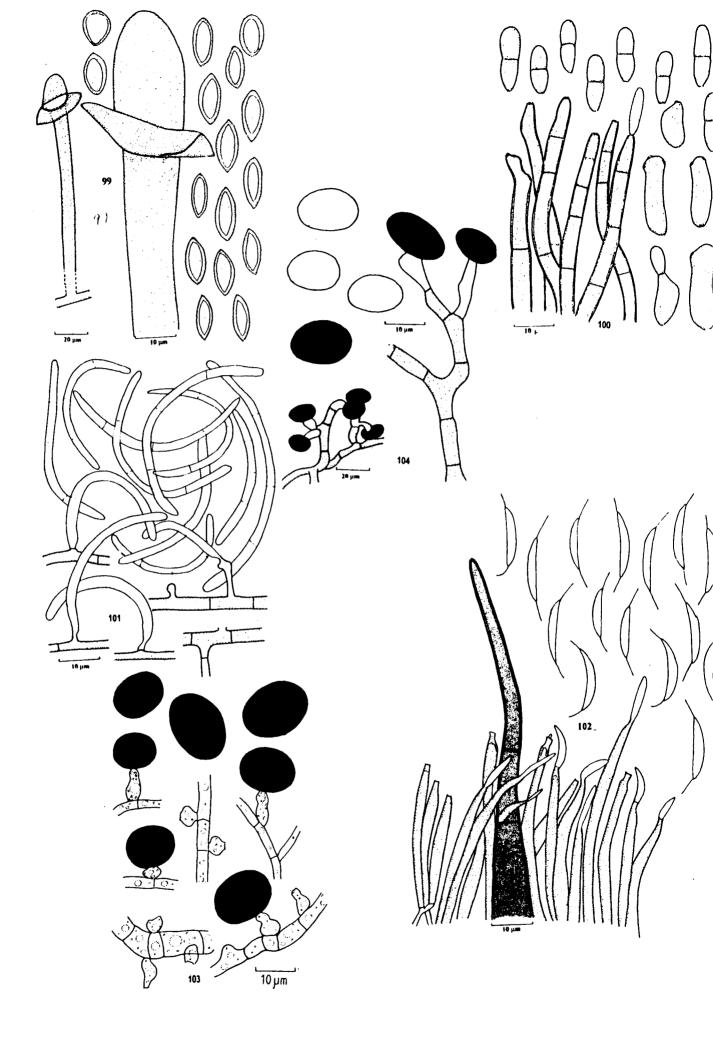


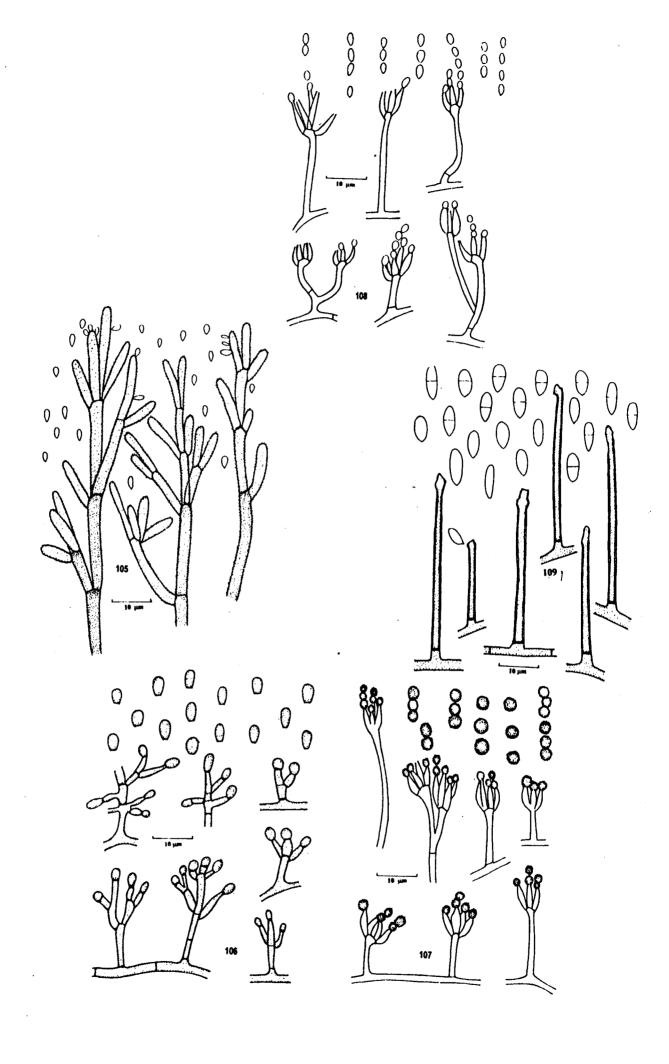


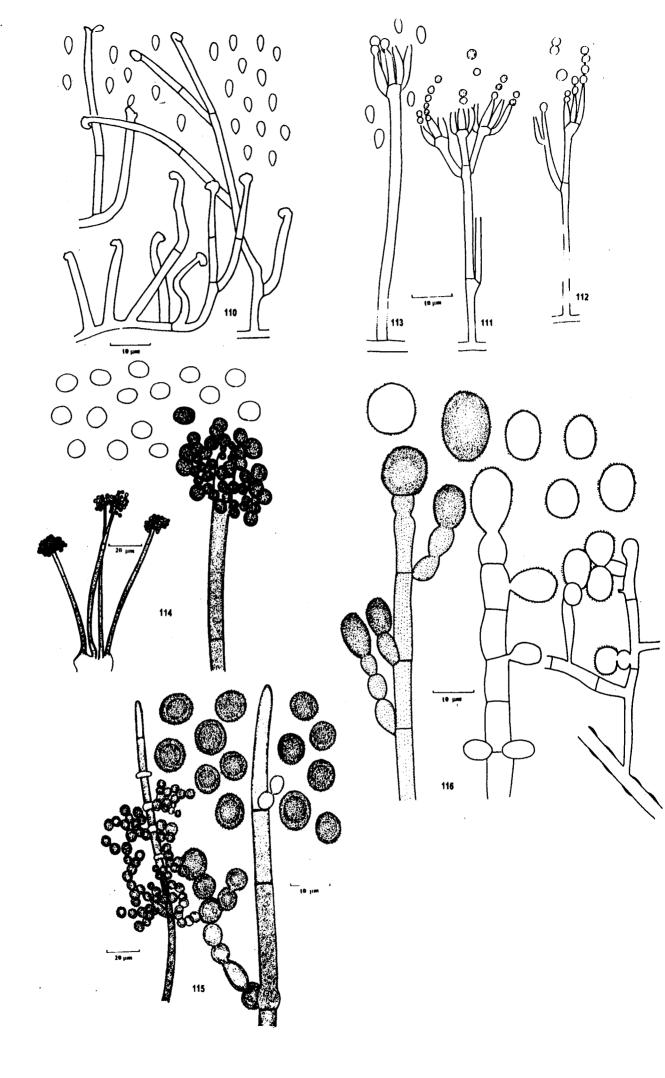


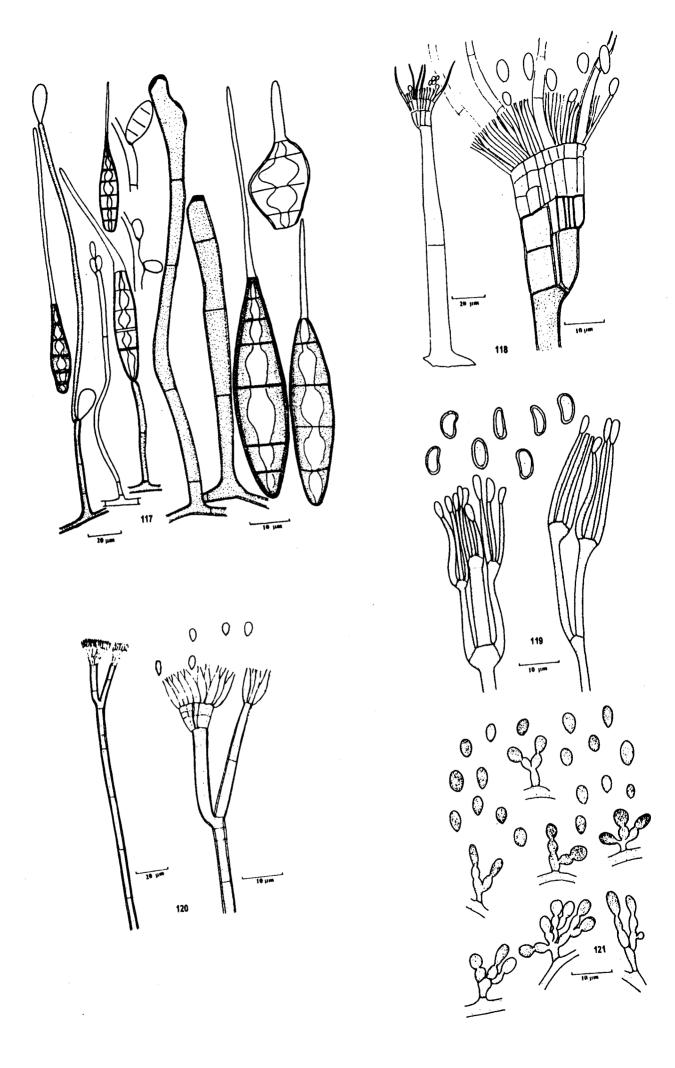


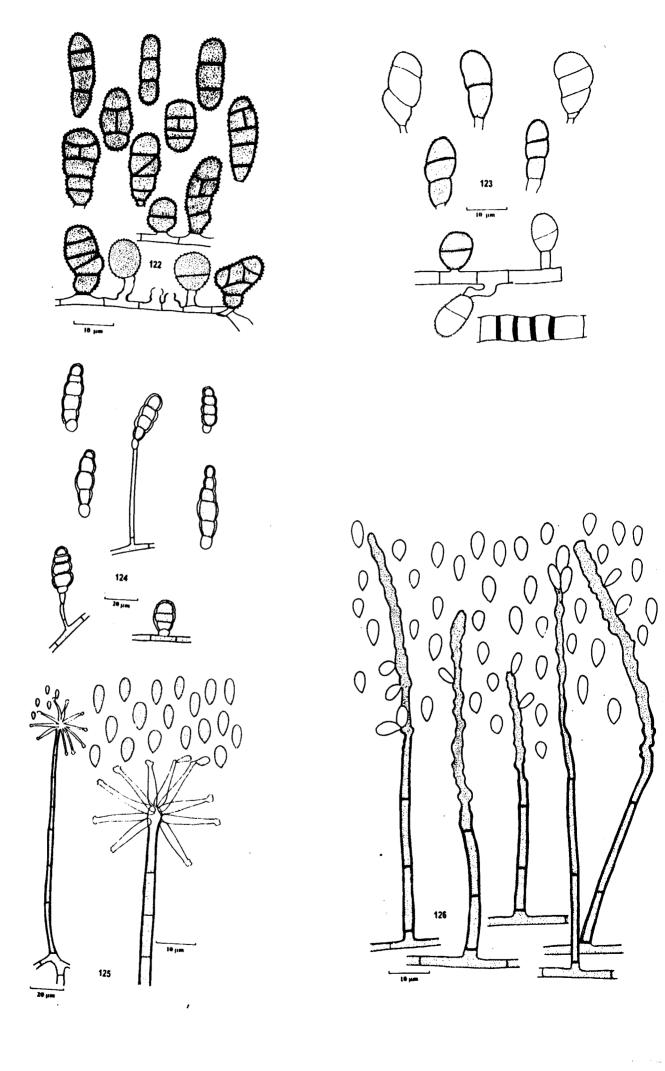


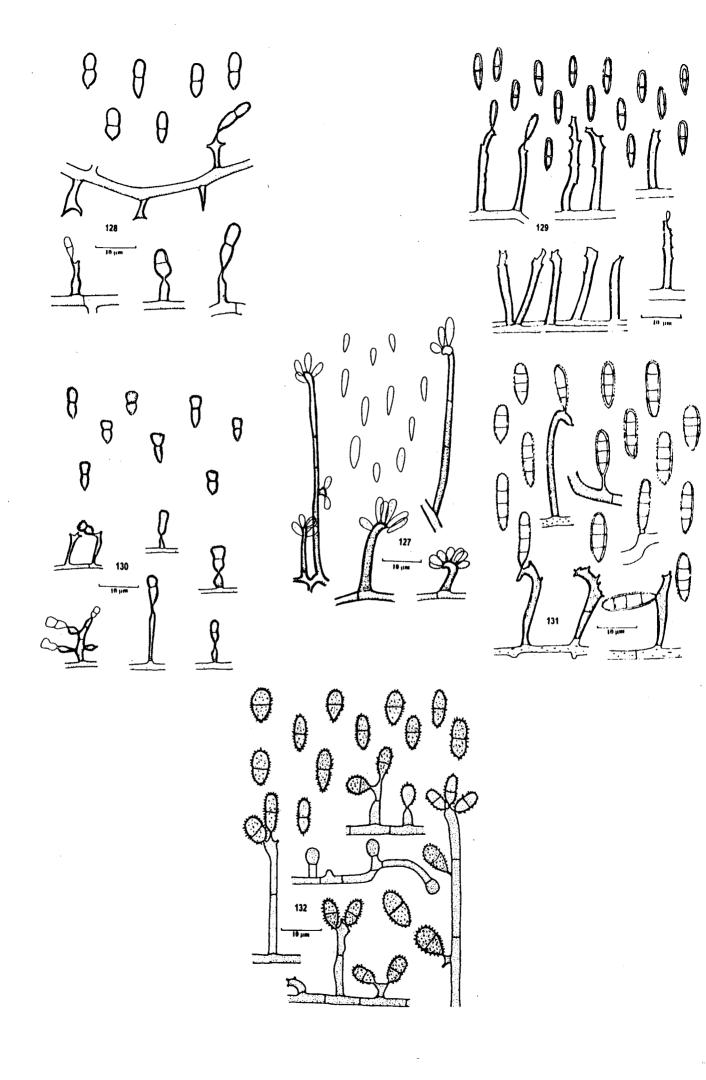


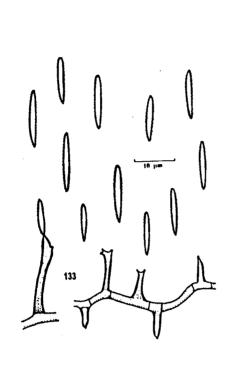


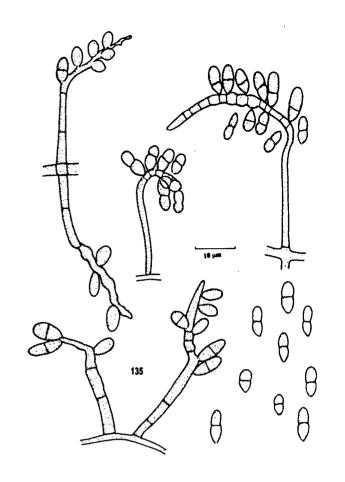


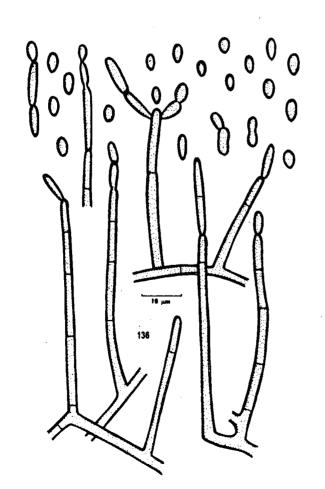


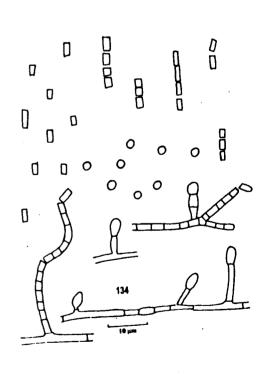


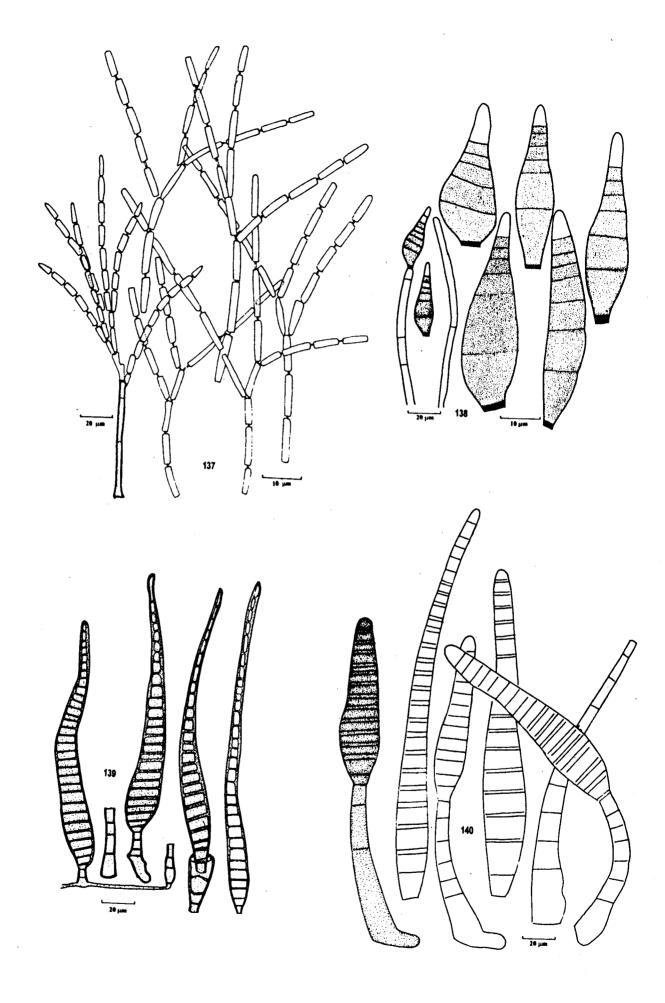


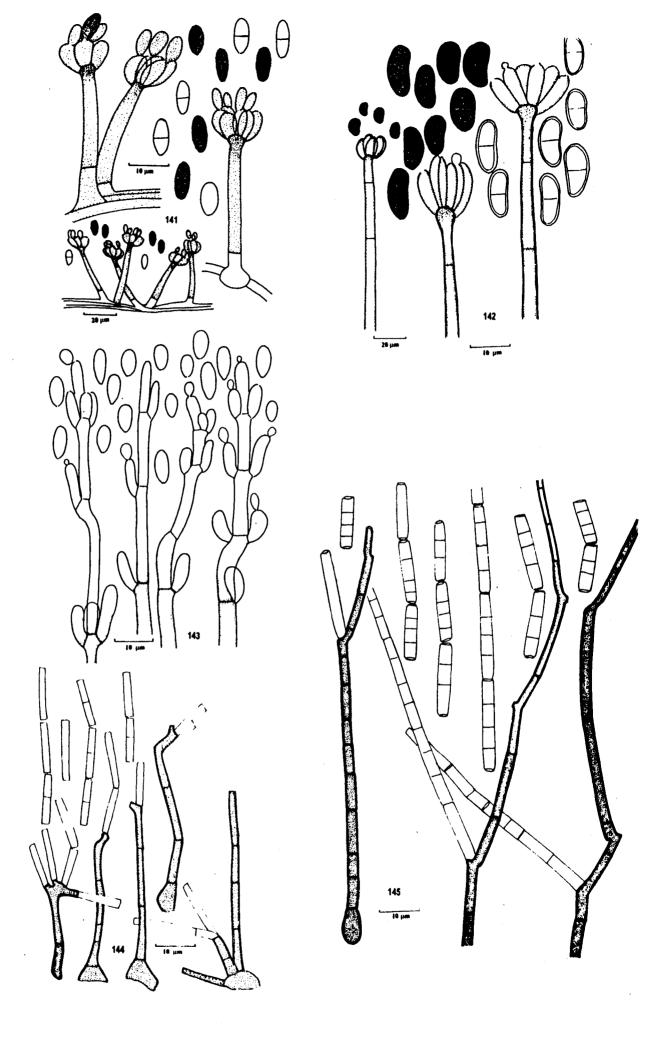


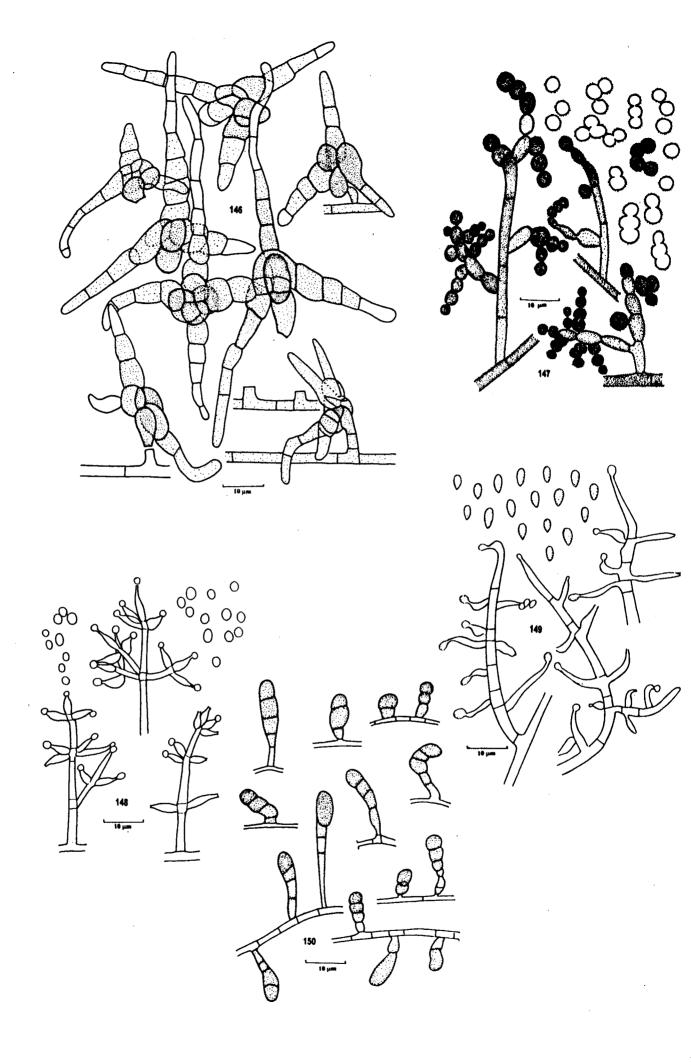


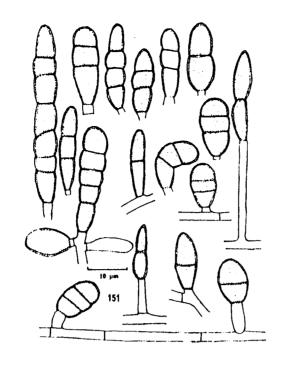


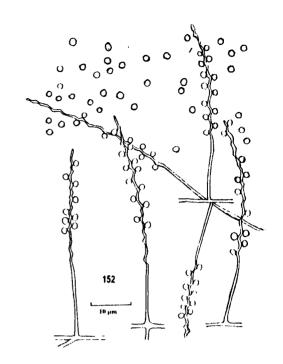


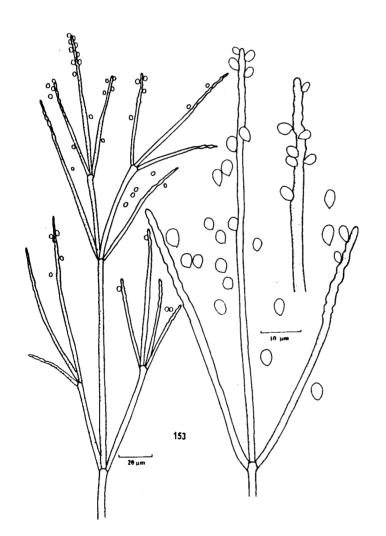


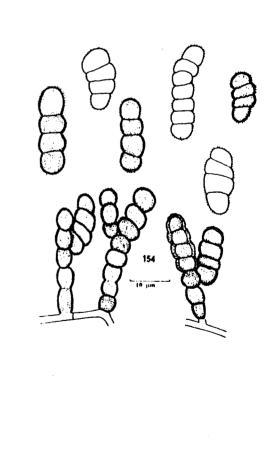


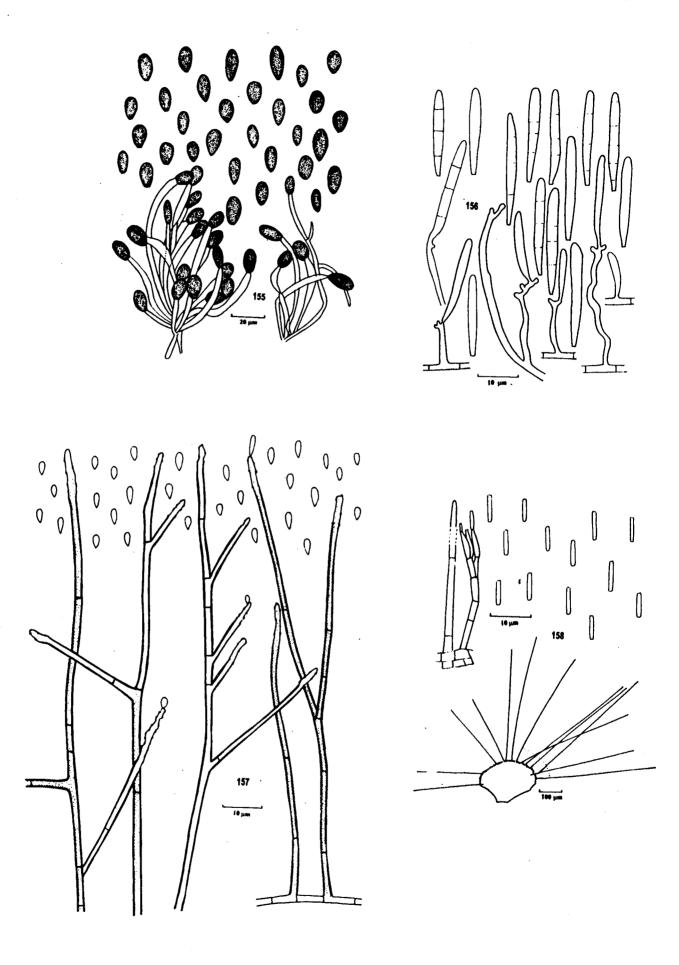


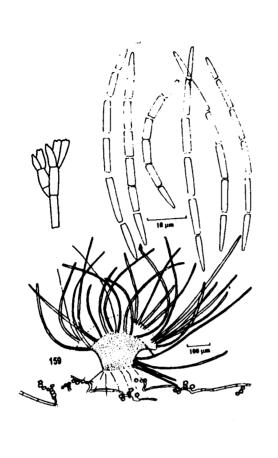


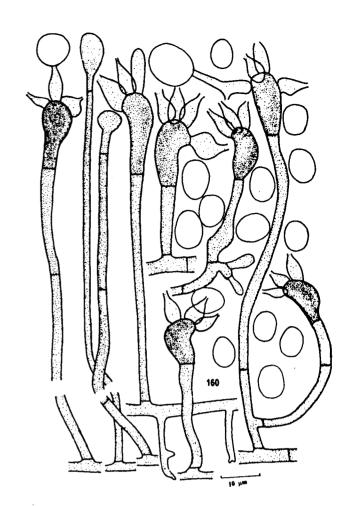


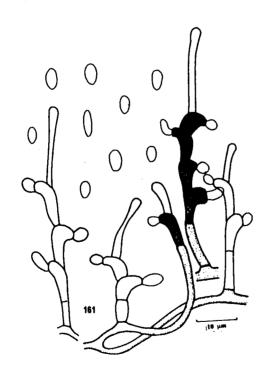


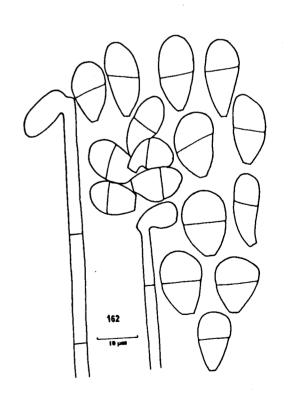




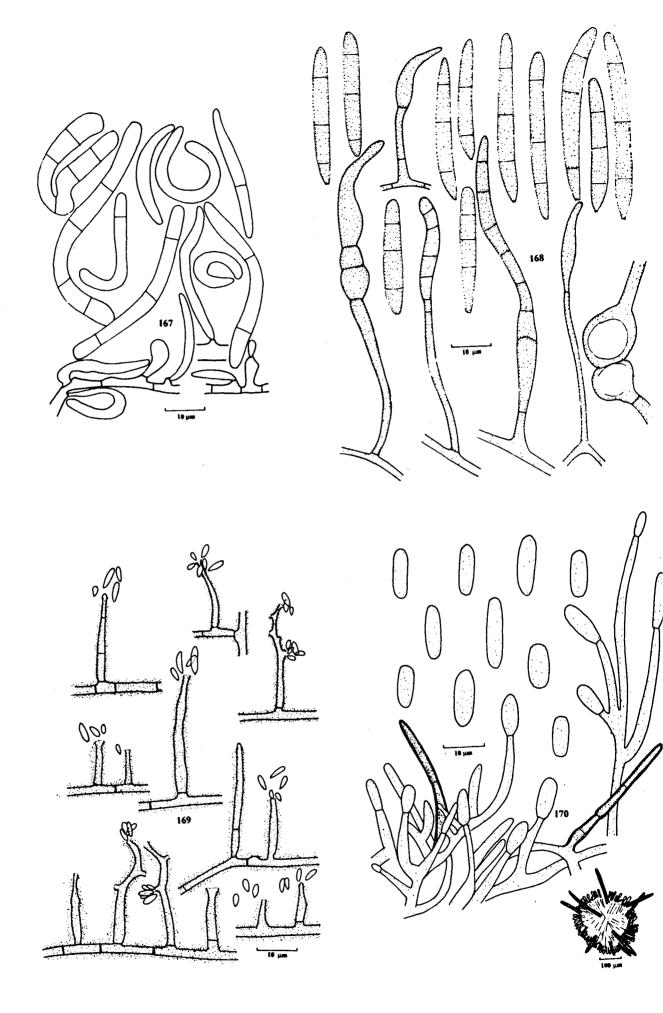


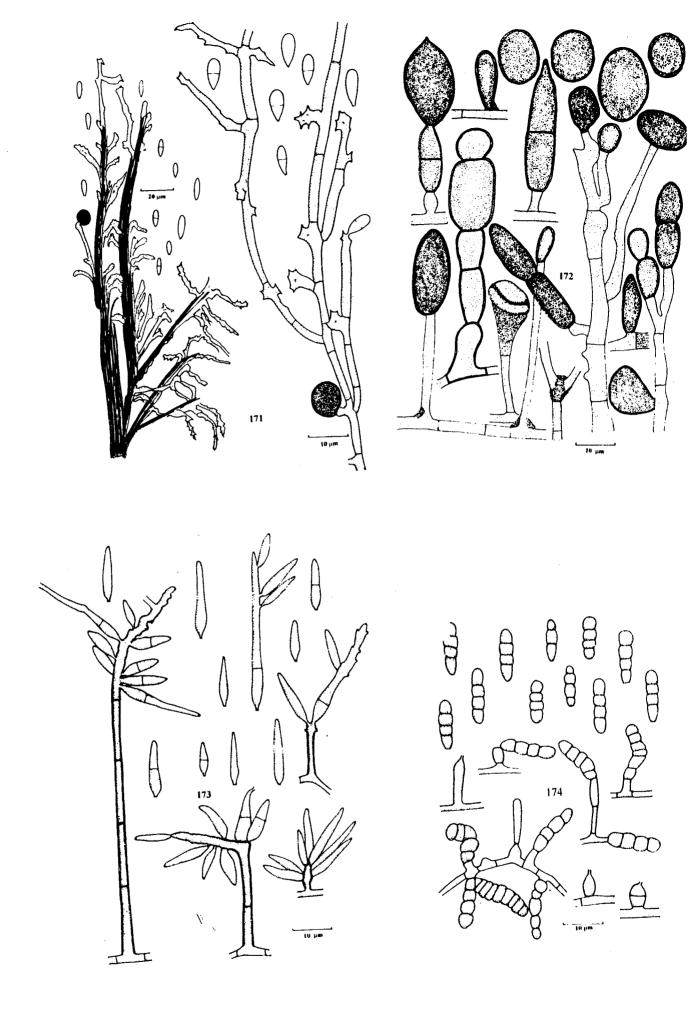


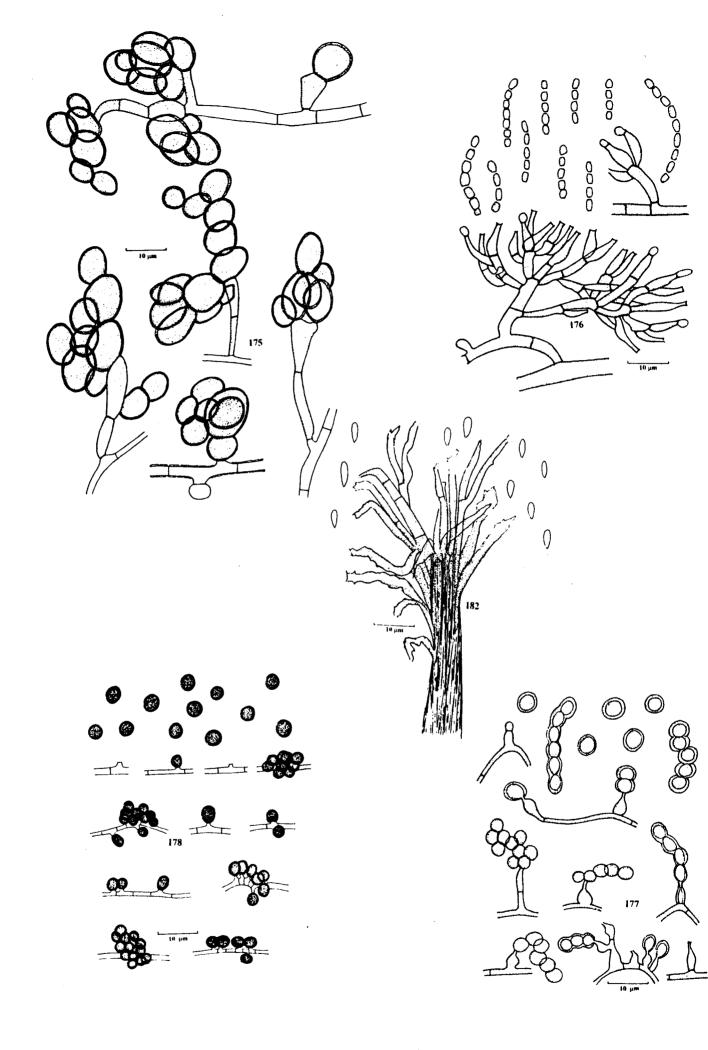


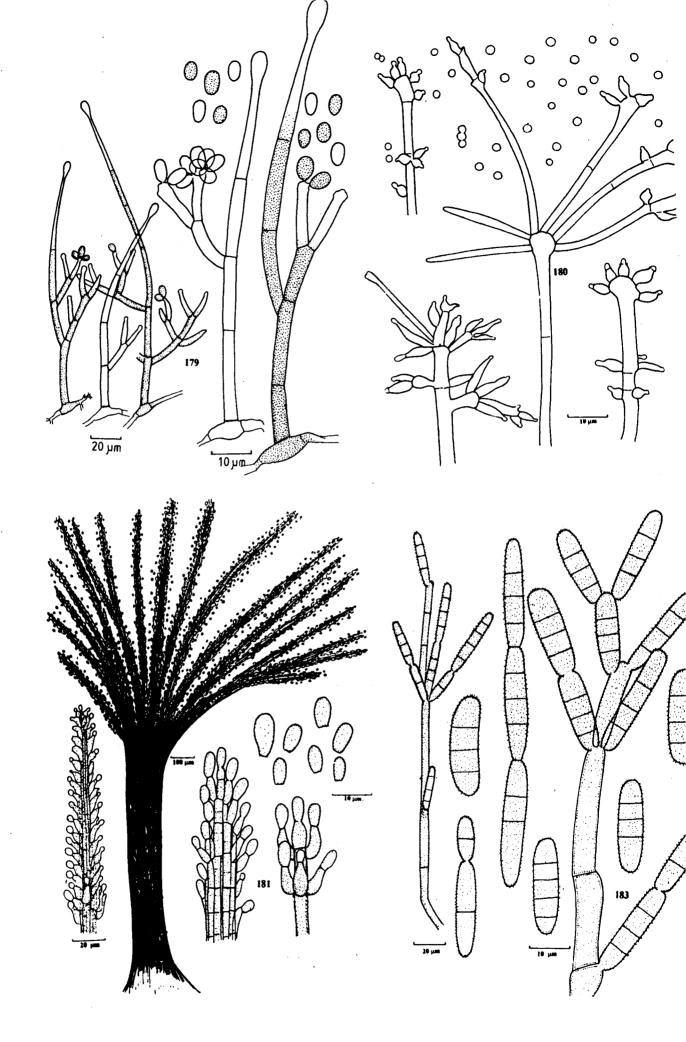












PART 2. ASSOCIATION OF THE FUNGI WITH THE TWO PLANTS AND THEIR NEIGHBOURHOOD

The ecological association of fungi with plants, that is - saprophytic, parasitic and mutualistic, has been a topic of considerable significance and recognised since long. As saprophytes, the fungi along with other decomposing organisms such as bacteria, viruses and nematodes are attributed to guaranty for complete decomposition of the plant parts. As parasites, the fungi subsist on the plants and plant parts. Both partners are known to derive advantage in mutualistic associations such as mycorrhizae or lichens (Kendrick, 1992).

The ecology of saprophytic fungi associated with plants and plant parts has been the subject of several investigations and this is reviewed in an earlier chapter. The hitherto carried out studies have revealed that there are fungi exhibiting substrate/host/ familial specificity, relation to localities and biogeographic regions, response to season and other environmental factors and specificity restricted even to plant parts. It was also possible to deduce indicators of litter decomposition at different time period. All groups of fungi, namely, the Zygomycetes, Ascomycetes, Basidiomycetes and Deuteromycetes have representives in the litter mycoflora. It is well known that fungi from air and soil participate in the decomposition of leaf litter (Dix and Webster, 1995).

The recent revelation is that the endophytic fungi, which live as symptomless inhabitors in the living tissues of plant parts, also contribute to the success of plant growth. The endophytes mainly include Ascomycetes and Deuteromycetes (Hyphomycetes and Coelomycetes). Most of these fungi within the host tissue remain as nonsporulating mycelial types throughout their life cycle (Petrini, 1986).

In the present study, investigation on the fungi occurring in association with two plant species, namely *Ficus benghalensis* and *Carissa congesta* and their neighbourhood, that is, the underneath soil and ambient air at a height of 1 M above

the ground level, was carried out at monthly intervals over a period of 24 months, i.e. Jan. 1997 to Dec. 1999, from two places, namely Taleigao and Verna in Goa.

The two plants subjected for investigation were different in several respects. The selection of distinct plants was aimed primarily to achieve information on commonness and contrast in the qualitative and quantitative occurrence of fungi in relation to substrate/host/family. The choice of two distantly located places namely Taleigao and Verna, was aimed at comparison of the results from the point of spacial difference.

Two methods of recovery techniques, namely moist-chamber and particle plating were used to isolate the fungi from the leaf litter. In the moist chamber technique, the resident fungal propagules present on the surface and subsurface grew and sporulated well, in the presence of high humidity and ambient temperature. After a few days of incubation, these fungi were available for isolation, though individual fungal structures were not discernible. The fungal fruiting structures were transferred to microslides and subjected to taxonomic indentification.

In the particle plating method, dilute solution (0.05 ml) of litter particles of 100-250 µm size when spread evenly and wide apart on MEA plates, allowed the fungal mycelia present on the particles to grow into separate colonies. The growth of fungi was visible on the second day and isolations were done uniformly on the second, eighth and fifteenth day of incubation. It was an exercise with ease the individual colonies could be picked up. In order to restrict the repetitive appearance of similar fungi, the isolates were first transferred to MEA plates cut into separate sectors and only representative colonies showing distinctiveness in colony morphology and cultural characters were housed in MEA slants. While the isolates grew into mature colonies in the slants, these fungi were identified.

The exercise resulted with the recovery of a total of 5000 individual colonies (Table 4.2.1). These were assignable to 228 species of fungi belonging to 120 genera which included Mucorales, Hyphomycetes, Coelomycetes and Ascomycetes (Table 4.1).

In the isolation of endophytic fungi, the mycelia growing along the edges of leaf bits were picked up on the 7th and 15th days of incubation. In similar studies carried out in the temperate countries (Fisher and Petrini, 1988), it was reported that the endophytic fungi commenced growth only after eight days of incubation. The delay in commencement of growth was attributed to the sudden shift from endophytic (within the leaf) to epiphytic (on the agar medium) environment. Such a delay in colony growth was not observed in this study and fungal colonies were visible even on the fourth day.

For statistical analysis of their occurrence, the isolates of fungi recovered during the study were segregated into following 9 broad groups (G1 - G9) based on the colony characters, spore ontogeny and pigmentation (Carmichael *et al.*, 1980; Hawksworth *et al.*, 1995).

Moniliaceous phialidic (G1): These were deuteromycetous fungi belonging to moniliaceous Hyphomycetes. The colonies/mycelia of these were white and with or without exudates. The conidiogenous apparatus were phialidic and colourless. The conidia were hyaline and slimy or dry.

G2 - <u>Dematiatious phialidic</u>: These were deuteromycetous fungi belonging to dematiaceous (=pigmented) Hyphomycetes. The colonies/mycelia of these were variedly coloured and pigmented, ranging from pale brown to black and with or without exudates. The conidiogenous apparatus were phialidic and dematiaceous. The conidia were pigmented, slimy or dry.

- G3 <u>Moniliaceous blastic</u>: Deuteromycetous fungi belonging to moniliaceous Hyphomycetes. The colonies/mycelia of these were white and with or without exudates. The conidiogenous apparatus and blastic conidia were colourless. The conidia were dry.
- G4 <u>Dematiaceous blastic</u>: Deuteromycetous fungi belonging to dematiaceous Hyphomycetes. The colonies/mycelia of these were variedly coloured and pigmented, ranging from pale to black and with or without exudates. The conidiogenous apparatus were dematiaceous and blastic. The conidia were dry.
- G5 <u>Dematiaceous tretic</u>: Deuteromycetous fungi belonging to dematiaceous Hyphomycetes. The colonies/mycelia of these were variedly coloured and pigmented, ranging from pale to dark and with or without exudates. The conidiogenous apparatus were dematiaceous and tretic. The conidia were slimy or dry.
- G6 Moniliaceous nonsporulating forms: In the absence of any spores or spore-bearing strucures, the taxonomic identify of these morphotypes was not known. In other words, these could be members of any taxonomic Division in the fungal kingdom. The colonies/mycelia of these were white and with or without exudates. These fungi did not sporulate in culture and remained sterile throughout the period of the study.
- G7 <u>Dematiaceous nonsporulating forms</u>: Similar to G6, in the absence of any spores or spore-bearing strucures, the identify of these morphotypes also was not known; they could be members of any taxonomic Division. The colonies/mycelia were variedly coloured and pigmented, ranging from pale to dark and with or without exudates. These fungi did not sporulate in culture and remained sterile throughout the period of the study.

G8 and G9 - Colourless and pigmented miscellanous forms: These belonged to Mucorales, Ascomycetes and Coelomycetes and were considered together as miscellaneous However, as in other groups, colour of the colonies/mycelia and fruitng structures such as sporangiophores and sporangia in case of mucorales, ascocarp and ascospores in the ascomycetes and pycnidia and pycnospores in case of coelomycetes were highlighted into two separate entities: colourless (G8) and pigmented (G9).

During the period of study, the seasons were considered as follows:

Period (months in 1997-98)	Season	Treatment
November - February	Winter	S1
March - May	Summer	S2
June - October	Monsoon	S 3
November - February	Winter	S4
March - May	Summer	S5
June - October	Monsoon	S 6

The data on fungal isolates obtained during the 24 month study were subjected to '3- factor factorial completely randomised design analysis' defined in the Statistical Package for Social Sciences to analyse the significance of fungi with reference to group, impact of the seasons, relation of the substrates, effect between groups and seasons, relation between groups and substrate, effect between seasons and substrate and impact of interaction beween the groups, substrates and seasons.

The results obtained are given in Table 4.2.1. explained below and the relavent ANOVA tables are given in the appendix

(i) Group effect:

Analysis of variance tests showed the interaction amongst various fungal groups as highly significant. The average number of fungal isolates was highest for G2: dematiaceous phialidic (0.9147), followed in a descending order of significance by G9: dematiaceous miscellaneous (0.8905), G4: dematiaceous blastic (0.8211), G1: moniliaceous phialidic (0.8166), G5: dematiaceous tretic (0.7721), G6: moniliaceous nonsporulating forms (0.7709), G7: dematiaceous nonsporulating forms (0.7515), G8: moniliaceous miscellaneous (0.7447) and represented least in G3: moniliaceous blastic (0.7221).

(ii) Effect of seasons:

ANOVA tests showed, on comparison, seasonal effect to be highly significant. In both years of analysis, abundance of fungi followed a simillar trend although the average number of fungi isolated in 1998 was slightly lower than in 1997. The recovery of highest number was during monsoons: S3 and S6 (0.8234 in S3 and 0.8106 in S6), followed in by the post monsoon months or winter (0.8080 in S1 and 0.8004 in S4) and the least during summer (0.7823 in S2 and 0.7780 in S4)

(iii) The substrate effect:

Substrate effect on ANOVA tests appeared to be highly significant. The average number of fungi isolated from leaf litter was the maximum (0.8188), followed closely by soil (0.8173), fresh leaf (0.7885) and the least by air (0.7676).

(iv) Group vs season effect:

Dematiaceous phialidic group showed the highest average(1.0218) of fungal propagules in the monsoon months of 1997 while moniliaceous blastic group showed

the least number (0.7164) in the monsoon months of 1997 and 1998 and summer months of 1998 when subjected to ANOVA for comparisons of group and seasons interactions.

(v) Group vs substrate effect:

The comparison of interaction amongst groups and substrate as revealed by ANOVA is highly significant. Moniliaceous phialidic group on soil was the highest (1.0063) and moniliaceous blastic group on air and soil and moniliaceous miscellaneous fungi on air were the least (0.7071).

(vi) Season vs substrate effect:

The result of ANOVA on interactions amongst season and substrate appeared to be highly significant. Soil in the monsoon months of 1997 showed to harbour the highest average of fungal propagules (0.8658) followed by in the post monsoon months of 1997 (0.8616), leaf litter in the monsoon months of 1998 (0.8399) and (0.8362) for 1997 and soil in monsoon of 1998 (0.8300), leaf litter in the post monsoon months/winter of 1998 (0.8158), fresh leaf in monsoon of 1997 (0.8124) and the least in air in the post monsoon months/winter of 1998.

(vii) Group - substrate - season effect:

Three way interactions among group, season and substrate revealed a great significance from ANOVA. The presence of dematiaceous phialidic fungi on fresh leaves in the monsoon months of 1997 was the highest (1.233) while dematiaceous tretic typeon leaf litter in the summer months showed the least of fungal isolates (0.0855).

Analysis of endophytic fungi:

The 3² factor factorial completely randomised design showed the distribution and diversity of endophytes in relation to plant parts. The two plants, *Carissa congesta* and *Ficus benghalensis*, were subjected to the design analysis by considering different parts of the fresh leaf as 'Factor-1' and seasons as 'Factor-2'. Effect of these factors and their interaction were studied using ANOVA and the results are explained below:

Factor-1:

Part I

Base of the leaf, including petiole

Part II

Middle portion of leaf

Part III

Tip of the leaf

Factor-2:

Season I

November-February (Post Monsoon/Winter)

Season II

March - May (Summer)

Season III

June -October (Monsoon)

A. Impact on Carissa congesta (Table:4.2.2; relavent ANOVA tables given in the appendix)

a) Moniliaceous phialidic (G1):

The basal area of the leaf showed highest average of fungal isolates (0.7731) followed by the middle portion (0.7208) and the tip(0.7071). Further, the basal part of the leaf exhibited highest average of fungal isolates in the monsoons (0.8640), followed by in the winter and summer (0.7483). The least average of fungal isolates (0.7071) was seen in the basal part in the winter, the middle in summer and monsoons and the tip portion in all the seasons.

b) Dematiaceous phialidic (G2):

The basal part of the leaf showed highest average of fungal isolates (1.4798), followed by the middle (1.0456) and the tip part of the leaf (0.8662). The highest average of fungal isolates were observed in summer (1.4275), followed by monsoons (1.0783) and winter (0.8858). The highest number of fungal isolates on the basal part of the leaf (2.1774) was in the summer, followed by the basal region in the monsoons (1.2988). Middle parts showed moderate values of of fungal isolates in the monsoon and summer(1.1223 and 1.1079). The least number was observed on the tip portion in winter.

c) Moniliaceous blastic (G3):

The highest average number of fungal isolates of this group was in the monsoon months (0.8106) followed by summer and winter (0.7071).

d) Dematiaceous blastic (G4):

Interactions amongst different leaf parts, seasons and different parts and seasons in ANOVA tests were highly significant. The basal parts of the leaf showed the highest average number of fungal isolates (0.9729), followed by the middle part (0.8834) and the tip (0.7516). The highest average of fungal isolates was observed in the monsoon (0.9503), followed by in the winter (0.8494) and the least in the summer (0.8081). The highest average number of fungal isolates was on the basal part of the leaf in monsoonmonths, followed by the middle part in the monsoon (0.9652) and the least on the tip of the leaf in summer (0.7243).

e) Dematiaceous tretic (G5):

The basal part of the leaf showed the highest average number of fungal isolates (0.7657) followed by in the middle and tip part (0.7071).

f) Moniliaceous nonnsporulating morphotypes (G6):

The basal part of the leaf showed highest average for the number of fungal isolates (0.7757), followed by the middle and tip part (0.7071).

g) Dematiaceous nonsporulating morphotypes (G7):

The basal part of the leaf showed the highest average number of fungal isolates (0.7966), followed by the middle (0.7603) and the least on the tip of the leaf (0.7144).

h) Moniliaceous miscellaneous (G8):

ANOVA tests showed no significance at all.

i) Dematiaceous miscellaneous group (G9):

The highest average number of fungal isolates were observed on the basal part of the leaf (1.336), followed by the middle (1.113) and the least on the tip of the leaf (0.7296). The highest average number of fungal isolates was observed on the basal part in summer (1.4395), followed by in the monsoon (1.3314) and winter (1.2371). Moderate average numbers of fungal isolates were observed on the middle part, the highest being in the monsoon (1.1796). The least was observed on the tip part of the leaf in the monsoons (0.7206).

B. <u>Impact on Ficus benghalensis:</u> (Table: 4.2.3 with relavent ANOVA tables given in appendix)

j) Moniliaceous phialidic (G1):

ANOVA test did not show any significance.

k) Dematiaceous phialidic (G2):

The interactions among the different parts of the leaf and among the different seasons was highly significant as revealed by ANOVA. The middle part of the leaf showed highest average number of fungal isolates (0.9216), followed by the basal part (0.86) and the tip (0.7217). The highest average number of isolates was obtained in the monsoon (0.9362), followed by in winter (0.8107) and the least in summer (0.7564).

1) Moniliaceous blastic (G3):

This group of fungi was not represented in the recovery exercise and therefore not subjected to analysis.

m) Dematiaceous blastic (G4):

ANOVA exhibited significance for both the main effects, that is, different parts of the leaf and the seasons, as well as the interactions between the different parts of the leaf and the seasons. The basal part showed highest average number of fungal isolates (0.9468), followed by the middle part (0.8976) and the least average number of isolates were in the tip region of the leaf (0.7406). The highest average number of fungal isolates were obtained in winter (0.923), followed by in the monsoon (0.9032) and the least in the summer (0.759). The highest average number of fungal isolates was observed on the basal part of the leaf in winter (1.0485) followed by in the monsoon (1.0003). A similar trend but moderate average numbers of fungal isolates was observed in the middle part in winter (0.9774), followed by in the monsoon (0.9520) and the least (0.7216) was in the summer on the tip of the leaf.

n) Dematiaceous tretic(G5):

ANOVA test did not show any significance for this group.

o) Moniliaceous nonsporulating forms (G6):

The test showed significance for the interaction between the different parts of the leaf. The basal part showed a higher average number of fungal isolates (0.8614), followed by the middle part (0.7731) and the least in the tip of the leaf (0.7483).

p) Dematiaceous nonsporulating morphotypes (G7):

The basal part of the leaf showed a higher average number of fungal isolates (0.8074), followed by the middle (0.7308) and the least in the tip portion of the leaf (0.7137).

q) Moniliaceous miscellaneous (G8):

ANOVA test did not show any significance.

r) Dematiaceous miscellaneous (G9):

ANOVA test showed significance for both the main effects studied, that is the different parts of the leaf and the interaction among the different seasons studied. The basal parts of the leaf showed the highest average number of fungal isolates (1.0469), followed by the middle part(0.9037) and the least by the tip part(0.7201). The highest average number of fungal isolates was observed in the monsoon (0.9125), followed by the summer (0.9010) and the least in the winter (0.8572).

Association of fungi on flowers and fruits of Carissa congesta and fruits of F. benghalensis:

Flower and fruits of *C. congesta* appear during the months of March to August. While the fruits of *F. benghalensis* appear between the months of December to July. The results are computed accordingly.

The flower and fruits of *C. congesta* and *F. benghalensis* were plated to assess the diversity and abundance of microfungi on the floral parts of these plants. The flowers and fruits were teased apart (pedicel, calyx and corolla), surface sterilized in 70% ethanol for 10secs, 50secs in Sodium hypochlorite followed by 10secs in 70%ethanol a second time. The surface sterilized bits were plated and fungi isolated as when they appeared. The list of fungi that appeared on the floral parts of *C. congesta* is given below.

Table: 4.2.4 List of fungi on the floral parts of C. congesta

Acremonium sp.1

Acremonium sp.2

Ascochytula sapindae

Botryodiploidea theobromae

Camarosporium indicum

Chaetomium nigricolor

Cladosporium sp.2

Curvularia trifolii

Diatrypella indica

Fusarium decemcellulare

Fusarium culmorum

Gliomastix murorum

Nigrospora sphaerica

Paecilomyces sp.3

Pestalotiopsis palmarum

Pestalotiopsis versicolor

Undetermined hyphomycete sp.8

NS. 23

NS. 72

NS. 87

NS. 91

NS. 121

Seventeen species belonging to 14 genera and 5 nonsporulating forms were recovered. Of these 10 species in 8 genera were hyphomycetes, 5 species in 4 genera were coelomycetes, and 3 species in 3 genera of ascomycetes. The highest number of species was found on the pedicel, followed by on the calyx and the least on the corolla (Fig. 4.2.1) A high number of species was obtained in the months of March to April and a lesser number in May (Fig. 4.2.2)

The fruits of *C. congesta* harbored a total of 3 species of fungi, namely, *Acremonium* sp.2, *Cladosporium* sp.2 and *Fusarium decemcellulare*.

The fruits of F. benghalensis also harbored 3 species, namely, Acremonium sp.1, Cunninghamela echinulata and Fusarium decemcellulare.

PART 3: <u>ANALYSIS OF THE FUNGAL ISOLATES FOR ENZYME</u> <u>ACTIVITIES</u>:

Of the 60 isolates subjected to enzyme assay, 29 showed positive activity against lignin and these were subjected for laccase and xylanase activity tests.

The results of each of these qualitative enzyme tests are given in Table: 4.3.1-4.3.7

The results showed that on the whole maximum percentage of isolates exhibited protease (63.3%) activity, followed by laccase (62.1%), pectinase (52%), ligninase

(48.3%), amylase (33.3%), xylanase (31%) and the least number (15%) were with cellulase activity. The total isolates associated with *Carissa congesta* showed highest percentage for cellulase activity (88.9%), followed by amylase (75%), ligninase (72.4%), protease (71%), pectinase (68%), xylanase (66.7%) and laccase activity (61.1%). The total positive isolates associated with *Ficus benghalensis* showed high percentage for laccase (50%), ligninase (48%), pectinase (45.2%), amylase (45%), xylanase (44%), protease (42%) and the least of cellulase (22%) (Table 4.3.8).

The isolates tested for laccase assay showed promising results and therefore all the 18 fungi were subjected to further test and quantitative analysis. These included 9 hyphomycetous fungi, namely Beltraniella buloloensis, Cladosporium sp.2, Cylindrocarpon ianthothele, Fusarium decemcellulare, F. phragmitis, Gliomastix murorum, Periconia byssoides and Undetermined hyphomycete sp.13, 4 coelomycetous fungi, namely Ajrekarella polychaetriae, Ascochyta caricae, Camarosporium indica and Coniochaeta fuckelii. The remaining 5 isolates were nonsporulating forms labelled as NS. 47, NS 34, NS.121, NS. 45, and NS. 48 (Table 4.3.9)

The highest activity of laccase was exhibited by the isolate NS. 48 (1166.6nkat), followed by NS. 47 (183.5nkat), Camarosporium indicum (140.5nkat), Coniochaeta fuckelii (110.1nkat), Periconia byssoides (78.6nkat), Undetermined hyphomycete sp.13 (48nkat), Ajrekarella polychaetriae (46.9nkat), Ns. 45 (26.3nkat), Gliomastix murorum (26.1nkat), Cylindrocarpon ianthothele (23.4nkat), Sporidesmium altum (22.2nkat), Ascochyta caricae (19.3nkat), Fusarium phragmitis (19.2nkat), Cladosporium sp.2 (18.9nkat), NS. 34 (16.2nkat), Fusarium decemcellulare (14.5nkat), Beltraniella buloloensis (13.5nkat) and the least by NS. 121 (4.7nkat).

The largest group showing laccase activity were the isolates tested on the 5th day (Fusarium phragmitis, NS. 47, Cylindrocarpon ianthothele, Beltraniella buloloensis, NS 34, Ascochyta caricae, and Periconia byssoides); followed by 3rd day (Coniochaeta fuckelii, Gliomastix murorum, Sporidesmium altum, NS.121, and Fusarium decemcellulare), followed by 11th day (Cladosporium sp.2, Camarosporium indicum and NS. 45); followed by 7th day (NS. 48); followed by 9th day (Periconia byssoides); followed by 13th day (Undetermined hyphomycete sp.13) and by 15th day (Airekarella polychaetriae).

The fungal isolates showing the highest laccase activity (1166.6nkat) and the least activity (4.7nkat) both were isolated from Carissa congesta. Further, the maximum number of fungal isolates showing laccase activity were also isolated from C. congesta alone (viz., NS. 47, Ajrekarella polychaetriae, Cladosporium sp.2, Undetermined hyphomycete sp.13, NS 34, Ascochyta caricae and NS. 45) followed by those that were isolated from and common to both plant species (viz. Fusarium phragmitis, Cylindrocarpon ianthothele, Coniochaeta fuckelii, Gliomastix murorum and Fusarium decemcellulare. The least number of strains showing laccase activity were those isolated from Ficus benghalensis which included Sporidesmium altum, Camarosporium indicum and Periconia byssoides.

Of the isolates investigated, maximum number of strains showed laccase activity in the range of 0 - 20 nkat. Of these, 5 were isolated from Carissa congesta and two strains common to both, C. congesta and Ficus benghalensis. This was followed by 6 strains in the range of 21-50nkats. Of these 3 were isolated from Carissa congesta, 1 from Ficus benghalensis and 2 strains common to Carissa congesta and Ficus benghalensis. Only one strain isolated from F. benghalensis showed activity in the range of 51-80 nkats. One strain isolated from both Carissa

congesta and Ficus benghalensis showed activity in the range of 81-110nkats. In the range of 111-140 nkats a single isolate from F. benghalensis showed activity while single strains isolated from Carissa congesta showed activity in the range of 171-200nkats and 1161-1190nkats each (Table 4.3.10).

Quantitative analysis for laccase on fungi associated with *Carissa congesta* showed that 77.7% of fungal isolates had low activity (0-380nkat), while 5.5% showed high activity (771-1190nkat). Analysis for laccase activity on fungi associated with *Ficus benghalensis* showed that 44.45% of fungal isolates showed low activity (0-380nkat) while none showed moderate or high activity (Table 4.3.11).

Based on this study, the relation between different enzymes was analysed using Statistica ver. 5 by employing cluster analysis and subjecting the data to unweighted pair group average by using Euclidean distances (Fig. 4.3.1 and Table 4.3.12) shows that amylase and cellulase are more closely related than ligninase, laccase and xylanase. And these two clusters are more closely related to each other and they are also related to pectinase. All these branches in turn are distantly related to protease. The tree diagram also shows that amylase and cellulase are most distantly related to protease.

The relationship between different fungi and the enzymes they produce and the correlation between the two are analysed by subjecting 60 isolates of fungi to cluster analysis following the Tree Joining Linkage (unweighted pair-group average, Euclidean distances)(Fig. 4.3.2). The study resulted in 12 groups of clusters. The main branch of the tree has three nonsporulating forms of fungi, NS. 30, NS. 46, and NS. 120. These fungi are related in the similarity that they produced amylase, protease and cellulase. This formed the first cluster.

This branch is related to a larger group consisting of several clusters, the first of these consists of *Ajrekarella polychaetriae* and NS. 121. And these 2 are related that they produced protease, ligninse, laccase and xylanase. This cluster is closely related to the group formed by NS. 2 and *Cylindrocarpon ianthothele* wherein pectinase and ligninase are produced by both these strains. This in turn is related to the strains NS. 45, NS. 39 and *Beltraniella buloloensis* which commonly produced the following enzymes protease, ligninase and laccase. This formed the second cluster.

Botryodiploidea theobromae, Gliomastix murorum and Aspergillus sp.7 produced several enzymes, however the common one is amylase. This formed the third cluster.

Nonsporulating form (NS) 32, Fusarium decemcellulare, and Undetermined sp.12 and Cladosporium sp.2 similarly produced ligninase and xylanase. Sporidesmium altum, Nigrospora sphaerica, NS. 119 and NS. 34 produce ligninase and laccase commonly. NS.42, Cladosporium sp.5, NS. 33, NS. 118 and NS. 37 produced pectinase besides other enzymes. Neocercosporella indica, NS. 35, NS. 40, Scolecobasidium humicola and Gonytrichum sp. are similar in the fact that they produced protease. Nonsporulating form 117 produced only amylase. Chaetomium nigricolor, NS.38, Scolecobasidium constrictum, Mucor flavus, Nigrospora endophytica, and Trichoderma sp.1 produced no enzymes. Fusarium tabacinum and Fusarium solani produced only pectinase. This formed the fourth cluster.

Fusarium phragmitis, NS. 116 and Ascochytula sapindae produced ligninase besides protease in the first two strains. Camarosporium indicum, Periconia byssoides, Weisneiriomyces javanicus produce amylase, pectinase and ligninase commonly, while Trichocladium album produced only pectinase and ligninase. This formed the fifth cluster.

Nonsporulating form (NS) 36, Ascochyta caricae, NS. 31, Aspergillus sp.6, Coniochaeta fuckelii, Arxiella terrestris and Chaetomella cycadina produced pectinase and protease commonly besides other enzymes. This formed the sixth cluster.

Nonsporulating form 41 produced amylase, pectinase, protease, cellulase and ligninase. This is related to Beltrania rhombica, NS. 49 and NS. 48 which produces pectinase, protease and ligninase. This formed the seventh cluster.

As seen from the Fig. 4.3.2, cluster 3 to cluster 6 are related. This is referred as cluster 8. Cluster 8 is related to cluster 7. This cluster is 9. Cluster 9 is related to cluster 2. This is called cluster 10. Cluster 10 is related to cluster 1. Cluster 1 and 7 are most distantly related.

DISCUSSION:

1. Diversity of fungi on two plants in relation to their neighbourhood:

As can be seen from the results on the diversity studies on fungi, a large number of taxa of fungi have been isolated. In all, 228 species of fungi belonging to 120 genera, which included Mucorales, Hyphomycetes, Coelomycetes and Ascomycetes (Table 4.1.1) were recovered. Of these, 177 taxa belonged to Hyphomycetes and are accommodated in 81 genera. Sixteen hyphomycetous fungi were not assigned to any known taxa since these were unlike any known species of fungi and appropriate literature was not available for identification. Four species belonging to 2 genera of Mucoraceous fungi encountered during the study are described. A total of 12 species belonging to 10 genera of Ascomycetes and 35 species belonging to 27 genera of Coelomycetes are only listed out. A total of 121 isolates did not sporulate in culture or on the substrate and therefore they are not

described in detail but recognised only as 'nonsporulating forms'. These are recognised as morphotypes based on cultural characters such as colour, shape and size of the colony and presence or absence of exudates, but in the absence of any sporulating features, the taxonomy of these fungi remained undetermined.

Of the 228 species of fungi recovered, 22 species and 5 genera were new to science. These are the following: Acronidiellina indica sp. nov., Cercospora carriseae sp. nov., Cirrenalia indica sp. nov., Dicyma carisseae sp. nov., Doratomyces indicus sp. nov., Gonatobotryum bimorphospora sp. nov., Hyaloscolecobasidium indicum Gen. et sp. nov., Idriella mucoidea sp. nov., Idriella multiseptata sp. nov., Kumbhamaya indica Gen. et sp. nov., Moorella ficusensis sp. nov., Neocercosporella indica Gen. et sp. nov., Nigrospora endophytica sp. nov., Paracylindrocladia indica Genb. et sp. nov., Parahumicola endophytica Gen. et sp. nov., Phialocephala carisseae sp. nov., Phialocephala nephrospora sp. nov., Phialomyces microsporus sp. nov., Sclerographium goanensis sp. nov. and Spadicoides indicus sp. nov.

Undoubtedly this is an extremely large number of named and new taxa of fungi appeared in any known similar studies carried out earlier elsewhere. In the present study, two techniques, moist-chamber incubation and particle plating, were used for isolation of fungi. It is possible that partile-plating technique, which was introduced to studies such as this only recently, allowed the recovery of maximum diversity of fungi from the leaf particles wherein several unknown fungi also exhibited themselves. A total of 16 taxa of well sporulating but unknown fungi have been recovered in this exercise.

In this investigation, many common genera of litter fungi belonging to Hyphomycetes (Beltrania, Beltraniella, Helminthosporium, Idriella, Phialocephala, Gyrothrix, Periconia, Scolecobasidium, Vanakripa, Volutella, Weisneriomyces and Zygosporium), Coelomycetes (Ajrekarella, Ascochytula, Botryodiplodia, Camarosporium,

Coniochaet, Diplodia, Discosia, Lasidiploidea, Monochaetia, Neottiospora, Phyllostica, Robillarda, Seimatosporium, Stagonospora, Trichosprerma and Vasudevella), Ascomycetes (Diatrype, Diatrypella, Hypoxylon, Guignardia, Lophiostoma and Xylaria), Mucorales (Mucor) and sterile strains were recovered. Several conventionally uncommon genera such as Aspergillus, Curvularia, Fusarium, Paecilomyces, Penicillium, Tritirachium, Torula, Botryodiplodia, Chaetomella, Chaetomium, etc. which are generally referred as soil fungi were also encountered in the litter.

In the particle-plating technique followed, the particles were repeatedly washed in tap water and sterile distilled water before being spread on isolation media plates. Chances of soil fungi appearing along with the particle-inhabiting litter fungi are therefore very less. It can be safely deduced from the results that the fungi so far considered as soil flora are new records for the litter.

The species diversity estimation studies revealed that there are a few abundant species and a high proportion of rare species of fungi associated with the two plant species. The fungi present on all substrates, belonged to 17 genera of Hyphomycetes and 3 Coelomycetes. In all, 121 nonsporulating forms were recovered. The identity of these is not known. From careful observations, it is stated here that these nonsporulating forms also exhibited common and distinctiveness in their cultural appearance on both plant and plant parts, air and soil (Fig. 4.2.9)

A sizable number of endophytic fungi isolated from leaves of both plants at both locations appeared in the litter layer as well. These include Alternaria alternata, Arxiella terrestris, Aspergillus sp.6, Beltrania rhombica, Cercospora carriseae, Curvularia crepinii, C.lunata, Cylindrocladium parvum, Fusarium decemcellulare, F. solani, Gliomastix murorum, G. uniseptata, Hyaloscolecobasidium indicum, Idriella fertilis, I. lunata, I. mucoidea, I. ramosa, Nigrospora sphaerica, Nodulisporium honiaraense, Paecilomyces sp.1 and3, Pseudobotrytis terrestris, Scolecobasidium constrictum, S.humicola, S.triangularis, S.

variabile, S. verrucosum, S.longisporum, Stachybotrys atra, Wiesneriomyces javanicus, a few Undetermined Hyphomycete taxa, Ascochytula sapindae, Botryodiplodia theobromae, Discosia atroceras, Pestalotiopsis palmarum, Phomopsis filiformis, Robillarda sp., Septoria arcuata, Guignardia sp. and Xylaria sp.

This observation was similar to the studies made by Boddy and Griffith, (1989), Parkinson and Kendrick (1960), Wildman and Parkinson (1979) and Bills and Polishook (1994) wherein they found that endophytic fungi are often recovered from the upper layers of forest litter and are associated with litter decomposition. It can be stated that the endophytes, which survive as latent internal colonizers of fresh leaves later take an active part in the degradation of organic matter in the litter. This kind of shift in function, from hidden colonization to active degradation, may be considered as an ecological adaptation associated with litter decomposition. It is also possible that some of the litter degrading fungi might have adjusted and taken refuge as endophytes during some part in the year. There are recent studies indicating that the endophytes are certainly not veiled refuges in the leaf tissues but an active defence system against herbivory by insects and higher animals (Petrini, 1986).

From the results obtained it is clear that collecting localities spaced apart as organised in this work has no major impact to offer. The density of species at different seasons was similar in both localities (Fig.4.2.5).

2. Interaction of fungi with the substrate in relation to season:

As can be seen from the analysis of group effect, the most dominant group of fungi in this study was the dematiaceous phialidic group (G2) and the least was moniliaceous blastic (G3). The coloured fungi in general (G2, G9 and G4) were the dominant groups. In earlier studies on litter fungi (Vittal, 1973; Sudha, 1978) similar results were obtained. As in other plants, colour or pigments in fungi confers

protection from stress of light and other harmful effects. It is therefore apparent that dematiaceous or pigmented fungi were well adapted as dominent litter colonisers (Fig. 4.2.6)

As revealed in this study, seasonal effect on the occurrence of fungi is highly significant. The recovery of fungi, especially the dematiaceous phialidic forms, was highest during monsoons followed by the post monsoon months and the least in summer. It seems natural that high humidity combined with high ambient temperature of the monsoon months in this part of the tropical region favoured growth and sporulation of a large number of fungi on plant substrates. In a recent study, however, Bhat and Kaveriappa (1999) indicated that occurrence of litter fungi was highest during premonsoon summer months (Fig. 4.2.7)

As in similar other studies (Sudha, 1978), the number of fungi isolated from leaf litter of both plants was more than those on soil and in air. Along with the soil and air mycoflora, the endophytic fungi represented in low magnitude. The complete decomposition of leaf litter is the combined result of colonisation by a large number of fungi, besides other microorganisms. Although fungi visit other substrates and environment in the neighbourhood such as the soil and air, the eventual site of survival for dematiaceous fungi is the litter. It is therefore not surprising to come across a large number of fungi in the leaf litter.

3. Endophytic fungi in different plant parts:

In Carissa congesta, the basal part of the leaf showed maximum diversity of ascomycetous and hyphomycetous fungi, during pre-monsoon and monsoon months, than in the middle and tip portion, whereas in winter and summer months the density of fungal species was less. In Ficus benghalensis, during pre-monsoon and monsoon,

higher density of hyphomycetous species was observed in the middle part than the tip and basal regions of the leaf. The ascomycetous fungi were in high density in the basal parts of the leaf.

In the structure and dynamics of leaf growth and function, basal portion is the most enduring part and it is natural to expect occurrence of maximum species diversity of fungi in this part. Endophytic fungi get into the leaf tissue at an early period of leaf growth (Fisher *et al.*, 1986; Petrini and Fisher, 1986) and it is also possible that during the course of its colonisation, the fungi consume considerable time and apparently are unable to reach the tip or edge of the leaf.

The fruits and flowers of both plants had their mycoflora to exhibit. In general, these fungi were those, which appeared in litter or soil sometime, or the other.

4. Enzyme activities:

Of the 60 isolates subjected to enzyme assay, 29 showed positive activity against lignin and these were subjected for laccase and xylanase activity tests. The results of each of these qualitative enzyme tests are given in Table No. 4.3.1-4.3.7. The results showed that on the whole maximum percentage of isolates exhibited protease activity, followed by laccase, pectinase, ligninase, amylase, xylanase and the least were with cellulase activity. The total isolates associated with *Carissa congesta* showed highest percentage for cellulase activity, followed by amylase, ligninase, protease, pectinase, xylanase and laccase activity. The total positive isolates associated with *Ficus benghalensis* showed high percentage for laccase, followed by ligninase, pectinase, amylase, xylanase, protease and the least of cellulase.

The isolates tested for laccase showed promising results. These included 9 Hyphomycetes (Beltraniella buloloensis, Cladosporium sp.2, Cylindrocarpon

ianthothele, Fusarium decemcellulare, F. phragmitis, Gliomastix murorum, Periconia byssoides and Undetermined hyphomycete sp.13) and 4 coelomycetous fungi (Ajrekarella polychaetriae, Ascochyta caricae, Camarosporium indica and Coniochaeta fuckelii). The remaining 5 isolates were nonsporulating forms labelled as NS. 47, NS 34, NS.121, NS. 45, and NS. 48 (Table: 4.3.9)

Some of the isolates are powerful producers of laccase. Examples from this study include the isolate NS. 48, NS. 47, Camarosporium indicum, Coniochaeta fuckelii, Periconia byssoides, Undetermined hyphomycete sp.13, Ajrekarella polychaetriae, NS. 45 and Gliomastix murorum. The largest group showing laccase activity were the isolates tested on the 5th day (Fusarium phragmitis, NS. 47, Cylindrocarpon ianthothele, Beltraniella buloloensis, NS 34, Ascochyta caricae, and Periconia byssoides), followed by 3rd day (Coniochaeta fuckelii, Gliomastix murorum, Sporidesmium altum, NS.121, and Fusarium decemcellulare).

The fungal isolates showing the maximum number and highest and least laccase activity were isolated from Carissa congesta. Very few of the isolates from Ficus benghalensis exhibited laccase activity. Of the isolates investigated, maximum number of strains showed very low laccase activity, in the range of 0-20 nkat. Five of these were from Carissa congesta and two strains common to C. congesta and Ficus benghalensis. Six strains were in the range of 21-50nkats activity. Of these 1 was from F. benghalensi, 3 from Carissa congesta and 2 strains common to both plants. Only one isolate from F. benghalensis showed activity in the range of 51-80nkats. One strain isolated from both F. benghalensis and Carissa congesta and showed activity in the range of 81-110nkats. In the range of 111-140nkats a single isolate from F. benghalensis showed activity. Sngle strains isolated from Carissa congesta showed activity in the range of 171-200nkats and 1161-1190nkats.

Quantitative analysis for laccase of fungi associated with Carissa congesta showed that nearly 78% of fungal isolates had low, while about 6% showed high activity. Analysis for laccase activity on fungi associated with Ficus benghalensis showed about 45% of fungal isolates with low and none with neither moderate nor high activity.

The results of enzyme studies on subjecting to cluster analysis showed that some of these were grouped together while some remained wide apart in their activity. The analysis helped to estimate the extent of enzyme activity of the fungi. For example, *Beltrania rhombica*, *Beltraniella bulolensis* and *Weisneriomyces javanicus* produce several enzymes and this is an indication that they are the colonizers and powerful degraders of litter.

All these assays indicate that fungi are armoured with a variety of enzymes, which aid them in their saprophytic mode of activities. Isolation of fungi alone will not give any indication of their activities. These fungi will have to be tested both qualitatively and quantitatively for enzymes, as done in this work. Only on assaying individual plants and associated each fungus, one can build a profile of their enzymes.

Although the enzymes of fungi are for their own growth and sustenance, these can be used for advantage. Building up an 'enzyme profile catalogue' along with collection of 'pure cultures' and setting up of a 'taxonomy database' are all positive indicators in an effort of conservation and utilization of fungal biodiversity in our reduced country. Further, study such as this will have a far-reaching implication in our efforts of building up of biotechnology resource supply bank in India.

Epilogue:

Hitherto diversity, ecology and creativity studies on fungi of India, barring a few detailed investigations, were cursory in nature and too inadequate both in space and time. Comprehensive floristic surveys on saprophytic and endophytic fungi of plant and plant parts, as carried out in this work, are very few. Documentation and maintenance of pure cultures of fungi in state-of-the-art culture collections are not attempted at all. Screening of fungi for enzymes and other secondary metabolites were so far not an organized industry in our country.

Progressive pharmaceutical, agriculture, food and dye industries are now concentrating their effort on finding out the useful properties of microbial secondary metabolites, especially enzymes and other properties elaborated by fungi of litter, endophyte, dung and other less-known substrates and habitats. The knowledge on the extent of fungal diversity and their ecological and chemical function is therefore of paramount importance. It is therefore necessary and urgent that using efficient techniques such as the 'particle-plating', besides the conventional moist-chamber incubation, the fungi should be isolated in pure form from all substrates and habitats, maintained in culture repositories and documented in a systematic manner, as illustrated in this work. Such documented information and culture facilities established of the fungal-resources will be very useful and invaluable for future biotechnology and utilization in our country.

To achieve high fungal sourcing from habitats and substrates that deem to harbour diverse species, the methods of sampling, isolation and documentation should be efficient and ingenious. The methodology adapted in this work could be a model for future fungal systematics in our country. The results obtained in this work in the form of maximum recovery of diverse species, elucidation of a sizable number of new taxa, documentation of informations on the enzyme profile and eventually housing of

a large collection of pure isolates of fungi in a culture collection - are all of no small measure. From this point, the work carried out in this thesis is a novel contribution. The strength of this work lies on the information documented on the biodiversity of fungi of India with special reference to this part of the country, better understanding on the ecological relation of occurrence of fungi on plants and their neighbourhood and the enzyme profile built up for several of those isolated fungi. All these are small but valuable additions to the knowledge and growth of science in our country.

Investigations on the basis of biology of fungi, through systematic surveys, taxonomic characterization, establishment of pure cultures, unraveling the ecological relations with habitats and substrates and elucidation of fungal creativity, all have led to understanding of fungi in a big way which are useful even from the industrial point. For example, strain selection and indentity of taxa are an important and crucial first step in the screening and chemical characterization programme of the biotechnological industries.

The fungi are producers of extremely potent natural and semisynthetic compounds of high therapeutic value such as cephalosporin, cyclosporin, mevinolin, paspalitrems, tremorgenic neurotoxins, taxol and so on. A basic axiom that would guide the search for chemical diversity among organisms from nature is that elaboration of specialized metabolites is often genotypically specific, or atleast taxonomically limited, and therefore, chemical creativity may be sought by surveying metabolic products of diverse and substrate- or habitat-specific organisms. It is believed that fungi inhabiting difficult ecological niches are highly creative in their chemical machinery. This is to say that ecological and taxonomic observations on fungi will have much more dictation in impacting the chemical discovery process. Ecological and taxonomic phenomena, when translated into technological practice, new metabolic leads generally get expressed.

Group I: (Moniliaceous phialidic) 30 25 No. of isolates 20 15 10 5 C. congesta ■ F.benghalensis 0 **S1 S2 S**3 **S4 S**5 Season Group II: (Dematiaceous phialidic) 200 No. of isolates 100 100 50 C. congesta F.benahalensi 0 S1 S2 S3 **S4** S5 **S6** Season Group III: (Moniliaceous blastic) 200 No. of isolates 150

C. congesta F.benghalensis

100

50

S2

S3

S4

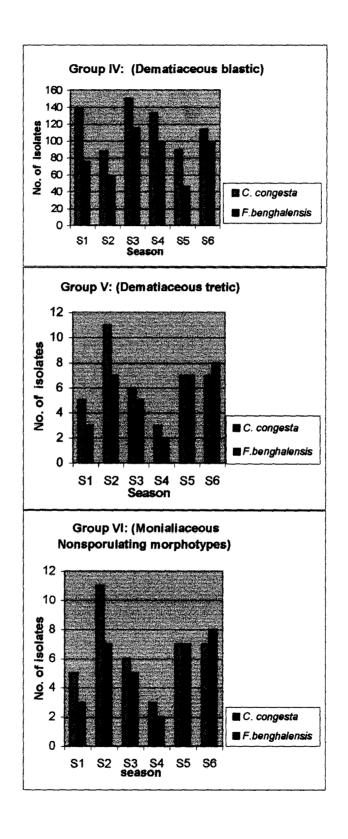
Season

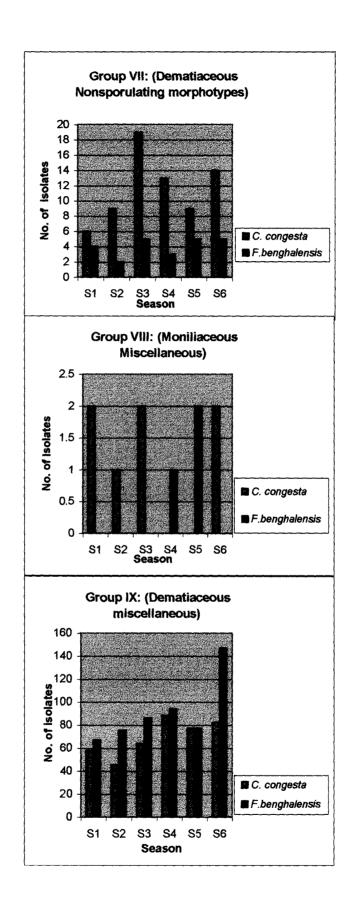
S5

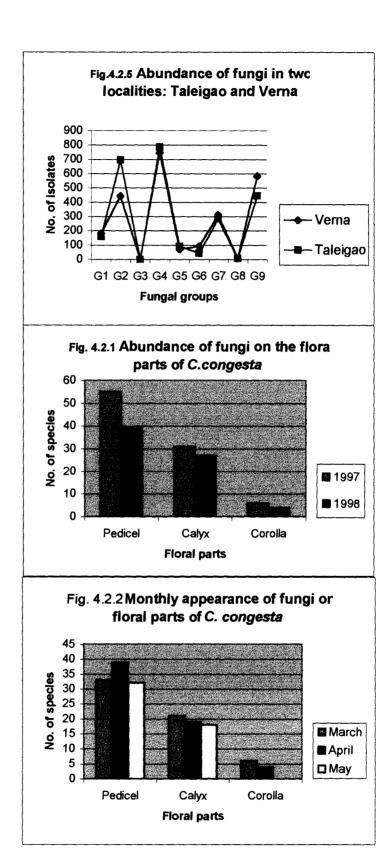
S6

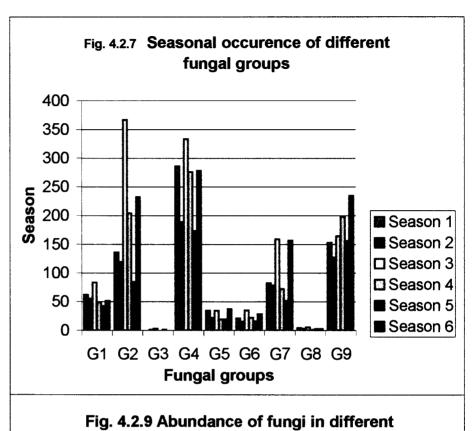
S1

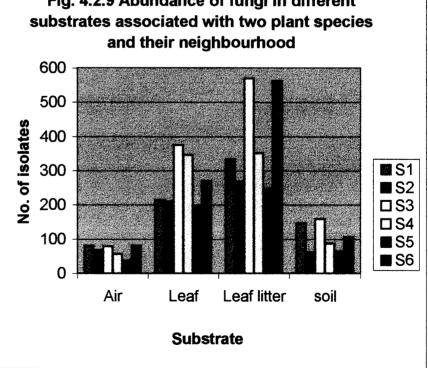
Fig. 4.2.6 Seasonal Occurrence of fungi











LIGNOLYTIC ACTIVITY

SR. NO.	TAXONOMY	ACTIVITY	REMARKS
1.	NS. 33		
2.	Weisneiriomyces	+	general decolorisation along edge., 3/4
 	javanicus	<u> </u>	of colony dark brown
3.	NS. 39	+	thin halo around the colony
4.	Scolecobasidium constrictum	-	_
5.	Ascochyta caricae	+	general decolorisation along edge of colony, darker at centre
6.	NS. 37	_	-
7.	Periconia byssoides	++	general decolorisation except for concentric bands which are darker brown
8.	Coniochaeta fuckelii	_	-
9.	Scolecobasidium humicola	_	_
10.	Ascochytula sapindae	++	general decolorisation, central patches darker brown
11.	Neocercosporella indica	_	-
12.	Fusarium tabacinum	+ .	general decolorisation, central colony light brown
13.	Arxiella terrestris	_	_
14.	NS. 42	_	_
15.	NS. 45	+	thin halo around colony
16.	Scolecobasidium variable	_	_
17.	NS. 116	++	general decolorisation, colony reddish brown in centre
18.	NS. 34	++	general decolorisation along edge of colony, central dark brown
19.	Aspergillus sp.7		_
20.	Gliomastix murorum	+	thin halo around colony
21.	Cladosporium sp.5	_	_
22.	Chaetomium nigricolor		
23.	Trichocladium album	+	general decolorisation along edge of colony, central medium brown
24.	Ajrekavella polychaetriae	++	General decolorisation prominent along the edges and interspaces of concentric circles of colony; central region of colony pinkish, after which it is dark brown
25.	Fusarium solani	_	_
26.	Fusarium decemcellulare	+	pale decolorisation along edge, centrally dark pinkish
27.	Camarosporium indicum	++	general decolorisation prominent along edge, central dark brown

28.	Trichoderma sp.1		
29.	NS. 30		
30.	Nigrospora endophytica		
31.	NS. 46		<u> – </u>
32.	Botryodiploidea	++	general decolorisation along edge,
	theobromae		central dark brown
33.	Sporidesmium altum	++	general decolorisation along edge,
			central dark brown
34.	Mucor flavus		
35.	NS. 35		
36.	Beltrani rhombica	+	very light halo around edge of colony
37.	Fusarium phragmitis	++	general decolorisation
38.	Cladosporium sp.2	++	general decolorisation at edge of
			colony, dark brown at centre
39.	Undetermined	+	general decolorisation along only edge
	hyphomycete sp. 12		³ / ₄ of colony dark brown
40.	Nigrospora sphaerica	++	general decolorisation along periphery
			and interspaces; concentric circles dark
	1 1		brown
41.	NS. 41	+	pale halo around colony
42.	Pseudobotrytis terrestris		_
43.	NS. 119	+	thin halo around colony
44.	Chaetomella cycadina		_
45.	NS. 47	++	general decolorisation, almost ½ plate
	110.17		decolorised
46.	NS. 31		
47.	Beltraniella bulolensis	+	general decolorisation along edge of
• • •			colony
48.	Gonytrichum sp.		_
49.	NS. 38		_
50.	NS. 118		
51.	Aspergillus sp.6		
52.	NS. 40		
53.	NS. 121	+	thin halo around colony
54 .	Cylindrocarpon	++	general decolorisation, central region
J 4 .	ianthothele	1 1	colony light brown
55.	NS. 33		Colony light blown
<u>56.</u>	NS. 49		thin halo around colony
		т	tilli lialo aroulid cololly
<u>57.</u>	NS. 36		4.5.1.1
58.	NS. 48	<u> </u>	thin halo around colony
59.	NS. 120		
60.	NS. 117		
€. • .	11 2 2 1 3 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1	-	{

CELLULOLYTIC ACTIVITY

SR.	TAXONOMY	COLONY	CLEARANCE	REMARKS
NO.		DIAM.	ZONE DIAM.	
1.	Fusarium solani	5.2 cm	-	colony shows irregular patches of yellow
2.	Nigrospora endophytica	6.0 cm		colony shows irregular patches of yellow
3.	NS. 30	2.1 cm	1.9 cm	3/4 of colony decolorised from the centre, just a little before the periphery
4.	NS. 120	1.3 cm	2.7 cm	1.3 cm diam. of colony highlighted yellow, also clearance zone
5.	Aspergillus sp.6	2.7 cm	0.2 cm	Clearance zone very faint, 2.0 cm diam. of colony decolorised from centre after which it is red
6.	Trichocladium album	5.0 cm	-	4.7 cm diam. from inner decolorised yellow after which red.
7.	Trichoderma sp.1	2.3 cm	_	2.0 cm diam. decolorised from centre after which red.
8.	Beltrani rhombica	0.8 cm	_	_
9.	Periconia byssoides	1.5 cm		-
10.	NS. 36	0.8 cm	-	_
11.	NS. 116	6.5 cm	-	entire colony decolorised yellow
12.	Chaetomella cycadina	6.0 cm	-	colony decolorised yellow in patches within a diam. of 4.5 cm diam. from centre
13.	NS. 49	1.8 cm	_	1.2 cm diam. of colony from centre decolorised
14.	NS. 34	3.4 cm	-	2.8 cm diam decolorised yellow from centre
15.	Sporidesmium altum	4.6 cm	_	3.4 cm diam. yellow
16.	NS. 119	1.0 cm	_	edge of colony decolorised yellow, 0.1 cm radius in thickness
17.	Gliomastix murorum	0.9 cm	_	_
18.	Ascochytula sapindae	5.6 cm	_	colony decolorised yellow in patches
19.	Neocercosporella indica	0.8 cm	_	_
20.	NS. 39	0.7 cm	_	_
21.	Scolecobasidium constrictum	0.6 cm	_	-
22.	NS. 48	0.6 cm	_	-
23.	NS. 35	1.3 cm	_	edge of colony about 0.3 cm radius decolorised yellow

24.	Fusarium	2.3 cm	_	1.9 cm decolorised yellow from
	decemcellulare			centre
25.	Aspergillus sp.7	2.0 cm	_	1.5 cm diam. decolorised from centre, though centre is darker brown
26.	Weisneiriomyces javanicus	1.1 cm	-	Colony decolorised in patches
27.	Fusarium tabacinum	5.2 cm		colony decolorised in patches
28.	Mucor flavus	6.8 cm	-	colony decolorised in very very few and tiny patches
29.	Gonytrichum sp.	1 cm		_
30.	Pseudobotrytis terrestris	1.3 cm	_	_
31.	NS. 45	0.5 cm	_	
32.	Cladosporium sp.2	6.5 cm	_	_
33.	NS. 38	0.6 cm	_	
34.	NS. 42	1.4 cm	-	0.6cm diam. of colony decolorised yellow from centre
35.	Botryodiploidea theobromae	1.2 cm	0.5 cm	halo very faint, centre green- brown, 0.2 cm in middle of colony or at the edge of 1 cm diam. from centre decolorised yellow
36.	NS. 41	1.4 cm	0.12 cm	
37.	NS. 37	3.0 cm	0.5 cm	colony color red., pale yellow clearance zone around colony
38.	NS. 47	5.0 cm	0.1 cm	patches of yellow a little before periphery of colony
39.	Camarosporium indicum	5.5 cm	_	Patches of yellow all over the colony
40.	NS. 117	4.5 cm	_	4.0 cm diam. from centre yellow
41.	Ajrekavella polychaetriae	8.0 cm	_	entire colony yellow
42.	Beltraniella bulolensis	2.7 cm	_	only edge of colony yellow
43.	Cylindrocarpon ianthothele	7.5 cm		_
44.	Ascochyta caricae	1.5 cm	_	only 0.1 cm of edge yellow
45.	NS. 33	2.6 cm		2.5 cm diam. from centre yellow
46.	Cladosporium sp.5	2.6 cm	0.4 cm	4cm diam. decolorised halo around colony
47.	Scolecobasidium variable	1.0 cm	——————————————————————————————————————	_
48.	NS. 118	2.5 cm		0.1 cm of edge yellow
49.	Nigrospora sphaerica	6.0 cm	_	_
50.	NS. 32	7.2 cm	_	6.0 cm diam. from centre of colony yellow
	NS. 46	2.7 cm	1.4 cm	1.4 cm diam. halo around colony

52.	Scolecobasidium humicola	2.0 cm	_	_
53.	NS. 121	3.0 cm		_
54.	Arxiella terrestris	3.9 cm	_	_
55.	NS. 40	2.5 cm		
56.	Coniochaeta fuckelii	3.8 cm	_	
57.	Undetermined hyphomycete sp. 12	3.2cm	_	_
58.	NS. 31	6.1 cm	_	_
59.	Fusarium phragmitis	8.5 cm		_
60.	Chaetomium nigricolor	6.5 cm	_	_

Where, Clearance zone 0.1 - 0.6cm +

0.7 - 1.2cm ++

1.3 - 1.8cm +++

1.9 - 2.4cm ++++

2.5 - 3.0cm +++++

Table: 4.3.3

XYLANASE ACTIVITY

SR.	TAXONOMY	COL	CZ	REMARKS
NO.		DIAM	DIAM	
1.	NS. 45	1.3 cm	_	_
2.	Ascochytula sapindae	1.0 cm		colony whitish
3.	Weisneiriomyces	0.8 cm	-	_
	javanicus			
4.	NS. 48	1.0 cm		
5	Sporidesmium altum	2.5 cm		light green pale yellow
6.	Cylindrocarpon ianthothele	3.9 cm	_	pinkish yellow
7.	Botryodiploidea theobromae	2.7 cm	_	whitish green
8.	Fusarium phragmitis	3.7 cm	<u></u>	pale yellow pink
9.	NS. 41	0.9 cm		Greenish
10.	NS. 39	0.5 cm		Greenish
11.	Fusarium decemcellulare	5.5 cm	1.2 cm	-
12.	Nigrospora sphaerica	1.1 cm	_	_
13.	NS. 47	2.5 cm	_	Off-white
14.	Trichocladium album	3.0 cm	_	Pale yellow green
15.	Ascochyta caricae	3.0 cm		yellow brown
16.	Camarosporium indicum	2.2 cm	_	pale orange
17.	NS. 121	3.9 cm	0.3 cm	reverse of colony pale orange
18.	NS. 32	2.3 cm	0.2 cm	colony partly orangish brown, partly olive green
19.	Cladosporium sp.2	4.8 cm	1.4 cm	olive green
20.	NS. 34	2.1 cm	0.1 cm	olive green
21.	Beltrani rhombica	0.8 cm	_	Greenish
22.	Gliomastix murorum	0.9 cm		Greenish
23.	Beltraniella bulolensis	4.2 cm	0.2 cm	pale green with whitish
24.	Undetermined hyphomycete sp. 12	2.0 cm	1.0 cm	(yellow pigment)
25.	Periconia byssoides	1.7 cm	_	pinkish light green
26.	NS. 119	1.2 cm	0.4 cm	
27.	Ajrekavella	2.0 cm	0.5 cm	Whitish
	polychaetriae			
28.	NS. 49	1.4 cm	_	_
29.	NS. 116	8.0 cm		_

Where, Clearance zone 0.1 - 0.6cm + 0.7 - 1.2cm ++

1.3 - 1.8cm +++

LACCASE ACTIVITY

Table: 4.3.4

SR.	TAXONOMY	COLONY	HALO	REMARKS
NO.		DIAM.	DIAM.	
1.	Gliomastix murorum	1.2 cm	0.6 cm	Violet
2.	NS. 48	1.6 cm	1.0 cm	Violet
3.	NS. 119	2.1 cm	0.8 cm	pinkish purple
4.	NS. 49	1.2 cm	0.6 cm	Violet
5.	Beltraniella	3.0 cm	1.4 cm	Shows inner green halo (1 cm) then l.
	bulolensis			pink (0.4 cm)
6.	NS. 41	0.9 cm	_	_
7.	NS. 47	2.5 cm	-	_
8.	NS. 34	2.0 cm	0.8 cm	bright green
9.	Ajrekavella	1.7 cm	0.2 cm	inner green halo after which a very
	polychaetriae			faint pink halo
10.	Cladosporium sp.2	2.3 cm	0.4 cm	bright green
11.	Beltrani rhombica	2.0 cm	_	_
12.	Fusarium phragmitis	2.5 cm		
13.	Undetermined	1.7 cm	_	-
	hyphomycete sp. 12			
14.	Trichocladium album	2.0 cm	0.3 cm	bright green, irregularly developed
				around colony in patches
15.	Ascochyta caricae	2.6 cm	0.6 cm	Bright green
16.	NS. 45	0.8 cm	1.4 cm	inner violet (1 cm) & outer light pink
				(0.4 cm)
17.	Ascochytula sapindae	4.5 cm	–	_
18.	NS. 39	0.7 cm	1.6 cm	inner violet (1.0 cm) outer l. pink (0.6
				cm)
19.	Periconia byssoides	1.7 cm	_	_
20.	NS. 32	2.3 cm	0.6 cm	bright green
21.	Weisneiriomyces	0.9 cm	0.4 cm	Violet
	javanicus			
22.	NS. 116	8.2 cm		_
23.	Sporidesmium altum	2.4 cm	1.2 cm	bright green
24.	Cylindrocarpon	3.6 cm	_	_
	ianthothele			
25.	Nigrospora sphaerica	4.1 cm	1.0 cm	bright green, regularly not around col.
				but along or within periphery
26.	Botryodiploidea	3.1 cm	0.4 cm	bright green within periphery
	theobromae			
27.	Fusarium	1.8 cm	_	-
	decemcellulare			
28.	NS. 121	0.7 cm	0.4 cm	green around colony
29.	Camarosporium	1.5 cm	-	_
117h -	indicum		<u> </u>	

Where, Clearance zone 0.1 - 0.6cm +

^{0.7 - 1.2}cm ++

^{1.3 - 1.8}cm +++

Table: 4.3.5 **PECTOLYTIC ACTIVITY**

SR.	TAXONOMY	COLONY	CLEARANCE
NO.	IAAONOWII	DIAM	ZONE DIAM.
1.	Scolecobasidium	1.5 cm	LONE DIAM.
1.	constrictum	1.5 cm	. -
2.	Cladosporium sp.5	2.3 cm	0.8 cm
3.	Arxiella terrestris	2.0 cm	0.4 cm
4.	NS. 46	1.6 cm	
5.	NS. 31	3.5 cm	0.2 cm
6.	NS. 117	1.6 cm	
7.	NS. 36	2.8 cm	0.4 cm
8.	Gonytrichum sp.	2.3 cm	· —
9.	NS. 49	2.2 cm	2.0 cm
10.	NS. 41	1.3 cm	1.0 cm
11.	Camarosporium indicum	4.1 cm	0.2 cm
12.	Beltrani rhombica	1.7 cm	2.2 cm
13.	Trichocladium album	5.9 cm	0.4 cm
14.	Aspergillus sp.6	4.4 cm	0.2 cm
15.	NS. 118	2.6 cm	0.8 cm
16.	Chaetomium	7.2 cm	_
	nigricolor		
17.	Chaetomella cycadina	8.4 cm	0.4 cm
18.	Coniochaeta fuckelii		
19.	NS. 30	2.2 cm	0.4 cm
20.	NS. 40	2.7 cm	0.4 cm
21.	Beltraniella	3.5 cm	_
	bulolensis		
22.	NS. 35	2.3 cm	0.3 cm
23.	Pseudobotrytis	1.7 cm	_
	terrestris		
24.	Neocercosporella indica	1.1 cm	-
25.	Periconia byssoides	6.5 cm	1.0 cm
26.	Fusarium phragmitis	8 cm	
27.	NS. 34	5.0 cm	_
28.	Ajrekavella	4.1 cm	0.3 cm
- -	polychaetriae		
29.	Ascochytula sapindae	8.9 cm	
30.	Nigrospora endophytica	8.8 cm	_
31.	Sporidesmium altum	3.4 cm	0.8 cm
32.	Gliomastix murorum	1.7 cm	
33.	Scolecobasidium variable	1.5 cm	· <u>-</u>
34.	NS. 33	2.7 cm	1.0 cm
J.⊤.	1 110.00	4., OIII	1.0 0111

35.	Cladosnovium on 2	2 8 am	
36.	Cladosporium sp.2	2.8 cm	0.2 cm
30.	Botryodiploidea	8.0 cm	0.2 cm
27	theobromae		
37.	Nigrospora sphaerica	5.1 cm	
38.	Fusarium solani	7.6 cm	0.2 cm
39.	NS. 120	2.0 cm	
40.	Undetermined	0.9 cm	-
<u> </u>	hyphomycete sp. 12		
41.	NS. 116	8.9 cm	<u> </u>
42.	Cylindrocarpon	8.7 cm	_
	ianthothele		<u> </u>
43.	Fusarium	3.3 cm	-
	decemcellulare		
44.	NS. 45	0.7 cm	_
45.	NS. 39	0.9 cm	_
46.	Weisneiriomyces	2.5 cm	0.4 cm
	javanicus		
47.	Ascochyta caricae	8.0 cm	0.2 cm
48.	NS. 119	1.3 cm	0.6 cm
49.	NS. 37	2.3 cm	1.0 cm
50.	Fusarium tabacinum	6.5 cm	0.2 cm
51.	Mucor flavus	8.8 cm	_
52.	NS. 38	0.8 cm	_
53.	NS. 32	7.2 cm	_
54.	Trichoderma sp.1	2.8 cm	_
55.	NS. 47	5.0 cm	0.2 cm
56.	Aspergillus sp.7	3.5 cm	0.4 cm
57.	NS. 48	1.2 cm	2.0 cm
58.	NS. 42	2.7 cm	1.4 cm
59.	Scolecobasidium	3.2 cm	_
	humicola		
60.	NS. 121	0.4 cm	

Where, Clearance zone 0.1 - 0.6cm +

0.7 - 1.2cm ++

1.3 - 1.8cm +++

1.9 - 2.4cm ++++

AMYLOLYTIC ACTIVITY

SR.	TAXONOMY	COLONY	HALO	REMARKS
NO.		DIAM.	DIAM.	
1.	NS. 33	3.1 cm	0.2 cm	colony whitish, only peripheral edge of colony pale yellow
2.	Weisneiriomyces javanicus	2.7 cm	0.4 cm	pale yellow in patches
3.	NS. 39	1.8 cm		
4.	Scolecobasidium constrictum	1.5 cm		-
5.	Ascochyta caricae	8.8 cm	-	entire colony yellow
6.	NS. 37	2.3 cm	-	entire colony whitish
7.	Coniochaeta fuckelii	4.0 cm	0.1 cm	Colony yellowish
8.	Periconia byssoides	2.0 cm	0.6 cm	entire colony pale yellow
9.	Scolecobasidium humicola	2.3 cm		-
10.	Ascochytula sapindae	8.3 cm		entire colony whitish
11.	Neocercosporella indica	1.3 cm	-	-
12.	NS. 32	8.2 cm	_	_
13.	Arxiella terrestris	4.1 cm	-	entire colony whitish
14.	NS. 45	1.2 cm		only peripheral edge of colony pale yellow
15.	Scolecobasidium variable	1.6 cm	0.2 cm	_
16.	NS. 116	8.7 cm	-	entire colony whitish
17.	NS. 34	6.6 cm	-	
18.	Aspergillus sp.7	2.3 cm	1.8 cm	entire colony pale yellow
19.	Gliomastix murorum	1.7 cm	2.2 cm	_
20.	Cladosporium sp.5	3.2 cm	0.4 cm	entire colony yellow
21.	Chaetomium nigricolor	6.7 cm		central region (4 cm diam.) of colony pale yellow
22.	Trichocladium album	7.5 cm		entire colony pale yellow
23.	Ajrekavella polychaetriae	6.0 cm	-ranna	entire colony whitish
24.	Fusarium solani	8.8 cm	-	entire colony whitish
25.	Fusarium decemcellulare	6.4 cm		edge of col. whitish
26.	Camarosporium indicum	4.4 cm	0.2 cm	entire colony yellow
27.	Trichoderma sp.1	8.7 cm		central region (3.5 cm diam.) has a bluish tinge, rest of the white.
28.	NS. 30	4.4 cm	0.2 cm	entire colony yellow
29.	Nigrospora endophytica	3.5 cm	-	entire colony pale yellow
30.	NS. 46	2.8 cm	0.1 cm	entire colony yellow
			~~~~~	

31.	Botryodiploidea theobromae	1.8 cm	1.8 cm	_
32.	Sporidesmium altum	8.9 cm	-	edge of colony pale yellow
33.	Mucor flavus	8.8 cm		entire colony whitish
34.	NS. 35	4.0 cm		entire colony yellow
35.	Fusarium phragmitis	8.7 cm		entire colony whitish
36.	Cladosporium sp.2	4.5 cm	-	
37.	Undetermined	3.5 cm	0.5 cm	_
	hyphomycete sp. 12			
38.	Nigrospora sphaerica	2.1 cm	0.6 cm	entire colony yellow
39.	NS. 41	2.0 cm	0.2 cm	entire colony pale yellow
40.	Pseudobotrytis	3.0 cm		<del>-</del>
	terrestris			
41.	NS. 119	1.6 cm	0.6 cm	_
42.	NS. 47	6.5 cm	-	entire colony pale yellow
43.	NS. 31	6.0 cm	-	central region (4.1 cm diam.) of
				colony yellowish
44.	Gonytrichum sp.	3.3 cm		entire colony yellowish
45.	NS. 38	2.0 cm	-	
46.	NS. 118	3.4 cm		entire colony yellow
47.	Aspergillus sp.6	2.9 cm		entire colony whitish
48.	NS. 40	3.0 cm		pale yellow at periphery of colony
49.	NS. 121	3.1 cm	_	peripheral region of colony pale
				yellow
50.	Cylindrocarpon	8.8 cm	-	entire whitish
	ianthothele			
51.	NS. 49	3.0 cm	0.6 cm	entire whitish
52.	NS. 36	3.3 cm	0.8 cm	
53.	NS. 48	1.4 cm		only edge of colony yellow
54.	NS. 120	2.4 cm	1.2 cm	-
55.	NS. 117	3.2 cm	0.6 cm	_
56.	Fusarium tabacinum	8.9 cm	_	entire whitish
57.	NS. 42	2.3 cm	_	
58.	Beltrani rhombica	3.0 cm	-	
59.	Chaetomella cycadina	2.9 cm	_	
60.	Beltraniella bulolensis	3.2 cm	-	~

Where, Clearance zone 0.1 - 0.6cm +

0.7 - 1.2cm ++

1.3 - 1.8cm +++

1.9 - 2.4cm ++++

### Table: 4.3.7

## PROTEOLYTIC ACTIVITY

SR.	TAXONOMY	COLONY	CLEARANCE	REMARKS
NO.		DIAM.	ZONE DIAM.	
1.	Mucor flavus	8.0 cm		
2.	Fusarium tabacinum	6.5 cm	_	
3.	Chaetomium	7.8 cm		
	nigricolor	710 011		
4.	Ascochytula sapindae	7.8 cm	-	
5.	Trichoderma sp.1	8.0 cm		
6.	Fusarium solani	7.3 cm	_	_
7.	Trichocladium album	5.0 cm	_	_
8.	NS. 116	5.9 cm	0.2 cm	
9.	Fusarium phragmitis	6.9 cm	0.2 cm	
10.	Nigrospora	6.5 cm	-	_
	endophytica			
11.	Scolecobasidium	1.3 cm		-
l	constrictum			
12.	Scolecobasidium	1.2 cm	-	-
	variable			
13.	Gliomastix murorum	2.0 cm	_	_
14.	NS. 30	3.4 cm	1.2 cm	
15.	Undetermined	0.5 cm	-	<b>-</b> .
	hyphomycete sp. 12			
16.	NS. 38	2.5 cm	-	_
17.	Cladosporium sp.2	1.4 cm	_	_
18.	NS. 35	2.9 cm	0.2 cm	
19.	NS. 47	5.7 cm		entire colony cleared
20.	Periconia byssoides	5.0 cm	_	_
21.	Weisneiriomyces javanicus	2.9 cm	-	<u>-</u>
22.	Fusarium	5.2 cm		_
	decemcellulare			**
23.	NS. 32	7.0 cm	_	-
24.	NS. 117	1.4 cm	-	
25.	Pseudobotrytis	1.2 cm	0.30 cm	
	terrestris			
26.	Nigrospora sphaerica	2.0 cm	_	_
27.	Cladosporium sp.5	1.5 cm	-	_
28.	Beltraniella bulolensis	6.0 cm	2.0 cm	
29.	Chaetomella cycadina	5.4 cm	1.6 cm	strong
30.	Ascochyta caricae	5.0 cm	1.0 cm	
31.	NS. 36	3.3 cm	1.8 cm	
32.	Coniochaeta fuckelii	2.5 cm	1.8 cm	
33.	NS. 118	3.0 cm	0.2 cm	
	A			

34.	Cylindrocarpon ianthothele	5.7 cm	2.2 cm	
35.	Botryodiploidea theobromae	6.0 cm	1.4 cm	
36.	NS. 45	0.9 cm	3.0 cm	
37.	Ajrekavella polychaetriae	3.9 cm	1.6 cm	
38.	NS. 34	6.0 cm	0.4 cm	
39.	Neocercosporella indica	1.1 cm	0.8 cm	
40.	NS. 120	2.0 cm	1.0 cm	
41.	NS. 39	1.1 cm	2.0 cm	
42.	Scolecobasidium humicola	3.0 cm	0.2 cm	
43.	Gonytrichum sp.	2.5 cm	0.6 cm	
44.	NS. 49	2.2 cm	0.6 cm	
45.	Aspergillus sp.7	2.9 cm	0.6 cm	
46.	Aspergillus sp.6	4.2 cm	1.2 cm	
47.	Sporidesmium altum	5.9 cm	0.4 cm	
48.	Camarosporium indicum	4.9 cm	0.10 cm	
49.	NS. 119	1.5 cm	0.4 cm	
50.	NS. 41	1.5 cm	0.8 cm	
51.	NS. 37	2.3 cm	0.2 cm	
52.	NS. 46	2.8 cm	0.4 cm	
53.	NS. 48	6.7 cm	1.0 cm	
54.	NS. 40	3.6 cm	0.2 cm	
55.	Arxiella terrestris	3.0 cm	1.6 cm	
56.	NS. 121	0.3 cm	2.0 cm	
57.	NS. 31	4.7 cm	1.0 cm	
58.	Beltrani rhombica	4.0 cm		entire colony decolorized in layers
59.	NS. 33	2.9 cm	0.2 cm	
60.	NS. 42	2.1 cm		_

Where, Clearance zone 0.1 - 0.6cm +

0.7 - 1.2cm ++

1.3 - 1.8cm +++

1.9 - 2.4cm ++++

Table: 4.3.8 Positive isolates associated with F.benghalensis and C. congesta

Enzyme	Total isolates screened	Total isolates showing activity	% positive isolates	C.congesta associated positive isolates	F.benghalensis associated positive isolates	
Pectinase	60	31	51.6	67.7	45.1	
Amylase	60	20	33.3	75.0	45.0	
Protease	60	38	63.3	71.1	42.1	
Cellulase	60	9	15.0	88.8	22.2	
Ligninase	60	29	48.3	72.4	48.2	
Laccase	29	18	62.0	61.1	50.0	
Xylanase	29	9	31.0	66.6	44.4	

Table: 4.3.9 Days on which maximum laccase activity was observed.

Taxonomy	Laccase activity in nkats	Day on which optimum laccase activity is observed	Plant from which the fungal strain is obtained.
Fusarium phragmitis	19.7	- 5	Carissa & Ficus
NS. 47	183.6	5	Carissa
Ajrekavella polychaetriae	47.0	. 15	Carissa
Cylindrocarpon ianthothele	23.4	5	Carissa & Ficus
Camarosporium indicum	110.1	3	Carissa & Ficus
Beltraniella bulolensis	13.5	5	Carissa
Gliomastix murorum	26.2	3	Carissa & Ficus
Cladosporium sp.2	19.09	11	Carissa
Undetermined hyphomycete sp. 12	48.0	13	Carissa
NS. 34	16.2	5	Carissa
Ascochytula sapindae	19.3	5	Carissa
Sporidesmium altum	22.2	3	Ficus
NS. 116	4.8	3	Carissa
Botryodiploidea theobromae	140.5	11	Ficus
Fusarium decemcellulare	14.5	3	Carissa & Ficus
NS. 45	26.3	11	Carissa
NS. 48	1166.7	7	Carissa
Periconia byssoides	78.6	5,9	Ficus

Table: 4.3.10 Plants showing strains with laccase activity.

Laccase activity	Carissa congesta	Ficus benghalensis	Carissa & Ficus
>01	Beltraniella bulolensis, Cladosporium sp. 2, NS. 34, Ascochytula sapindae, NS. 116,		Fusarium phragmitis, Fusarium decemcellulare,
>20	Ajrekavella polychaetriae, Undetermined hyphomycete sp.12 NS. 45,	Sporidesmium altum	Cylindrocarpon ianthothele, Gliomastix murorum,
>50		Periconia byssoides	<del></del>
>80			Camarosporium indicum
>110		Botryodiploidea theobromae	
>140			
>170	NS. 47		
>1160	NS. 48		

## Where,

>01 = 0-20 nkats >20 =21-50 nkats >50 =51-80 nkats >80 =81-110 nkats >110 =111-140 nkats >140 =141-170 nkats >170 =171-200 nkats >1160=1161-1190nkats

Table: 4.3.11 Laccase activity in the two plant species.

Experimental plants	Laccase activity	No. of isolates	% of isolates	
Carissa congesta	Low	14	77.77	
	Moderate	0	0.00	
	High	1	5.55	
Ficus benghalensis	Low	8	44.44	
	Moderate	0	0.00	
	High	0	0.00	

#### Where,

0 - 380 nkats = Low 381-770 nkats = Moderate 771-1190 nkats = High

Table: 4.3.12 Qualitative estimation of enzymatic activity of fungal strains obtained from two plant species.

TAXONOMY	PLANT	AMYLASE	PECTINASE	PROTEASE	CELLULASE	LIGNINASE	LACCASE	XYLANASE
NS. 48	Carissa	_	++++	++		+	++	_
NS. 49	Carissa	+	++++	+		+	+	
Beltraniella bulolensis	Carissa	·		++++	_	+	+++	+
NS. 41	Carissa	+	++	++	+	+		
NS. 47	Carissa	_	+	++++	+	++		
NS. 34	Carissa	_	<del>-</del>	+	_	++	++	+
Ajrekavella	Carissa		+	+++	_	++	+	+
polychaetriae								
Cladosporium sp.2	Carissa		_		_	++	+	+++
Beltrani rhombica	Carissa	_	++++	+++	_	+		_
Undetermined	Carissa	+		_	_	+	_	++
hyphomycete sp.12								
Trichocladium album	Carissa	<u> </u>	+			+	+	_
NS. 45	Carissa	<del>-</del>		+++++	_	+	+++	-
Ascochytula sapindae	Carissa		_			++	_	_
NS. 116	Carissa		_	+	_	++		_
Fusarium solani	Carissa		+		_			
Trichoderma sp.1	Carissa		_		_			
NS. 30	Carissa	+	+	++	++++			
Nigrospora endophytica	Carissa	<del>-</del>	_	_		<u> </u>		
NS. 46	Carissa	+		+	+++			
Mucor flavus	Carissa	_	_					
Pseudobotrytis terrestris	Carissa	_	<del></del>	+		-		:
Chaetomella cycadina	Carissa	_	+	+++	_			
Gonytrichum sp.	Carissa	_	_	+	_			
Aspergillus sp.6	Carissa	_	+	++	+			
NS. 40	Carissa		+	+	_			
NS. 120	Carissa	++	_	++	+++++			
NS. 117	Carissa	+			-			

Scolecobasidium constrictum	Carissa	<del></del>		_	_	-		
NS. 37	Carissa	-	++	+	+			
Coniochaeta fuckelii	Carissa	+	+	+++				
Scolecobasidium	Carissa			+	-	_	1	
humicola								
NS. 42	Carissa		+++					
Scolecobasidium	Carissa	+		_	-	-	}	
variable						<u> </u>		
Aspergillus sp.7	Carissa	+++	+	+	<u></u>			
Ascochyta caricae	Ficus		+	++	-	+	+	
NS. 39	Ficus		~	++++		+	+++	
Periconia byssoides	Ficus	+	++			++		
NS. 32	Ficus				_	+	+	+
Sporidesmium altum	Ficus		++	+	-	++	++	
Botryodiploidea	Ficus	+++	+	+++	+	++	+	
theobromae							ļ	
NS. 121	Ficus			++++		+	+	+
NS. 35	Ficus	_	+	+	_		ļ	
NS. 31	Ficus		+	++				
NS. 38	Ficus							
NS. 36	Ficus	++	+	+++		-	<u> </u>	
Fusarium tabacinum	Ficus		+				ļ	
NS. 33	Ficus	+	++	+				
Neocercosporella indica	Ficus	_		++	_			
NS. 119	Ficus	+	+	+		+	++	+
Gliomastix murorum	Carissa	++++	-	-	-	+	+	
	& Ficus							
Fusarium phragmitis	Carissa	-	_	+		++	_	_
•	& Ficus							
Weisneiriomyces	Carissa	+	+	-	-	+	+	
javanicus	& Ficus		<u> </u>	<u> </u>		1,	<u> </u>	l

Cylindrocarpon ianthothele	Carissa & Ficus	_	_	++++	_	++	_	_
Nigrospora sphaerica	Carissa & Ficus	+	_		_	++	++	
Fusarium decemcellulare	Carissa & Ficus	_	_	_	<u>-</u>	+	-	++
Camarosporium indicum	Carissa & Ficus	+	+	+	_	++		_
NS. 118	Carissa & Ficus	_	++	+	_	_		
Arxiella terrestris	Carissa & Ficus	_	+	+++	_	_		
Cladosporium sp.5	Carissa & Ficus	+	++	_	+	_		
Chaetomium nigricolor	Carissa & Ficus	_	_		_	_		

Clearance zone, 0.1 - 0.6 cm = +

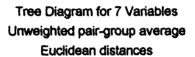
0.7 - 1.2 cm = ++

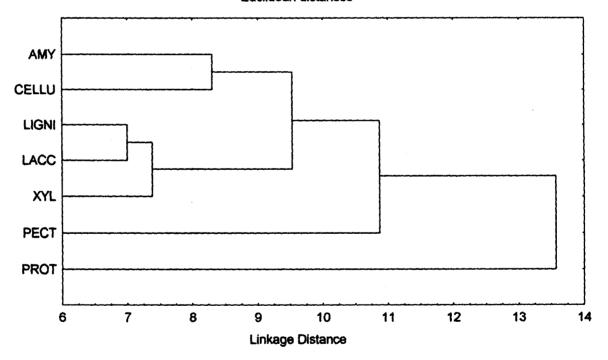
1.3 - 1.8 cm = +++

1.9 - 2.4 cm = ++++

2.5 - 3.0 cm = +++++

Fig. 4.3.1





Tree Diagram for 60 Cases
Unweighted pair-group average
Euclidean distances

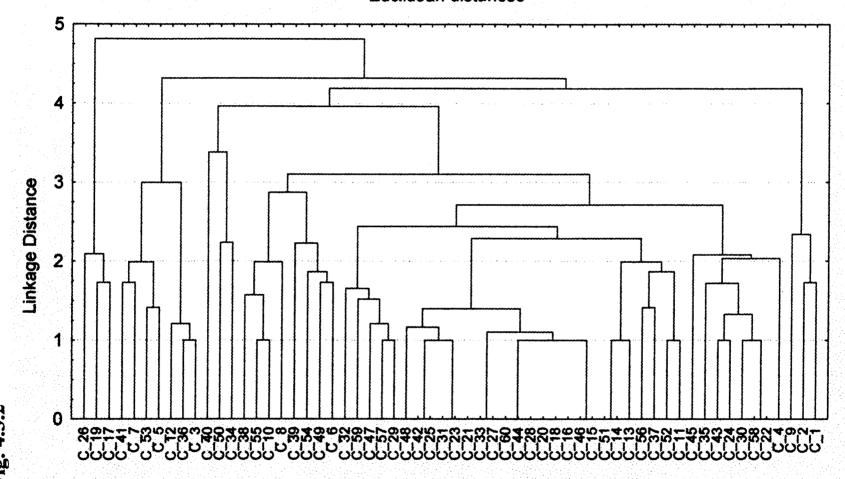


Table 4.2.1 3 factor factorial

			Group 1				······································	Group 2					Group 3		
	Sub1	Sub2	Sub3	Sub4	Avg	Subl	Sub2	Sub3	Sub4	Avg	Sub1	Sub2	Sub3	Sub4	Avg
	0.3731	0.0817	0.1434	0.6608	0.2161	0.088	0.2815	0.4134	0.3069	0.3008	-1E-05	0.1159	-1E-05	-1E-05	0.0401
S1	(0.9344)	(0.7627)	(0.8021)	(1.0774)	(0.8462)	(0.7668)	(0.8840)	(0.9557)	(0.8983)	(0.8949)	(0.7071)	(0.7848)	(0.7071)	(0.7071)	(0.7349)
	0.3705	0.0404	0.1708	0.3286	0.1698	0.1153	0.1378	0.4014	0.1271	0.2217	-1E-05	-1E-05	-1E-05	-1E-05	-1E-05
S2	(0.9330)	(0.7351)	(0.8190)	(0.9103)	(0.8184)	(0.7844)	(0.7986)	(0.9494)	(0.7919)	(0.8495)	(0.7071)	(0.7071)	(0.7554)	(0.7071)	(0.7244)
	0.2254	0.0414	0.1722	0.5349	0.1767	0.0767	1.0203	0.3532	0.5024	0.5441	-1E-05	-1E-05	0.0373	-1E-05	0.0132
S3	(0.8517)	(0.7358)	(0.8199)	(1.0173)	(0.8226)	(0.7594)	(1.2330)	(0.9237)	(1.0012)	(1.0218)	(0.7071)	(0.7071)	(0.7330)	(0.7071)	(0.7164)
	0.1222	0.0245	0.1362	0.4318	0.1306	0.1271	0.6097	0.3879	0.1516	0.3859	-1E-05	-1E-05	0.0706	-1E-05	0.0248
S4	(0.7888)	(0.7242)	(0.7976)	(0.9653)	(0.7941)	(0.7919)	(1.0534)	(0.9423)	(0.8072)	(0.9412)	(0.7071)	(0.7071)	(0.7554)	(0.7071)	(0.7244)
	0.0826	0.0332	0.18	0.5994	0.1628	0.0377	0.1382	0.3321	0.141	0.1892	-1E-05	-1E-05	0.0373	-1E-05	0.0132
S5	(0.7633)	(0.7302)	(0.8246)	(1.0485)	(0.8141)	(0.7333)	(0.7989)	(0.9122)	(0.8006)	(0.8302)	(0.7071)	(0.7071)	(0.7330)	(0.7071)	(0.7164)
	0.1374	0.0082	0.1582	0.538	0.1463	0.0744	0.6306	0.3765	0.3059	0.4036	-1E-05	-1E-05	0.0373	-1E-05	0.0132
S6	(0.7984)	(0.7129)	(0.8113)	(1.0188)	(0.8039)	(0.7579)	(1.0633)	(0.9362)	(0.8977)	(0.9506)	(0.7071)	(0.7071)	(0.7330)	(0.7071)	(0.7164)
	0.2139	0.038	0.16	0.5126	0.1668	0.0861	0.4446	0.3772	0.2501	0.3367	-1E-05	0.0185	0.042	-1E-05	0.0214
Avg	(0.8449)	(0.7335)	(0.8124)	(1.0063)	(0.8166)	(0.7656)	(0.9719)	(0.9366)	(0.8661)	(0.9147)	(0.7071)	(0.7201)	(0.7362)	(0.7071)	(0.7221)

			Group 4					Group 5			· · · · · · · · · · · · · · · · · · ·		Group 6		
	Sub1	Sub2	Sub3	Sub4	Avg	Sub1	Sub2	Sub3	Sub4	Avg	Sub1	Sub2	Sub3	Sub4	Avg
	0.1366	0.1775	0.255	0.2234	0.2053	0.444	0.0297	0.0562	0.1964	0.1151	-1E-05	0.0364	0.0477	0.09	0.0426
S1	(0.7979)	(0.8231)	(0.8689)	(0.8505)	(0.8398)	(0.9716)	(0.7278)	(0.7458)	(0.8345)	(0.7843)	(0.7071)	(0.7324)	(0.7401)	(0.7681)	(0.7366)
	0.0881	0.1876	0.1357	0.122	0.145	0.0489	0.0098	0.1488	0.05	0.0693	0.1235	0.0246	0.1225	-1E-05	0.069
S2	(0.7669)	(0.8292)	(0.7973)	(0.7887)	(0.8031)	(0.7409)	(0.7140)	(0.8055)	(0.7416)	(0.7545)	(0.7896)	(0.7243)	(0.7890)	(0.7071)	(0.7543)
	0.1025	0.0661	0.3692	0.2071	0.1934	0.2649	-1E-05	0.1061	0.27	0.1098	0.1467	0.0364	0.2321	0.2184	0.145
S3	(0.7762)	(0.7524)	(0.9323)	(0.8409)	(0.8327)	(0.8746)	(0.7071)	(0.7785)	(0.8775)	(0.7809)	(0.8042)	(0.7324)	(0.8556)	(0.8476)	(0.8031)
	0.0782	0.2053	0.2418	0.092	0.1824	0.444	0.0098	0.0385	0.1461	0.094	-1E-05	0.0864	0.1089	0.2809	0.1075
S4	(0.7604)	(0.8398)	(0.8613)	(0.7694)	(0.8261)	(0.9716)	(0.7140)	(0.7338)	(0.8038)	(0.7707)	(0.7071)	(0.7658)	(0.7803)	(0.8837)	(0.7794)
	0.0682	0.1701	0.1397	0.122	0.1374	0.1247	-1E-05	0.136	-1E-05	0.0645	0.0607	0.0351	0.133	-1E-05	0.0676
S5	(0.7538)	(0.8186)	(0.7998)	(0.7887)	(0.7984)	(0.7904)	(0.7071)	(0.7975)	(0.7071)	(0.7513)	(0.7488)	(0.7315)	(0.7956)	(0.7071)	(0.7534)
	0.1406	0.0446	0.366	0.1597	0.1828	0.3993	0.0098	0.1279	0.1747	0.1254	0.1529	0.0477	0.1844	0.2487	0.1381
S6	(0.8004)	(0.7380)	(0.9306)	(0.8122)	(0.8263)	(0.9483)	(0.7140)	(0.7924)	(0.8214)	(0.7908)	(0.8080)	(0.7401)	(0.8273)	(0.8653)	(0.7988)
	0.102	0.1403	0.2482	0.1535	0.1742	0.2795	0.0098	0.1016	0.1363	0.0961	0.0788	0.0442	0.1368	0.1344	0.0943
Avg	(0.7759)	(0.8002)	(0.865)	(0.8084)	(0.8211)	(0.8829)	(0.7140)	(0.7756)	(0.7977)	(0.7721)	(0.7608)	(0.7377)	(0.7980)	(0.7965)	(0.7709)

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			Group 7					Group 8					Group 9		
	Subl	Sub2	Sub3	Sub4	Avg	Sub1	Sub2	Sub3	Sub4	Avg	Subl	Sub2	Sub3	Sub4	Avg
	0.019	0.0389	0.0889	0.1134	0.0642	-1E-05	0.06	0.06	0.3334	0.0866	0.0243	0.4417	0.387	0.3516	0.3431
S1	(0.7204)	(0.7341)	(0.7674)	(0.7832)	(0.7511)	(0.7071)	(0.7483)	(0.7483)	(0.9129)	(0.7659)	(0.7241)	(0.9704)	(0.9418)	(0.9228)	(0.9182)
	0.0616	0.0265	0.0767	0.019	0.048	-1E-05	0.06	-1E-05	0.3334	0.0643	0.0604	0.3165	0.2642	0.0242	0.2145
S2	(0.7494)	(0.7256)	(0.7594)	(0.7204)	(0.7403)	(0.7071)	(0.7483)	(0.7071)	(0.9129)	(0.7512)	(0.7486)	(0.9036)	(0.8742)	(0.7240)	(0.8453)
	0.0851	0.0449	0.1456	0.0934	0.0927	-1E-05	-1E-05	0.06	0.433	0.0758	0.0485	0.4968	0.3662	0.0848	0.3172
S3	(0.7649)	(0.7382)	(0.8035)	(0.7703)	(0.7699)	(0.7071)	(0.7071)	(0.7483)	(0.9659)	(0.7588)	(0.7406)	(0.9984)	(0.9307)	(0.7647)	(0.9040)
	0.0095	0.0279	0.0887	0.0519	0.05	-1E-05	0.06	-1E-05	-1E-05	0.021	0.0121	0.5102	0.3325	0.1698	0.3172
S4	(0.7138)	(0.7266)	(0.7673)	(0.7429)	(0.7416)	(0.7071)	(0.7483)	(0.7071)	(0.7071)	(0.7218)	(0.7156)	(1.0051)	(0.9124)	(0.8184)	(0.9040)
	0.0237	0.0242	0.0675	0.0285	0.0399	-1E-05	-1E-05	0.1232	-1E-05	0.0424	0.0121	0.3449	0.377	0.0121	0.252
S5	(0.7237)	(0.7240)	(0.7533)	(0.7270)	(0.7348)	(0.7071)	(0.7071)	(0.7894)	(0.7071)	(0.7365)	(0.7156)	(0.9192)	(0.9365)	(0.7156)	(0.8672)
	0.0522	0.0567	0.1783	0.033	0.0946	-1E-05	-1E-05	-1E-05	0.2985	0.0385	0.0243	0.412	0.495	0.0242	0.3176
S6	(0.7431)	(0.7461)	(0.8236)	(0.7301)	(0.7711)	(0.7071)	(0.7071)	(0.7071)	(0.8936)	(0.7338)	(0.7241)	(0.9550)	(0.9975)	(0.7240)	(0.9042)
	0.0415	0.0364	0.107	0.0561	0.0648	-1E-05	0.0295	0.0395	0.2223	0.0546	0.0301	0.4189	0.369	0.1056	0.293
Avg	(0.7359)	(0.7324)	(0.7791)	(0.7457)	(0.7515)	(0.7071)	(0.7277)	(0.7345)	(0.8499)	(0.7447)	(0.7281)	(0.9586)	(0.9322)	(0.7782)	(0.8905)

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			Average		
	Sub1	Sub2	Sub3	Sub4	Avg
	0.1112	0.1343	0.1538	0.2424	0.1529
S1	(0.7818)	(0.7964)	(0.8086)	(0.8616)	(0.8080)
	0.0924	0.0854	0.1501	0.1056	0.112
S2	(0.7697)	(0.7651)	(0.8063)	(0.7782)	(0.7823)
	0.1025	0.16	0.1992	0.2496	0.178
S3	(0.7762)	(0.8124)	(0.8362)	(0.8658)	(0.8234)
	0.0816	0.1551	0.1503	0.1408	0.1406
S4	(0.7626)	(0.8094)	(0.8064)	(0.8005)	(0.8004)
	0.0448	0.0782	0.1655	0.0894	0.1053
S5	(0.7381)	(0.7604)	(0.8158)	(0.7677)	(0.7780)
	0.104	0.1195	0.2054	0.1889	0.1571
S6	(0.7772)	(0.7871)	(0.8399)	(0.8300)	(0.8106)
	0.0892	0.1217	0.1704	0.168	0.1408
Avg	(0.7676)	(0.7885)	(0.8188)	(0.8173)	(0.8005)

CD (0.05) for Groups 0.0203

CD (0.05) for Seasons 0.0166

CD (0.05) for Substrate 0.0135

CD (0.05) for Group x Season 0.0497

CD (0.05) for Group x Substrate 0.0406

CD (0.05) for Season x Substrate 0.0331

CD (0.05) for Group x Season x Substrate 0.0994

Table 4.2.2 Analysis

3² factorial CRD Experiment on (*C.congesta*)

	<del></del>					Group 6		Group 8	
	0.0976	1.6898	0.1023	0.4465			0.1345	0.05	1.2849
P1	(0.7731)	(1.4798)	(0.7761)	(0.9729)	(0.7657)	(0.7757)	(0.7966)	(0.7416)	(1.3360
	0.0195	0.5932	0.0499	0.2804	-1E-05	-1E-05	0.0780	-1E-05	0.7387
P2	(0.7208)	(1.0456)	(0.7416)	(0.8834)	(0.7071)	(0.7071)	(0.7603)	(0.7071)	(1.1130)
-	-1E-05	0.2503	-1E-05	0.0649		-1E-05	0.0103	-1E-05	0.0323
P3	(0.7071)	(0.8662)	(0.7071)	(0.7516)	(0.7071)	(0.7071)	(0.7144)	(0.7071)	(0.7296)
	0.0195	0.2846	-1E-05	0.2214	0.0279	0.0394	0.0614	-1E-05	0.5424
S1	(0.7208)	(0.8858)	(0.7071)	(0.8494)	(0.7266)	(0.7345)	(0.7493)	(0.7071)	(1.0210)
	0.0195	1.5377	-1E-05	0.1530	-1E-05	0.0394	0.0947	-1E-05	0.6672
S2	(0.7208)	(1.4275)	(0.7071)	(0.8081)	(0.7071)	(0.7345)	(0.7712)	(0.7071)	(1.0804)
	0.0766	0.6627	0.1570	0.4030	0.0568	0.0195	0.0634	0.05	0.6603
S3	(0.7594)	(1.0783)	(0.8106)	(0.9503)	(0.7462)	(0.7208)	(0.7506)	(0.7416)	(1.0772)
	-1E-05	0.4279	-1E-05	0.361	0.0863	0.123	0.0931	-1E-05	1.0304
P1S1	(0.7071)	(0.9633)	(0.7071)	(0.9279)	(0.7657)	(0.789)	(0.7701)	(0.7071)	(1.2371)
	0.06	4.2411	-1E-05	0.2857	-1E-05	0.123	0.1884	-1E-05	1.5721
P1S2	(0.7483)	(2.1774)	(0.7071)	(0.8864)	(0.7071)	(0.789)	(0.8297)	(0.7071)	(1.4395)
	0.2465	1.1869	0.3358	0.7195	0.1795	0.06	0.1239	0.1571	1.2726
P1S3	(0.8640)	(1.2988)	(0.9142)	(1.1043)	(0.8243)	(0.748)	(0.7899)	(0.8106)	(1.3314)
	0.0600	0.3219	0.0000	0.2592	0.0000	0.0000	0.0829	0.0000	0.6922
P2S1	(0.7483)	(0.9066)	(0.7071)	(0.8713)	(0.7071)	(0.707)	(0.7635)	(0.7071)	(1.0919)
	-1E-05	0.7274	-1E-05	0.1621	-1E-05	-1E-05	0.0923	-1E-05	0.6395
P2S2	(0.7071)	(1.1079)	(0.7071)	(0.8137)	(0.7071)	(0.707)	(0.7696)	(0.7071)	(1.0675)
	-1E-05	0.7596	0.1571	0.4316	-1E-05	-1E-05	0.0591	-1E-05	-0.4678
P2S3	(0.7071)	(1.1223)	(0.8106)	(0.9652)	(0.7071)	(0.707)	(0.7477)	(0.7071)	(0.1794)
	-1E-05	0.1203	-1E-05	0.0612	-1E-05	-1E-05	0.0104	-1E-05	0.0389
P3S1	(0.7071)	(0.7876)	(0.7071)	(0.7491)	(0.7071)	(0.707)	(0.7144)	(0.7071)	(0.7341)
	-1E-05								0.0389
P3S2	(0.7071)	(0.9972)	(0.7071)	(0.7243)	(0.7071)	(0.707)	(0.7144)	(0.7071)	(0.7341)
	-1E-05	<del></del>	<del></del>	<del> </del>	-1E-05	<del></del>		<del>  `</del>	
P3S3	(0.7071)					ì	j	ł	
Mean	(0.7337)	(1.1305)	<del></del>	<del>  `                                   </del>	<del> </del>	<del>                                     </del>	<del></del>		

Group 1	C.D. (0.05) for Part	0.05
•	C.D. (0.05) for Season	0.05
	C.D. (0.05) for Part x Season	0.09
Group 2	C.D. (0.05) for Part	0.10
	C.D. (0.05) for Season	0.10
•	C.D. (0.05) for Part x Season	0.17
Group 3	C.D. (0.05) for Part	0.09
	C.D. (0.05) for Season	0.09
	C.D. (0.05) for Part x Season	0.16
Group 4	C.D. (0.05) for Part	0.04
•	C.D. (0.05) for Season	0.04
	C.D. (0.05) for Part x Season	0.07
Group 5	C.D. (0.05) for Part	0.04
*	C.D. (0.05) for Season	0.04
	C.D. (0.05) for Part x Season	0.085
Group 6	C.D. (0.05) for Part	0.045
•	C.D. (0.05) for Season	0.045
	C.D. (0.05) for Part x Season	0.08
Group 7	C.D. (0.05) for Part	0.032
4	C.D. (0.05) for Season	0.032
	C.D. (0.05) for Part x Season	0.06
Group 8	C.D. (0.05) for Part	0.06
*	C.D. (0.05) for Season	0.06
	C.D. (0.05) for Part x Season	0.10
Group 9	C.D. (0.05) for Part	0.06
	C.D. (0.05) for Season	0.06
	C.D. (0.05) for Part x Season	0.10
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Table 4.2.3 **Analysis 3**² factorial CRD Experiment on (F.benghalensis)

	Group 1	Group 2	Group 4	Group 5	Group 6	Group 7	Group 8	Group 9
	0.1006	0.2396	0.3964	0.0560	0.2420	0.1518	0.1023	0.596
P1	(0.7750)	(0.8600)	(0.9468)	(0.7457)	(0.8614)	(0.8074)	(0.7761)	(1.0469)
	0.0611	0.3493	0.3056	0.0195	0.0976	0.0340	-1E-05	0.3167
P2	(0.7491)	(0.9216)	(0.8976)	(0.7208)	(0.7731)	(0.7308)	(0.7071)	(0.9037)
	0.0247	0.0208	0.0484	-1E-05	0.0599	0.0093	-1E-05	0.0185
P3	(0.7244)	(0.7217)	(0.7406)	(0.7071)	(0.7483)	(0.7137)	(0.7071)	(0.7201)
	0.0742	0.1572	0.3519	-1E-05	0.0599	0.0492	0.0499	0.2348
S1	(0.7578)	(0.8107)	(0.9230)	(0.7071)	(0.7483)	(0.7411)	(0.7416)	(0.8572)
	0.0122	0.0721	0.0760	0.0358	0.1771	0.0896	-1E-05	0.3118
S2	(0.7157)	(0.7564)	(0.7590)	(0.7320)	(0.8229)	(0.7679)	(0.7071)	(0.9010)
	0.1006	0.3764	0.3157	0.0394	0.1588	0.0517	0.0499	0.3327
S3	(0.7750)	(0.9362)			(0.81170			(0.9125)
	0.1572		1			i	0.1571	0.4411
P1S1	(0.8107)	(0.8525)	(1.0485)	(0.7071)	(0.7483)	(0.776)	(0.8106)	(0.9701)
	-1E-05	0.0931	0.1268	0.1111	0.46	0.235	-1E-05	0.6387
P1S2	(0.7071)	(0.7701)		(0.7817)	(0.9798)	(0.858)	(0.7071)	(1.0671)
	0.1516				1		1	0.7177
P1S3	(0.8072)		·	(0.7483)	<del></del>	<del>`</del>		
	0.0706	0.2613	Į.	1			1	
P2S1	(0.7554)	(0.8725)	(0.9774)	(0.7071)	(0.7483)	(0.733)	(0.7071)	(0.8846)
	0.0373	0.1273	0.0829	-1E-05	0.1111	0.037	-1E-05	0.327
P2S2	(0.7330)	(0.7920)	(0.7635)	(0.7071)	<del></del>	·	(0.7071)	<del>  \</del>
	0.0759		1					1
P2S3	(0.7589)	(1.1003)	(0.9520)	(0.7483)	(0.7894)	(0.727)	(0.7071)	(0.9172)
	-1E-05			i e				
P3S1	(0.7071)	(0.7071)	(0.7431)	(0.7071)	(0.7483)	(0.714)	(0.7071)	(0.7169)
	-1E-05	-1E-05		-1E-05	-1E-05	0.009	-1E-05	0.0279
P3S2	(0.7071)			(0.7071)			(0.7071)	
	0.0759			1	1		1	
P3S3	(0.7589)			(0.7071)	<del></del>	<del> </del>	(0.7071)	
Mean	(0.7495)	(0.8344)	(0.8617)	(0.7245)	(0.7943)	(0.751)	(0.7301)	(0.8902)

Group 1	C.D. (0.05) for Part C.D. (0.05) for Season C.D. (0.05) for Part x Season	0.08 0.08 0.16
Group 2	C.D. (0.05) for Part C.D. (0.05) for Season C.D. (0.05) for Part x Season	0.14 0.14 0.24
Group 3	Absent	
Group 4	C.D. (0.05) for Part C.D. (0.05) for Season C.D. (0.05) for Part x Season	0.12 0.12 0.2
Group 5	C.D. (0.05) for Part C.D. (0.05) for Season C.D. (0.05) for Part x Season	0.15 0.15 0.18
Group 6	C.D. (0.05) for Part C.D. (0.05) for Season C.D. (0.05) for Part x Season	0.18 0.18 0.30
Group 7	C.D. (0.05) for Part C.D. (0.05) for Season C.D. (0.05) for Part x Season	0.065 0.065 0.11
Group 8	C.D. (0.05) for Part C.D. (0.05) for Season C.D. (0.05) for Part x Season	0.01 0.01 0.08
Group 9	C.D. (0.05) for Part C.D. (0.05) for Season C.D. (0.05) for Part x Season	0.08 0.08 0.14

## CHAPTER V SUMMARY

The saprophytic fungi are versatile organisms and with their amazingly diverse species composition and ability to produce a variety of extra-cellular enzymes they act as regulators of the structure, function and dynamics of plant and animal community in nature. The nature and extent of mycoflora on plant litter, phylloplane, endophyte and soil is very complex and diverse and extensive literature is available on such studies carried out earlier. Amongst the microfungi isolated from varied substrates and habitats and so far recorded in literature, maximum species diversity was on decomposing plant litter.

This thesis embodies results of a study carried out during the last two years on the diversity, ecological association and activity of the sapophytic fungi associated with two locally grown plant species, *Ficus benghalensis* Linn. (F. Moraceae) and *Carissa congesta* Wight (F. Apocynaceae) and their immediate neighbourhood.

Partially decomposed leaf litter, young and mature fresh leaf, flower and fruit of *C.congesta* and *F. benghalensis* Linn., soil beneath the litter bed and air from 1 M above the ground level of the tree/bush canopy were the samples subjected for recovery of the associative fungi.

The leaf-litter and soil samples were subjected to particle plating (Bills & Polishook, 1994) and moist-chamber incubation (Hawksworth, 1974) techniques. Air samples were trapped in malt extract agar plates incorporated with antibiotics. Fresh leaves were subjected to a 3-step sterilization before the bits were planted in agar plates for the recovery of endophytic fungi. Conventional microscopic techniques and procedures were adapted for the study of fungi isolated.

A large number of isolates of fungi were recovered. All the sporulating fungi were identified down to the species level. Non-sporulating morphotypes were several and these were graded as 'dematiaceous' (coloured) and 'moniliaceous' (colourless) forms. Live cultures and herbarium specimens were maintained for all the fungi

described. Holotypes designated for the new taxa are housed in the herbarium of Goa University Fungus Culture Collection.

Sixty fungi that appeared commonly on leaf-litter, fresh leaf and soil samples and recovered in culture were subjected to amylase, cellulase, protease, pectinase, ligninase, laccase and xylanase enzyme assays using standard methods.

The data on fungal isolates were subjected to a '3- factor factorial completely randomised design analysis' defined in the 'Statistical Package for Social Sciences'. For endophytes, the '3² factor factorial completely randomised design analysis' of the SPSS was followed. To analyse the enzyme activity, 'Joining Tree Clustering' from "Statistica ver.5" was used. Square root transformations of the values obtained during the study was subjected to ANOVA Test. An exhaustive bibliography is given at the end of the thesis

About 5000 isolates of fungi were recovered. In all, these included 228 species of fungi belonging to 120 genera which included Mucorales, Hyphomycetes, Coelomycetes and Ascomycetes. Of these, 177 taxa belonging to 81 genera of Hyphomycetes, 4 species belonging to 2 genera of Mucorales, 12 species in 10 genera of Ascomycetes and 35 species belonging to 27 genera of Coelomycetes were recoverd. Sixteen hyphomycetes unlike any known species were not assigned to any known taxa since relevant literature was not available for identification. A total of 121 isolates did not sporulate in culture or on the substrate and recognised only as 'nonsporulating morphotypes' based on cultural characters. The detailed description, illustration and taxonomy of the mucoraceous and hyphomycetous fungi are given in the text. The Coelomycetes and Ascomycetes listed out here are not described in detail.

Of the 60 isolates subjected to enzyme assay, 29 showed positive activity against lignin and these were subjected for laccase and xylanase activity tests.

Twenty two species and 5 genera were new to science. These included: Acronidiellina indica sp. nov., Cercospora carriseae sp. nov., Cirrenalia indica sp. nov., Dicyma carisseae sp. nov., Doratomyces indicus sp. nov., Gonatobotryum bimorphospora sp. nov., Hyaloscolecobasidium indicum Gen. et sp. nov., Idriella mucoidea sp. nov., Idriella multiseptata sp. nov., Kumbhamaya indica Gen. et sp. nov., Moorella ficusensis sp. nov., Neocercosporella indica Gen. et sp. nov., Nigrospora endophytica sp. nov., Paracylindrocladia indica Genb. et sp. nov., Parahumicola endophytica Gen. et sp. nov., Phialocephala carisseae sp. nov., Phialocephala nephrospora sp. nov., Phialomyces microsporus sp. nov., Sclerographium goanensis sp. nov. and Spadicoides indicus sp. nov.

It is possible that particle-plating technique allowed the recovery of maximum diversity of fungi from the leaf particles. Eighteen taxa of well sporulating but unknown fungi have been recovered in this exercise.

In this investigation, many common (Hyphomycetes: Beltrania, Beltraniella, Helminthosporium, Idriella, Phialocephala, Gyrothrix, Periconia, Scolecobasidium, Vanakripa, Volutella, Weisneriomyces and Zygosporium, Coelomycetes: Ajrekarella, Ascochytula, Botryodiplodia, Camarosporium, Coniochaet, Diplodia, Discosia, Lasidiploidea, Monochaetia, Neottiospora, Phyllostica, Robillarda, Seimatosporium, Stagonospora, Trichosprerma and Vasudevella, Ascomycetes: Diatrype, Diatrypella, Hypoxylon, Guignardia, Lophiostoma and Xylaria, Mucorales: Mucor) and uncommon genera (Aspergillus, Curvularia, Fusarium, Paecilomyces, Penicillium, Tritirachium, Torula, Botryodiplodia, Chaetomella, Chaetomium, etc.) of fungi belonging to litter were recovered.

The species diversity studies revealed that there are a few abundant species and a high proportion of rare species of fungi associated with the two plant species.

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The fungi present on all substrates, belonged to 17 genera of Hyphomycetes and 3 Coelomycetes. In all, 121 nonsporulating forms were recovered. The identity of these is not known.

The endophytic fungi isolated belonged to genera Alternaria, Arxiella, Aspergillus, Beltrania, Cercospora, Curvularia, Cylindrocladium, Fusarium, Gliomastix, Hyaloscolecobasidium, Idriella, Nigrospora, Nodulisporium, Paecilomyces, Pseudobotrytis, Scolecobasidium, Stachybotrys, Wiesneriomyces, a few Undetermined Hyphomycete taxa, Ascochytula, Botryodiplodia, Discosia, Pestalotiopsis, Phomopsis, Robillarda, Septoria, Guignardia and Xylaria. Some of these were also recovered from the litter.

The endophytic fungi often recovered from the upper layers of forest litter are associated with litter decomposition. The endophytes which survive as latent internal colonizers of fresh leaves are believed also to take an active part in the degradation of the litter. This kind of shift in function, from hidden dormant colonization to active degradation, may be considered as an ecological adaptation associated with litter decomposition. It is also possible that some of the litter degrading fungi might have adjusted and taken refuge as endophytes during some part in the year. The endophytes are certainly not a group of veiled refuges in the leaf tissues but an active defense system against herbivory by insects and higher animals.

The most dominant group of fungi in this study was the dematiaceous phialidic group (G2) and the least was moniliaceous blastic (G3). The coloured fungi in general (G2, G9 and G4) were the dominant groups. As in other plants, colour or pigments in fungi confers protection from stress of light and other harmful effects. It is therefore apparent that dematiaceous or pigmented fungi were well adapted as dominant litter colonizers.

Seasonal effect on the occurrence of fungi was highly significant. The recovery of fungi, especially the dematiaceous phialidic forms, was highest during

monsoon followed by the post monsoon months and the least in summer. The high humidity combined with high ambient temperature of the monsoon months in this part of the tropical region favoured growth and sporulation of a large number of fungi on plant substrates.

The number of fungi isolated from leaf litter of both plants was more than those on soil and in air. Along with the soil and air mycoflora, the endophytic fungi were represented in low magnitude. Although fungi visit other substrates and environment in the neighbourhood such as the soil and air, the eventual site of survival for dematiaceous fungi is the litter. It is therefore not surprising to come across a large number of fungi in the leaf litter.

The basal part of the leaf showed maximum diversity of ascomycetous and hyphomycetous fungi, during pre-monsoon and monsoon months, than in the middle and tip portion. In the structure and dynamics of leaf growth and function, basal portion is the most enduring part and it is natural to expect occurrence of maximum species diversity of fungi in this part. Endophytic fungi get into the leaf tissue at an early period of leaf growth and it is possible that during the course of its colonisation, the fungi consume considerable time and apparently are unable to reach the tip or edge of the leaf.

Of the 60 isolates subjected to enzyme assay, 29 showed positive activity against lignin and these were subjected for laccase and xylanase activity tests. The results showed that on the whole maximum percentage of isolates exhibited protease activity, followed by laccase, pectinase, ligninase, amylase, xylanase and the least were with cellulase activity. The total isolates associated with *Carissa congesta* showed highest percentage for cellulase activity, followed by amylase, ligninase, protease, pectinase, xylanase and laccase activity. The total positive isolates

associated with *Ficus benghalensis* showed high percentage for laccase, followed by ligninase, pectinase, amylase, xylanase, protease and the least of cellulase.

For laccase, promising results were shown by 9 Hyphomycetes, 4 coelomycetous and 5 nonsporulating forms. Some of the isolates are powerful producers of laccase. These included Ajrekarella polychaetriae, Ascochyta caricae, Beltraniella buloloensis, Camarosporium indicum, Coniochaeta fuckelii, Cylindrocarpon ianthothele, Fusarium decemcellulare Gliomastix murorum, Periconia byssoides, Sporidesmium altum, Undetermined hyphomycete sp. 13 and the nonsporulating isolates NS.34, 45, 47, 48 and NS. 121.

The fungal isolates showing the maximum number and highest and least laccase activity were isolated from Carissa congesta. Very few of the isolates from Ficus benghalensis exhibited laccase activity. Quantitative analysis for laccase of fungi associated with Carissa congesta showed that 78% of fungal isolates had low and 6% had high activity whereas with fungi associated with Ficus benghalensis showed 45% with low and none with high activity.

All these confirmed that fungi armoured with variety of enzymes undertake saprophytic mode of activities. Further, isolation of fungi alone will not give any indication of their activities. These fungi when tested for enzymes, showed that they are also very creative.

Building up an 'enzyme profile catalogue' along with collection of 'pure cultures' and setting up of a 'taxonomy database', as did in this work is an effort of conservation and utilization of fungal biodiversity. Further, study such as this will have a far-reaching implication in our efforts of building up of biotechnology resource supply bank in India.

Comprehensive floristic surveys on saprophytic and endophytic fungi are very few. Documentation and maintenance of pure cultures of fungi in state-of-the-art culture collections as done here are not attempted earlier. Screening of fungi for enzymes and other secondary metabolites were also not an organized effort so far in our country. Therefore, documentation, culture collection and screening as carried out in this work will be very useful and invaluable for future biotechnology and utilization in our country.

The strength of this work lies on the information documented on the biodiversity of fungi of India with special reference to this part of the country, better understanding on the ecological relation of occurrence of fungi on plants and their neighbourhood and the enzyme profile built up for several of those isolated fungi. All these are small but valuable additions to the knowledge and growth of fungal science in our country.

## **ACKNOWLEDGEMENT**

## Acknowledgement

I express my sincere gratitude to my guide Prof. D.J.Bhat, Head, Department of Botany, Goa University for suggesting this topic and guiding me throughout the work.

I am grateful to Dr. Chandralata Raghukumar, Scientist, B.O.D., National Institute of Oceanography, Goa, for her support and guidance in conducting enzyme assays.

My heartfelt thanks are extended to Dr. R.N.S. Sundaram, Scientist; Mr. K. Ashok Kumar, Statistician, and Mr. R.G. Desai, Scientist, Indian Council of Agriculture Research, Old Goa, for their suggestions and assistance with statistical analysis.

I thank Prof. B.P.R. Vittal and Prof. J. Muthumary, C.A.S. in Botany, University of Madras, Chennai, for suggestions and help with identification of some of the fungi.

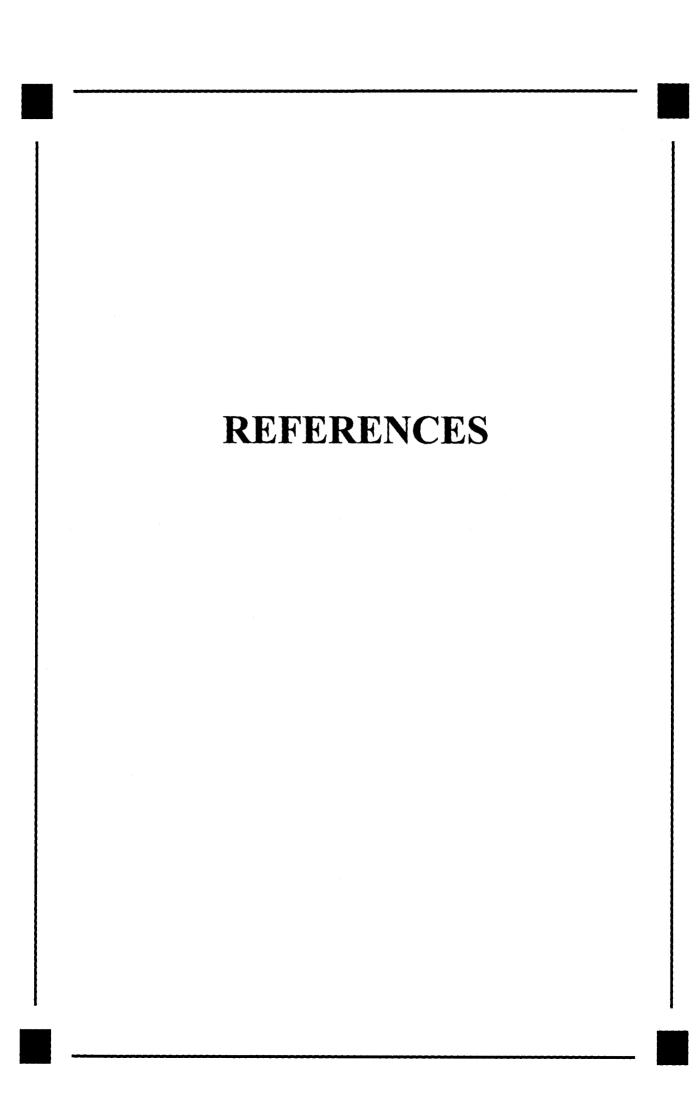
I thank Drs. M. Dreyfuss and C. Moller, Novartis Pharma Ltd., Switzerland, for suggestions and support.

I thank the Goa University authorities for providing me all facilities and a research fellowship during the tenure of this work. Grateful thanks are due to all my teachers in the Department of Botany, Goa University. Valuable suggestions and comments extended by Dr. Nandkumar Kamat, Research Scientist, Department of Botany, Goa University, is sincerely acknowledged.

My gratefulness also extends to my friends Gauri, Paranjalli, Alpha, Shyam, Glenda and Nilan.

I gratefully acknowledge the support my Parents, brothers and sister have given me all along.

Miriam Jacob



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# **APPENDIX**

**ANOVA**^a

					perimental Mo	ethod	
]			Sum of	df	Mean	F	Sig.
			Squares		Square		
SP1		GROUP	2.954	8	0.369	100.428	0.000
		SEASON	0.191	5	3.821E-02	10.391	0.000
		SUBSTRATE	.277	3	9.248E-02	25.151	0.000
}	2-Way Interactions	(Combined)	2.585	79	3.273E-02	8.900	0.000
		GROUP *					
}		SEASON	.369	40	9.213E-03	2.506	0.000
		GROUP *					
}		SUBSTRATE	2.066	24	8.610E-02	23.416	0.000
}		SEASON *					
j		SUBSTRATE	.150	15	1.002E-02	2.726	0.000
	3-Way Interactions	GROUP *					
		SEASON *	1.048	120	8.731E-03	2.374	0.000
		SUBSTRATE					
	Residual		1.986	540	3.677E-03		
	Total		9.041	755	1.198E-02		

SP1 by GROUP, SEASON, SUBSTRATE

## C.congesta

### ANOVA^a

				Expe	rimental Meth	od	
<u> </u>			Sum of Squares	df	Mean Square	F	Sig.
GR1	•	PART	3.641E-02	2	01.821E-02	3.891	0.030
		SEASON	1.489E-02	2	7.444E-03	1.591	0.218
	2-Way Interactions	PART *					
		SEASON	5.6953E-02	4	1.424E-02	3.042	0.029
	Residual		0.168	36	4.679E-03		
	Total		0.277	44	6.289E-03		

GR1 by PART, SEASON

## C.congesta

### ANOVA^a

			<u> </u>	Expe	rimental Meth	od	
			Sum of Squares	df	Mean Square	F	Sig.
GR2		PART	2.2986	2	1.493	86.017	0.000
		SEASON	2.262	2	1.131	65.162	0.000
Ì	2-Way Interactions	PART *	ı				
		SEASON	1.945	4	0.486	28.008	0.000
	Residual		0.625	36	1.736E-02		
	Total		7.818	44	0.178		

GR2 by PART, SEASON

## C.congesta

ANOVA^a

				Expe	rimental Meth	ıod	
			Sum of Squares	df	Mean Square	F	Sig.
GR4	•	PART	0.372	2	0.186	60.350	0.000
		SEASON	0.160	2	8.023E-02	26.040	0.000
	2-Way Interactions	PART *					
		SEASON	4.013E-02	4	1.003E-02	3.257	0.022
	Residual		0.111	36	3.081E-03		
	Total		0.683	44	1.553E-02		

GR4 by PART, SEASON

## C.congesta

### **ANOVA**^a

				Expe	rimental Meth	od	
			Sum of Squares	df	Mean Square	F	Sig.
GR8		PART	5.093E-02	2	2.546E-02	13.491	0.000
		SEASON	4.519E-03	2	2.260E-03	1.197	0.314
	2-Way Interactions	PART *					
		SEASON	5.968E-03	4	1.492E-03	0.791	0.539
	Residual		6.794E-02	36	1.887E-03		
	Total		0.129	44	2.940E-03		

GR8 by PART, SEASON

## C.congesta

ANOVA*

				Expe	rimental Meth	nod	
			Sum of Squares	df	Mean Square	F	Sig.
GR10	,	PART	2.822	2	1.411	227.508	0.000
l		SEASON	3.341E-02	2	1.670E-02	2.693	0.081
	2-Way Interactions	PART *					
		SEASON	0.105	4	2.613E-02	4.212	0.007
	Residual		0.223	36	6.202E-03		
	Total		3.183	44	7.235E-02		

GR10 by PART, SEASON

## F.benghalensis

**ANOVA**^a

				Expe	rimental Meth	od	
			Sum of Squares	df	Mean Square	F	Sig.
GR2		PART	0.314	2	0.157	16.904	0.000
		SEASON	0.255	2	0.128	13.720	0.000
	2-Way Interactions	PART *					
		SEASON	9.513E-02	4	2.378E-02	2.557	0.055
	Residual		0.335	36	9.302E-03		
	Total		1.000	44	2.272E-02		

GR2 by PART, SEASON

## F.benghalensis

**ANOVA**^a

				Expe	rimental Metl	nod	
ĺ			Sum of	df	Mean	F	Sig.
			Squares		Square		
GR7		PART	7.464E-02	2	3.732E-02	25.609	0.000
		SEASON	6.790E-03	2	3.395E-03	2.330	0.112
	2-Way Interactions	PART *					
		SEASON	1.258E-02	4	3.146E-03	2.159	0.093
	Residual		5.246E-02	36	1.457E-03		
	Total		0.146	44	3.329E-03		

GR7 by PART, SEASON

## F.benghalensis

### **ANOVA**^a

				Expe	rimental Meth	od	
			Sum of Squares	df	Mean Square	F	Sig.
GR9		PART	0.805	2	0.402	118.180	0.000
		SEASON	2.562E-02	2	1.281E-02	3.761	0.033
	2-Way Interactions	PART *					
		SEASON	2.519E-02	4	6.298E-03	1.849	0.141
	Residual		0.123	36	3.406E-03		
	Total		0.978	44	2.224E-02		

GR9 by PART, SEASON

### F.benghalensis

**ANOVA**^a

				Expe	rimental Meth	ıod	
			Sum of Squares	df	Mean Square	F	Sig.
GR4		PART	0.348	2	0.174	31.075	0.000
		SEASON	0.240	2	0.120	21.477	0.000
1	2-Way Interactions	PART *				ı.	
		SEASON	8.552E-02	4	2.138E-02	3.819	0.011
	Residual		0.202	36	5.599E-03		
	Total		0.876	44	1.990E-02		

GR4 by PART, SEASON

## F.benghalensis

### **ANOVA**^a

				Experimental Method					
			Sum of Squares	df	Mean Square	F	Sig.		
GR6		PART	0.106	2	5.308E-02	3.710	0.034		
		SEASON	4.858E-02	2	2.429E-02	1.698	0.197		
	2-Way Interactions	PART *							
		SEASON	0.107	4	2.683E-02	1.875	0.136		
	Residual		0.515	36	1.431E-02				
	Total		0.777	44	1.766E-02				

GR6 by PART, SEASON

PII: S0181158400001160

#### Two new endophytic conidial fungi from India

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Abstract — Two new taxa of endophytic conidial fungi isolated from fresh young leaves of Carissa carandas L. are described and illustrated. These include a new anamorph-genus Kumbhamaya and a new species of Gonatobotryum. © 2000 Adac / Éditions scientifiques et médicales Elsevier SAS

Anamorphs / new conidial fungi / hyphomycetes / taxonomy / endophytes / biodiversity / India

#### INTRODUCTION

The discovery of endophytic fungi has attained significance in recent years because they sometimes elaborate secondary metabolites useful in biocontrol, or in industry or as pharmaceuticals (Bills, 1995). During our investigation of the taxonomy and biology of endophytic fungi occurring in leaves of Carissa carandas L. (Apocynaceae), an evergreen shrub growing on lateritic soils on the west coast of India, several fungi were isolated in culture from fresh leaves gathered from Taleigao and Verna Plateau in Goa, among which the following two taxa of conidial fungi are believed to be new to science. They are described and illustrated below.

#### MATERIALS AND METHODS

Methods discussed by Petrini (1986) for isolation of endophytic fungi were followed in this work. Five randomly chosen disease-free mature and young leaves of C. carandas, freshly gathered and thoroughly washed in tap water, were surface sterilized, first in 70 % ethanol for 30 s, then in 4 % Sodium hypochlorite for 1 min. and finally in 70 % ethanol for 30 s. The surface sterilized leaves were cut into 1 mm² size pieces using a sterile razor blade. About 12 pieces were aseptically placed equidistant in rows in a 10 cm diam. Petri plate containing antibiotic incorporated malt extract agar medium (composition in 1 L: 5 g malt extract, 20 g agar, 1000 ml distilled water, 0.02 g each of bacitracin, neomycin, penicillin, polymixin, streptomycin and tetracycline). The process was repeated for other leaves. The plates were incubated at 23 °C. As the fungal colonies appeared from the edges of the leaf bits and extended into agar medium, generally after 5-7 days, these were aseptically transferred to agar slants.

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#### RESULTS AND DISCUSSION

Several interesting and rare fungi were recovered from leaves and the following two conidial fungi are described as new taxa.

Kumbhamaya M.Jacob et D.J. Bhat anam. gen. nov.

(Etym. Sanskrit: Kumbha = kettle or pitcher-shaped [ = pyriform] phialide; maya = numerous)

Ad fungos conidiales, hyphomycetes pertinens. Coloniae effusae, brunneae ad atro-brunneae. Mycelium densum, ramosum, septatum, ex hyphis brunneis ad atro-brunneis constans. Conidiophora mononematosa, indistincta, septata. Cellulae conidiogenae monophialidicae, in conidiophoris non-ramosis terminales incorporatae, lageniformes, vel laterales, pyriformes, ad basim elongatae, erectae ad curvatae, crassitunicatae, laeves, brunneae vel atrobrunneae, collari prominente et expanso, ad basim constrictae. Conidia solitaria, fusiformia, curvata vel sigmoidea, utrinque acutata, sursum rostrata, saepe ad basim recurvata, hyalina, crassitunicata, laevia, 1 – 3-septata, in massis mucosis aggregata.

Typus Kumbhamaya indica M.Jacob & D.J. Bhat.

Conidial fungi, hyphomycetes. Colonies effuse, medium to dark brown. Mycelium dense, with branched, thickly septate, medium to dark brown hyphae. Conidiophores mononematous, indistinct, septate. Conidiogenous cells monophialidic, integrated, unbranched, terminal or intercalary, kettle- to pitcher-like (= pyriform), often with an elongated base, straight to curved, thick-walled, smooth, medium to dark brown, with a distinct and flared collarette, constricted and narrow at the base. Conidia solitary, fusiform, curved to sigmoid, pointed at both ends, sharply beaked at the tip, often slightly recurved at the base, hyaline, thick-walled, smooth, colourless, 1 – 3-septate, aggregating in a slimy mass.

Several genera of mononematous hyphomycetes with conspicuous phialides bearing flared collarettes and producing elongate and nonsetulate conidia in slimy heads are known (Bhat and Kendrick, 1993; Carmichael et al. 1980; Ellis, 1971, 1976; Matsushima, 1971, 1975). These include Craspedodidymum Holubova-Jechova (1972), Dischloridium Sutton, Fusarium Link, Fusariella Sacc., Phialophora Medlar and Phialomyces Misra & Talbot (Carmichael et al., 1980). In Craspedodidymum, the apically inflated phialides with large funnel-shaped collarette, produce brown, non-septate conidia. In Dischloridium, the phialides lack a conspicuous collarette and the conidia are non-septate. In the moniliaceous genus Fusarium, the branched conidiophores terminating in apical or lateral phialides produce globose to obclavate, nonseptate, hyaline microconidia and fusiform, curved, septate, hyaline, apically and basally pointed macroconidia which superficially resemble those of Kumbhamaya. The hyaline phialides in Fusarium however are inconspicuous. In Fusariella, the phialides lack a conspicuous collarette and the conidia are fusiform, septate, curved, medium brown and produced in linearly arranged false chains. In Phialophora, the phialides are small and the conidia are nonseptate, ellipsoidal and inconspicuous. In Phialomyces, although the phialides typically have a flared collarette, the conidia are not only globose but also in chains. Kumbhamaya differs from all these genera in that the kettle or pitcher-shaped (= pyriform) phialides bearing conspicuous and flared collarette produce fusiform, curved septate, hyaline conidia which are pointed at both ends and they are never in chains.

#### Kumbhamaya indica M.Jacob et D.J. Bhat anam. sp. nov. (Figs 1, 2)

Coloniae in agaro maltoso effusae, reverso atrobrunneo. Mycelium aerit densum, hyphae ramosae, crassiseptatae, medio-brunneae vel atrobrunneae, 2.2 – 7.5 latae. Conidiophora mononematosa, indistincta, septata, 2.2 – 2.5 µm diam. Celli

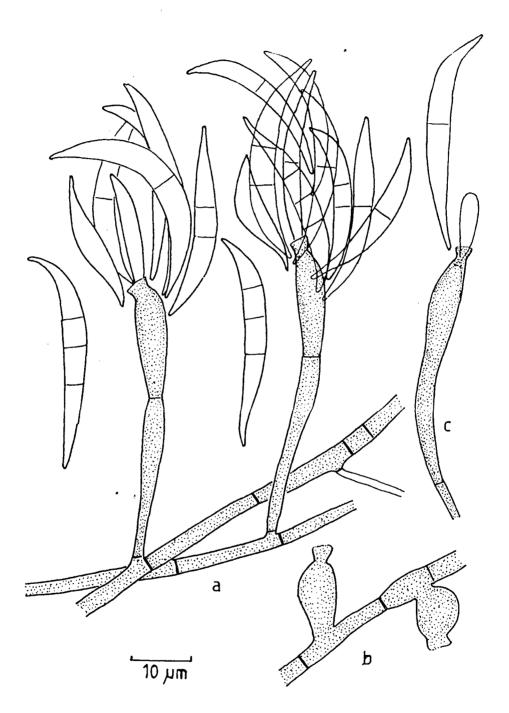


Fig. 1.a-c: Kumbhamaya indica. a: Note the slimy conidial mass at the tip of the phialides. b: Conidiophores. c: Conidial development.

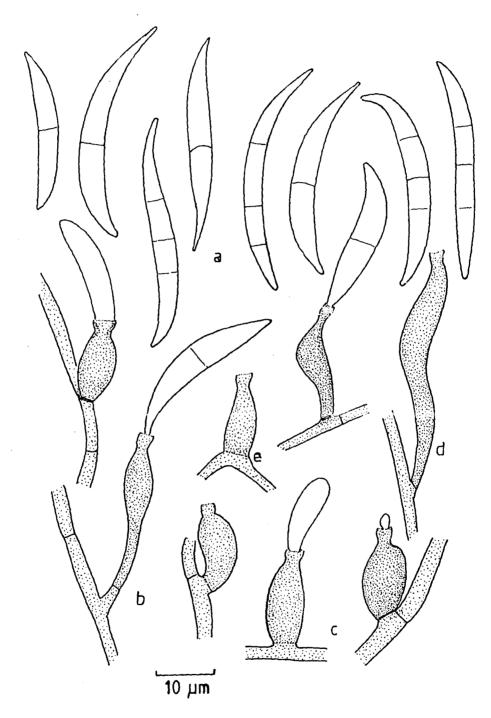


Fig. 2.a-e: Kumbhamaya indica. a: Conidia. b: Conidiophore with mature conidia. c-d: Conidiophores showing variations in size. e: Phialide showing wide collarette.

conidiogenae monophialidicae, in conidiophoris incorporatae, terminales vel laterales, pyriformes ad vermiformes, ad basim elongatae, rectae vel curvatae, crassitunicatae, laeves, brunneae vel atrobrunneae,  $12-50~\mu m$  longae et ad basim  $2.5-4.5~\mu m$  latae, in medio  $5.5-8.5~\mu m$  latae, collari prominente et expanso  $2-3.5\times1.5-2.5~\mu m$ . Conidia solitaria, fusiformia, curvata vel sigmoidea, utrinque acuta, falciformia, sursum rostrata, ad basim saepe recurvata, hyalina, crassitunicata, laevia, 1-3-septata,  $25-40\times3.5-5.5~\mu m$ , in massis mucosis aggregata.

Colonies on malt extract agar effuse, with moderately dense aerial mycelium, irregular at margin, medium brown at the centre, pale brown towards the periphery,  $2-3.5\,\mathrm{cm}$  diam. in 7 days; reverse of the colony dark brown. Mycelium densely branched, with thick septa, medium to moderately dark brown hyphae  $2.2-7.5\,\mu\mathrm{m}$  wide. Conidiophores mononematous, indistinct, septate,  $2.2-2.5\,\mu\mathrm{m}$  diam. Conidiogenous cells monophialidic, integrated, terminal or lateral, vase-like (= pyriform) to vermiform, often with an elongate base, straight to curved, thick-walled, smooth, medium to dark brown,  $12-50\,\mu\mathrm{m}$  long and  $2.5-4.5\,\mu\mathrm{m}$  wide at the base,  $5.5-8.5\,\mu\mathrm{m}$  wide in the middle, with a distinct and flared collarette  $2-3.5\times1.5-2.5\,\mu\mathrm{m}$ , constricted and narrow at the neck region. Conidia solitary, fusiform, curved to sigmoid, pointed at both ends, sharply beaked at the tip, occasionally recurved at the base, hyaline, thick-walled, smooth, mostly 3-septate, rarely 1-septate,  $25-40\times3.5-5.5\,\mu\mathrm{m}$ , aggregating in a slimy mass at the apex of the phialide.

Holotypus: Slide prepared from a dried culture grown on MEA medium and isolated from tip of young leaf of Carissa carandas, 14.11.1997. Verna Plateau, Goa, M. Jacob Herb. No.GUFCC-0233, PC; Paratype: Dried culture of the fungus grown on MEA medium and isolated from mature leaf of Bambusa sp., 24.2.1999, Mollem, Goa, Maria D'Souza, Herb. No. GUFCC-0238. Living cultures of same are also maintained at Goa University Culture Collection.

Gonatobotryum bimorphosporum M. Jacob et D.J. Bhat sp. nov. (Fig. 3)

Coloniae in agaro maltoso effusae, atrobrunneae, floccosae. Mycelium partim superficiale, partim immersum, hyphis marginem versus modo rhizoideorum compactis, septatis, ramosis, laevibus,  $4.5-10\,\mu m$  latis. Conidiophora distincta, mononematosa, recta vel flexuosa, brunnea, septata, non-ramosa, laevia,  $175-550\,\mu m$  longa,  $6.2-10\,\mu m$  lata, percurrenter elongascentes, in nodis terminalibus vel intercalaribus echinulata, ampullae conidiogenae  $13.5-45\times15-22\,\mu m$ . Cellulae conidiogenae polyblasticae, integratae, terminales vel intercalares, sphaericae vel subsphaericae, loci conidiogeni prominentes truncati. Conidia catenata, 0-septata, laevia, subhyalina vel brunnea, ad basim cicatricem ferentia, duarum formarum: conidia primaria e cellulis conidiogenis oriunda, elongato-ellipsoidea vel elongato-obclavata, 1-3 locos conidiogenos ferentia,  $7.5-11.5\times3-4.5\,\mu m$ ; conidia secundaria ex conidiis primariis oriunda, ellipsoidea, basipetalia,  $3-6.2\times2.5-3.5\,\mu m$ .

Colonies in malt extract agar effuse, moderately fast growing, dark brown, with irregular margin, with rough surface, with faint concentric zones, attaining 4-4.5 cm diam. after 7 days. Mycelium partly immersed, partly superficial, often compacted into root-like aggregates towards the periphery of the colony, with septate, repeatedly branched, smooth-walled, pale brown, hyphae  $4.5-10\,\mu\mathrm{m}$  wide. Conidiophores distinct, mononematous, flexuous to erect, brown, septate, unbranched, smooth,  $175-550\,\mu\mathrm{m}$  long,  $6.2-10\,\mu\mathrm{m}$  wide, percurrently regenerating, nodose and distinctly echinulate in the terminal and intercalary conidiogenous ampullae  $13.5-45\times15-22\,\mu\mathrm{m}$ . Conidiogenous cells polyblastic, integrated, terminal or intercalary, spherical to subspherical, ampullae with slightly raised and truncate conidiogenous loci. Conidia catenate, 0-septate, smooth, pale to moderately brown, cicatrized at the base, of two types. First-formed conidia

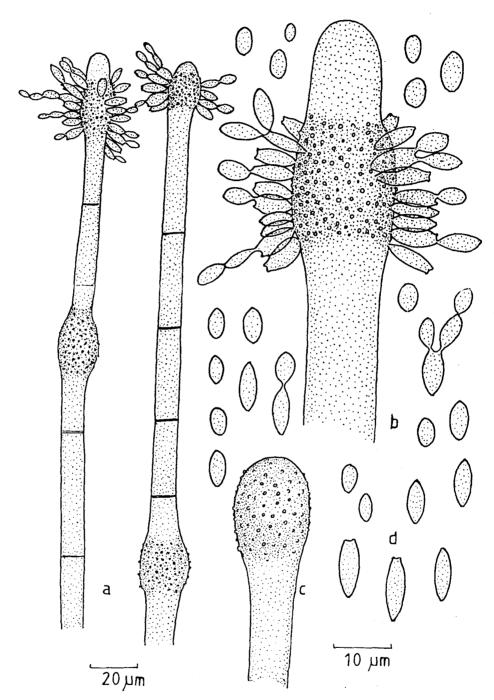


Fig. 3.a-d: Gonatobotryum bimorphosporum. a: Conidiophore with conidia. b: Polyblastic conidiogenous cells with 2 distinct type of conidia. c: Conidiogenous ampullae showing cicatrized scars. d: First formed and later formed conidia

arising synchronously and directly from conidiogenous cells, elongate-ellipsoidal to elongate-obclavate, with 1-3 apical conidiogenous loci,  $7.5-11.5\times3-4.5\,\mu\mathrm{m}$ ; later formed conidia arise from first-formed conidia, ellipsoidal, in branched basipetal chains,  $3-6.2\times2.5-3.5\,\mu\mathrm{m}$ .

Holotypus: Slide prepared from dried agar culture mat of the fungus isolated from fresh leaves of Carissa carandas, 22.11. 1998, Taleigao Plateau, M. Jacob Herb. No. GUFCC 0398, PC. Living culture of same maintained at Goa University Culture Collection.

The genus Gonatobotryum Saccardo, with G. fuscum (Sacc.) Sacc. as type species, is characterized by mononematous conidiophores producing integrated, terminal or intercalary, percurrent and polyblastic conidiogenous cells and catenate conidia (Carmichael et al., 1980; Ellis, 1971). The species so far described in the genus, viz. G. fuscum (Ellis, 1971), G. apiculatum (Peck) Hughes (Ellis, 1971) and G. indicum Manjal and Gill (Manjal and Gill,1963) are compared with G. bimorphosporum (Tab. 1). G. apiculatum, G. fuscum and G. indicum are saprophytic, parasitic or hyperparasitic and produce only one type of conidia whereas G. bimorphosporum is endophytic and produces two morphologically distinct types of conidia.

Tab. 1. Comparison of morphology and habitat of the species of Gonatobotryum

Species	Conidiophore	Ampullae	Conidia	Habitat Substrate/Host Distribution
G. fuscum (Sacc.) Sacc.	up to 700 µm long and 12 – 15 µm wide; Percurrently proliferating	up to 28 μm diam.	One type;  10-25 × 6-13 µm, Catenate, ellipsoidal to oblong, rounded at both ends.	Saprophytic on rotten wood of Quercus and Fagus; Hyperparasitic on Ceratocystis and other fungi; Europe and America.
G. apiculatum (Peck) Hughes	200 – 300 × 6 – 9 μm	9 – 15 μm diam.	One type; 5-12×2.5-6 µm In branched chains, limoniform.	Parasitic on leaves of <i>Hamamelis</i> ; also recorded on <i>Rhus</i> and soil; N. America.
G. indicum Munjal & Gill	250 – 420 × 7 – 9 μm	11 – 17 μm diam.	One type; 5.5 - 9 × 3.5 - 5 µm Ovate to oblong, One end broadly obtuse, the other tapering.	Saprophytic on dead twigs; India.
G. bimorphosporum sp. nov	175 – 560 × 6 – 10 μm	7.5	Two types: First formed conidia elongate - ellipsoidal to obclavate; -11.2 × 2.9 - 3.7 µm. Later-formed conidia ellipsoidal; 2 - 6.2 × 2.5 - 3.7 µm	Endophyte in leaves of Carissa carandas  L; India.

Acknowledgements. Financial assistance from the Department of Science and Technology, Govt. of India, New Delhi in the form of a research grant to Prof. D. J. Bhat is gratefully acknowledged.

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