STUDIES ON DIVERSITY, ECOLOGY AND BIOLOGY OF AQUATIC FUNGI OF SOME FRESHWATER STREAMS OF WESTERN GHAT FORESTS IN GOA STATE, INDIA

Thesis Submitted to THE GOA UNIVERSITY for the Award of The Degree of

DOCTOR OF PHILOSOPHY IN BOTANY

Ву

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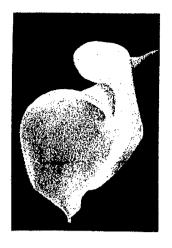
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DECLARATION

I hereby declare that the Ph.D. thesis entitled "STUDIES ON THE DIVERSITY, ECOLOGY AND BIOLOGY OF AQUATIC FUNGI FROM SOME FRESHWATER STREAMS OF WESTERN GHAT FORESTS IN GOA" 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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CERTIFICATE

I certifiy that the thesis entitled "STUDIES ON THE DIVERSITY, ECOLOGY AND BIOLOGY OF AQUATIC FUNGI FROM SOME FRESHWATER STREAMS OF WESTERN GHAT FORESTS IN GOA" submitted by Ms. Sreekala. K. Nair, is a record of research work done by her during the period from 1999-2002 when she worked under my supervision. The thesis has not formed the basis for the award of any degree, diploma or associateship to Ms. Sreekala. K. Nair.

I affirm that the thesis submitted by Ms. Sreekala. K. Nair incorporates the independent research work carried out by her under my supervision.

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CHAPTER 1: INTRODUCTION

1.1. Introduction:

Mycota that grow on decaying leaves, twigs and other plant parts submerged in water and complete the whole or part of their life cycle in streams, rivulets and rivers are called 'aquatic fungi' and these include members of Zygomycotina, Ascomycotina, Basidiomycotina and Deuteromycotina (Ingold, 1975). They are also referred as 'waterborne' or 'freshwater' fungi (Dix and Webster, 1995). The fungi growing in lentic water system such as ponds, tanks and lakes are largely zoosporic (Sparrow, 1960). Amongst the mycota of freshwater streams, hyphomycetous fungi are the largest group. In view of the pioneering contribution by Professor C.T. Ingold over a period of three decades (1942-1975) to the study of taxonomy and ecology of aquatic fungi, the freshwater hyphomycetes were often referred as 'Ingoldian fungi' (Webster and Descals, 1981). The fungi living in oceans and seas are another specialized group of aquatics called as 'marine fungi' (Kohlmeyer and Kohlmeyer, 1979).

Aquatic fungi in freshwater habitats, are represented by two major groups namely Saprolegniales (watermoulds) and aquatic Hyphomycetes. A number of fungi belonging to Ascomycotina and Mastigomycotina (zoosporic fungi) and a few Basidiomycetes also occur in freshwater habitats but in the dynamics of freshwater stream ecosystem, hyphomycetous fungi are considered as the most significant participants in view of their ability to digest a variety of submerged organic matter (Barlocher, 1982, 1985, 1992; Barlocher and Kendrick, 1973a, b, 1974, 1976;

Kaushik and Hynes, 1968, 1971; Graca et al., 1993a, b). The streams contain sufficient run off organic material which in the living or dead state serve as a constant source of nutrients for the fungi living in water.

1.2. <u>Definition</u>:

Although an aquatic fungus is defined as one capable of completing its life cycle underwater, it is now known that quite a few freshwater fungi collected as aquatic spora in water do not sporulate below the surface but produce their spores above water (Webster, 1981). Some aquatic species sporulate when placed in contact with droplets of water in a terrestrial habitat (Ando, 1992). Many conidial fungi produce their teleomorphs on submerged decaying litter when the streams dry out (Casas and Descals, 1997). Kendrick (1992) referred all these as 'amphibious fungi'.

1.3. Diversity:

Of the so far known 70,000 plus fungi, as of today, nearly 330 species of freshwater fungi have been described in 140 genera (Sridhar et al., 1992; Descals, per. comm.). Of these, majority are members of Deuteromycotina which reproduce by formation of mitospores or conidia. The conidia of freshwater hyphomycetes are small to unusually large, many measuring up to 100 µm diam. The aquatic spora exhibit distinctive shapes - namely elongated, sigmoid or helical; forked or branched - triradiate, tetraradiate or multiradiate, and/or often appendaged. These conidia, produced in abundance, are well-adapted to underwater dispersal and trapping but germinate soon after coming in

contact with any solid surface (Webster and Davey, 1984). Because of their characteristic conidial shape, the identification of Ingoldian fungi is often done by distinguishing their spores alone, which is not the case in most other groups of fungi (Dix and Webster, 1995).

1.4. Aquatic endophytes:

Riparian plants extend their roots into streams and rivers. The healthy and decaying roots of the plants are often colonized by aquatic fungi and so far, 20 species have been reported as aquatic root endophytes (Fisher et al., 1991; Sridhar and Barlocher, 1992; Raviraja et al., 1996). It has been pointed out that the submerged living roots of riparian trees provide a perennial source of space for survival of aquatic hyphomycetes (Iqbal et al., 1995). However, the role of these root-associated aquatic endophytes in plant nutrition, root absorption efficiency, protection against infection and root senescence is not known.

1.5. <u>Ecology</u>:

Freshwater fungi play an important role in conditioning and processing of leaf debris and render it more palatable and nutritionally enriched to aquatic invertebrates (Barlocher, 1985, 1992). The increased nutritive value of colonized leaves is associated with an increase in protein content. Presumably, the fungi are capable of obtaining soluble inorganic nitrogen available in low concentration in stream water and to form microbial biomass making use of carbon available in leaf tissues

(Webster and Descals, 1979). They are also known to produce an array of enzymes capable of macerating leaf tissues (Chamier and Dixon, 1982; Zemek et al., 1985). Due to their ability to grow at low water temperatures and to produce extracellular enzymes, aquatic hyphomycetes are considered important in release of nutrients locked up in the structural plant material (Chandrasekhar and Kaveriappa, 1988, 1991; Rodrigues et al., 1997). From the work of Graca (1993), it is now certain that the plant detritus is ingested by invertebrates only after colonization by these fungl. Most streams receive bulk of their carbon input not from attached algae or macrophytes but from the litter deposits of adjacent trees. Since the tree leaf litter is initially a relatively poor food source for many aquatic animals, it is believed that the fungi provide an essential link in food chain from the primary producers, i.e. the riparian vegetation, to the detritus consumers, i.e. the aquatic animals (Kaushik and Hynes, 1971; Barlocher and Kendrick, 1973 a, b; 1976; Webster and Descals, 1979; Graca, 1993).

1.6. Importance of the fungi:

It is now clearer that microfungi from the tropics are potential source of biotechnologically significant and pharmaceutically valuable compounds (Dreyfuss and Chapela, 1994; Bills, 1995); Cuomo et al., 1995). Realizing these exciting revelations, Rossman (1994) and Casas and Descals (1997) advocated for complete and urgent inventory, recovery and investigations on aquatic fungi of the streams of tropical forests, since this region of the world is presently under serious ecological stress.

1.7. The Present work:

Hitherto studies on the taxonomy, diversity, ecology, distribution and secondary metabolites of freshwater fungi have been confined largely to temperate region (Dix and Webster, 1995; Casas and Descals, 1997). The aquatic fungi of the streams of Western Ghat forests have been a subject of study since the publication of a preliminary survey of 11 streams of the region carried out by Subramanian and Bhat (1981) wherein Anguillospora longissima (de Wildem.) Ingold, Tetracladium setigerum (Grove) Ingold and Triscelophorus monosporus Ingold were reported for the first time from Dudhsagar Falls in Goa State. Since then, a number of publications appeared on the fungi of forest streams of Western Ghats in Karnataka and Kerala and these have been reviewed in detail by Sridhar et al. (1992).

1.7.1. Objectives:

In the absence of any detailed and intensive investigation on the mycota of freshwater streams of northern part of Western Ghats, an effort was made in this thesis to study the diversity, ecology and activity of the aquatic fungi of the streams of forests of Western Ghats in Goa region with following objectives: to study the following aspects.

- Taxonomy, cultural characters and diversity of aquatic fungi.
- Seasonal appearance, abundance and ecology.
- In situ survival mechanisms adapted for dry season by fungi.
- Effect of mining rejects on growth and sporulation.

• Enzyme production by aquatic fungi.

Hitherto known information on the diversity, taxonomy, ecology and activity of aquatic fungi has been reviewed in detail in the thesis. State-of-the-art techniques have been used in this work. The collection and isolation of aquatic fungi were done for two years during February 1999 to January 2001 from several streams in the State of Goa. The endophytic fungi isolated from submerged roots of riparian trees have been studied. Besides morphological features, details of cultural characters, habitat and substrate affinity were compiled on a specially prepared data-sheet. These were used in the diagnosis of all taxa recovered during the study. Standard and relevant taxonomic keys and literature were referred for identification of the fungi.

The results are presented in 4 parts in the thesis. In Part I, taxonomic diversity and habitat affinity of the fungi collected from 12 freshwater streams of Goa are detailed out. In Part II, seasonal appearance and abundance of aquatic fungi in three freshwater streams of the wildlife sanctuaries of Molem, Bondla and Cotigao in Goa during the two years study period are elaborated. The diversity and abundance of these fungi in different sites are statistically analyzed. In Part III, the survival mechanism adopted by these fungi for dry season is explained based on a specially designed *in situ* litter-bag experiment and analysis of root endophytes. The effect of mining rejects on growth and sporulation of aquatic hyphomycetes studied is also given. In Part IV, the results of study on the ability of aquatic fungi to produce different enzymes are elaborated.

A list of references is given at the end of the thesis. A few research publications accomplished during the study period are appended as appendix.

CHAPTER 2: REVIEW OF LITERATURE

A broad understanding of 'freshwater fungus' in a lotic system refers to any species that depends on freshwater of streams or rivers for performance of its whole or part of the life cycle (Ingold, 1975; Thomas, 1996). The freshwater fungi are aquatic or semi-aquatic in their habitat and depend on submerged organic matter for sustenance. Diverse fungi are known from freshwater stream habitats. These include representatives from different taxonomic groups, namely the zoosporic (Fuller and Jaworski, 1987; Sparrow, 1968), zygomycetous (Descals and Webster, 1984), ascomycetous (Hyde, 1993; Hyde et al., 1996; Jones, 1976; Shearer, 1993), basidiomycetous (Marvanova and Barlocher, 2000; Nawawi, 1985e; Nawawi and Kuthubutheen, 1988g; Nawawi and Webster, 1982; Nawawi et al., 1977a-b; Shaw, 1972) and conidial fungi (Cribb and Cribb, 1993; Ingold, 1975; Marvanova and Hywell-Jones, 2000; Nawawi, 1973a-d, 1974a-c, 1975a-b, 1976a-d, 1982, 1985a-d, 1987a-b; Nawawi and Kuthubutheen, 1987a-c, 1988af, 1989a-c, 1990a-d). Among these, moniliaceous conidial fungi, known as freshwater hyphomycetes, water-borne hyphomycetes Ingoldian fungi (Moniliales, Deuteromycotina) are the largest and well-documented group. Their biology and diversity is more understood and comprehensive than other groups in the aquatic environment.

Aquatic fungi of lotic environment have now become a discipline of their own. According to Barlocher (1992), this has been possible largely through contribution of three distinguished mycologists namely, C.T. Ingold (1942-1975) who discovered them, J. Webster (1973-1995) who worked out much of their biology and ecology and H.B.N. Hynes (1981-1983) who emphasized the role of these fungi in energy-flow of stream ecosystem and the food-chain linkage between leaves and invertebrates through fungal biomass.

2.1. Earlier Studies on Aquatic Fungi:

Researches on freshwater stream fungi began more than 50 years ago when Ingold (1942) published the first of his many papers from England and convincingly proved that fungi regularly occur on submerged decaying leaves, twigs, and wood of dicotyledonous trees and shrubs in running waters (Ingold, 1942; 1943a-c; 1944). Since then, there have been extensive studies and freshwater fungi were reported from several continents such as Africa, America, Asia, Australia and Europe (Ingold, 1975; Dix and Webster, 1995).

It is now evident that the freshwater fungi have a worldwide distribution. Diversity and distribution of these fungi have been studied by several workers around the world and a few notable names are given below country-wise:

List of important workers of freshwater fungi (Descals, 1995; Sati and Tiwari, 1997):

Country

Name of Researchers

England E. Descals; E. B. Garath-Jones; C.T. Ingold; D. Park; J. Webster.

Canada F. Barlocher; H.B.N. Hynes; B. Kendrick.

Czechoslovakia P. Marvan; L. Marvanova.

Germany G. R. W. Arnold.

India D.J. Bhat; K.R. Chandrashekar; K.M. Kaveriappa; C. Manoharachary; K.

Rajashekar; S.C. Sati; K.R. Sridhar; C.V. Subramanian; N. Tiwari

Africa S.O. Alasoadura; D.J. Bhat; C.T. Ingold; Le John; N. Singh.

Japan K. Ando; T. Matsushima; K. Miura; K. Tabaki.

Malaysia K. Kuthubutheen; A. Nawawi.

Pakistan S.H. Iqbal

Sweden, Spain A. Muller-Haeckel; S. Nilsson.

U.S.A. B.J. Dyko; R.D. Goos; H.J. Hudson R.H. Petersen; F.V. Ranzoni;

USSR I. O. Dudka; A. Menelik

Specific works included, of the temperate countries such as England (Ingold, 1975;

Descals and Webster, 1981; Shearer and Webster, 1985), Canada (Barlocher, et al., 1977,

1992; Michaelides and Kendrick, 1978), Italy (Del Frate and Caretta, 1983), France (Merce, 1987; Chauvet and Merce, 1988; Chauvet, 1989), Hungary (Gonczol, 1971, 1975, 1987), Czechoslovakia (Marvanova, 1973) Switzerland and Germany (Barlocher and Rosset, 1981; Wood-Eggenschwiler & Barlocher, 1983), USA (Metvalli and Shearer, 1989), northern Pakistan (Iqbal, 1992; Iqbal, et al., 1979, 1980, 1990), Australia (Shaw, 1972; Shaw and Sutton, 1985) and Japan (Ando, 1992).

Freshwater fungi have been reported and described from several parts of tropical and subtropical countries: Ethiopia (Bhat and Chien, 1990), Ghana, Ivory Coast, Zimbabwe (Ingold, 1956, 1958, 1959, 1960; Dixon, 1959; Le' John, 1965) Nigeria, (Alasoadura, 1968a-c; Singh, 1972, 1976; Singh & Musa, 1977), Cuba (Marvanova and Marvan, 1969), Dominican Republic (Betancourt, et al., 1986), Hawaiian Islands (Anastasiou 1964, Goos, 1970, 1978; Ranzoni, 1979), Indonesia (Nayo, 1975), Jamaica (Hudson and Sutton, 1964; Hudson, 1961; Crane and Dumont, 1975), Malaysia (Nawawi, 1985a-b, 1987; Kuthubutheen, 1987; Kuthubutheen and Nawawi, 1987, 1988a-b, 1990), Papua New Guinea and Solomon Islands (Tubaki, 1965; Matsushima, 1971a-b), Puerto Rico (Padgett 1976; Betancourt and Caballero 1983; Betancourt, et al., 1987; Betancourt and Justiniano, 1989), Taiwan (Matsushima, 1980), Thailand (Tubaki, et al., 1983) and Venezuela (Nilsson, 1962).

2.2. Aquatic Hyphomycetes:

Since the pioneering work of Ingold in the 1940s, a sizable number of novel genera and species of aquatic fungi have been reported from freshwater streams. Ingold's work of over three decades culminated with the publication of a pictorial guide entitled 'An

ilustrated guide to aquatic and water-borne hyphomycetes (Fungi Imperfecti) along with notes on their biology' (Ingold, 1975). Taxonomic keys and descriptions of these fungi were subsequently provided by Descals et al.(1995).

The conidia of freshwater fungi have very distinctive shapes: These may be branched, often with three or four arms (tri- or tetraradiate), worm-shaped (sigmoid) or in the form of a steep helix (helicoid) and appendaged (setulate). The conidia are usually large, many measuring 50-100 µm diam or more. The spores are produced in abundance and generally adapted to underwater germination, growth and trapping (Webster, 1959, 1987; Webster and Davey, 1984).

In their conidial state, aquatic fungi are classified in Hyphomycetes or Coelomycetes. An increasing number of anamorphs with their teleomorphs which belong to genera of Ascomycotina and Basidiomycotina, are being reported (Webster, 1992). Aquatic hyphomycetes are thus not a group of closely related organisms, but represent fungi which became adapted to aquatic habitats, both morphologically and physiologically. Their conidial phase were believed to have resulted from convergent evolution (Webster, 1987). The teleomorphs usually develop on twigs and branches previously submerged in streams and later become exposed as the stream water level falls. The ascospores and basidiospores of aquatic fungi show no unusual morphological adaptations. They are discharged into air and get disseminated long distances by air current (Dix and Webster, 1995).

Because of their characteristic conidial shape, aquatic hyphomycetes can be identified often from these spores alone, which is certainly not the case for most other groups of fungi. Further, the conidia readily get trapped in foam in streams, enabling easy

survey of distribution (Iqbal and Webster, 1973a). The important role which this group of fungi plays in conditioning and processing of leaf debris, making it palatable and nutritious to aquatic invertebrates is a feature of ecological interest (Dix and Webster, 1995).

So far, 328 species of freshwater hyphomycetes are known. New genera such as Canalisporium Nawawi and Kuthubutheen (1989a), Quadricladium Nawawi and Kuthubutheen (1989b), Crucella Marvanova and Suberkropp (1990), Nidulispora Nawawi and Kuthubutheen (1990a), Obeliospora Nawawi and Kuthubutheen (1990b), Candelosynnema Hyde and Seifert (1992), Isthmophragmospora Kuthubutheen and Nawawi (1992), Paracryptophiale Kuthubutheen and Nawawi (1994) and many more have been published during the recent past few years. Dix and Webster (1995) have concluded that there may be many more aquatic fungi awaiting isolation and identification.

Species of aquatic fungi that have worldwide distribution included Anguillospora crassa Ingold, Campylospora chaetocladia Ranzoni, Flagellospora penicilloides Ingold, Jaculispora submersa Hudson and Ingold, Lunulospora curvula Ingold, Tetrachaetum elegans Ingold, Tetracladium setigerum (Grove) Ingold, Tricladium angulatum Ingold, Triscelophorus monosporus Ingold and Varicosporium elodeae Kegel (Wood-Eggenschwiler and Barlocher, 1985). Some species appear to have been localised in a given climatic zone. Alatospora acuminata Ingold, Flagellospora curvula Ingold, Heliscella lugdunensis Sacc and Theny and Lemonniera aquatica De Wild are some of the dominant species in high North whereas Anguilospora aquatica S Nilsson, Brachiosphaera tropicalis Nawawi, Campylospora filicladia Nawawi, Ingoldiella hemata Shaw, Isthmotricladia gombakiensis Nawawi, Lunulospora cymbiformis Miura, Phalangispora constricta Nawawi & Webster, Pyramidospora casuarinae Nilsson,

Speiropsis pedatospora Tubaki, Tricladiomyces malaysianum (Nawawi) Nawawi, Triscelophorus acuminatus Nawawi are confined to tropics (Dix and Webster, 1995).

Traditionally, freshwater hyphomycetes were distinguished into two groups by their biological behaviour. These include 'strict aquatic' and 'aero-aquatic'. With the broadened concept of aquatics by Thomas (1996), two additional groups of freshwater hyphomycetes can be categorized, namely 'terrestrial aquatic' and 'submerged aquatic' hyphomycetes.

2.2.1. 'Ingoldian' Hyphomycetes:

Growing on submerged leaves and twigs, these fungi abound in fast flowing treelined streams and well-aerated rivers. They were relatively sparsely recovered on woody substrates (Ingold, 1975; Shearer and Webster 1991).

Two basic shapes of conidia, branched or sigmoid, have been recognised in them. In majority of the branched conidia, they are tetraradiate, that is, usually with four long arms diverging from a common point. Examples of this spore type are found in genera such as Alatopsora, Actinospora, Articulospora, Campylospora, Clavariopsis, Jaculispora, Lemonniera, Tetrachaetum, Tetracladium, Tricladium and Triscelophorus. Examples of branched conidia includes Dendrospora, Polycladium and Varicosporium. Amongst the Ingoldians, a number of species produce sigmoid conidia, i.e. long and worm-like usually with curvature in more than one plane. The sigmoid conidia are seen in genera such as Anguillospora, Flagellospora, Lunulospora and Mycocentrospora. Besides these three basic shapes, aquatic hyphomycetes with conidia in other shapes such as shrimp-like or fly-wheeled in Gyorffyella and spherical or ovoid in Margaritispora,

Dactyllela and Dimorphospora are known. It is significant to note that nearly all Ingoldian fungi have conidiophores and conidia that are hyaline and thin-walled (Ingold, 1975; Nawawi, 1985).

Several factors such as shape of conidia, mucilage secretion, appessorium formation on attachment to substrata, rapid colonization and sporulation once substrata become available, production of a wide range of extracellular enzymes, ability to grow and sporulate at temperatures down to 0°C and effective methods of transmission between unconnected water courses and ability to withstand drought, were attributed for the successful adaptation of aquatic hyphomycetes (Ingold, 1975; Read et al., 1991; Webster and Descals, 1981). The unique features of their conidial forms, two to several armed or sigmoid, have been shown to aid their dispersal (Iqbal and Webster, 1973a). Air bubbles generated in water can trap conidia and bring them to the surface. Trapping efficiency is said to be correlated with conidial shape and geometry. For example, tetraradiate conidia were trapped about three times as readily as sigmoid conidia and about 30 times as effectively as the ovoid forms (Ingold and Webster, 1973b).

2.2.2. 'Aero-aquatic' hyphomycetes:

Beverwijk (1951a-b; 1953; 1954) used the term 'aero-aquatic' for those hyphomycetes usually found in stagnant ponds, ditches or slow-running streams and are capable of vegetative growth on submerged leaves or woody substrates under semi-anaerobic conditions. In contrast to strict aquatics in which the whole cycle of conidium production, liberation and dispersal normally takes place below water, the aero-aquatics sporulate only when the substrate is exposed to air. The conidia, either tightly helicoid in

more than one plane or equipped with a special flotation device in the form of an intricate hyphal system, are mostly pigmented. Examples include genera such as *Arbuscula*, *Aegerita*, *Beverwykella*, *Cancellidium*, *Candelabrum*, *Clathrospherina*, *Clathrosporium*, *Cristulariella*, *Fusticeps*, *Helicodendron*, *Helicoon*, *Helicomyces*, *Mycoenterolobium*, *Nidulispora* and *Spirosphaera* (Abdullah and Webster, 1980; Carmichael et al., 1980; Fisher, 1977; Glen-Bott, 1951; Goh and Hyde, 1996; Goos et al., 1985; 1986; Tubaki, 1975; Webster and Davey, 1980).

These hyphomycetes with special floatation or air-trapping device were dispersed from one static water habitat to another by the topography, contour and environmental conditions of ponds or ditches and substrate-water interface (Fisher 1977; Subramanian, 1983; Webster and Descals, 1981).

2.2.3. 'Terrestrial aquatic' hyphomycetes:

Terrestrial aquatic hyphomycetes (Ando,1992) are represented by a number of conidial fungi isolated from rain drops associated with intact terrestrial plant parts, such as the leaf-surfaces or rainwater drained through intact tree trunks (Ando and Tubaki, 1984b). The genera described from such isolations include *Alatosessilispora*, *Arborispora*, *Curucispora*, *Microstella*, *Ordus*, *Tricladiella* and *Trifurcospora* (Ando, 1992; Ando and Kawamoto, 1986a, 1986b, Ando and Tubaki, 1984a; 1984b; 1984c; 1985; 1987). A major character of this group is that they produce staurosporous conidia similar in shape to those of the Ingoldian group, but lacking in conspicuous conidiophores (micronematous). The conidia are mostly subhyaline and thin-walled, though some dematiaceous species were also reported. Examples enclude *Ceratosporium cornutum*, *Tetraploa aristata* and

Tripospermum infalcatum. These fungi produce their conidia quickly in relation to their water sources (e.g. morning dew, mist and rain) of unpredictable frequency and duration. Another characteristic feature of these hyphomycetes is their staurosporous shape of conidia, which is adapted to hold water around the conidium, as long as possible. These characteristics made terrestrial-aquatic hyphomycetes better adapted to their unique environment (Ando, 1992).

2.2.4. 'Submerged-aquatic' hyphomycetes:

This group, first addressed by Ingold (1975) represents a heterogenous assemblage of fungi growing on submerged decaying plant materials. Most of the species are found on wood litter blocked by rocks in fast-flowing streams. These lignicolous or foliicolous hyphomycetes are nearly all dematiaceous and produce relatively thick-walled conidiophores or conidia. The conidiophores are macronematous, solitary or synnematous. The conidiogenous loci may be denticulate, cicatrized, tretic or phialidic. Although some species may sporulate under submerged conditions, a vast number sporulate when the substrates are no longer under water. Incubation of such woody substrates in moist chamber yields a great number species. Their conidia are capable of air dispersal or dispersed by some other mechanisms. These hyphomycetes may be regarded as 'facultative-aquatic', as compared to the aquatic Ingodian. The conidia of submergedaquatics are basically regular in shape, that is, ovoid, cylindrical, obclavate, pyriform or fusiform. However, branched conidia are common and may also be found in foam samples, for example, conidia of Caseresia sphagnnorum, Pleiochaete setosa and Tetraploa aristata (Kuthubutheen 1987a; 1987b; 1987c; 1987d; 1993; Kuthubutheen and Nawawi, 1987a; 1987b; 1988a; 1988b; 1988c; 1990; 1991a; 1991b; 1991c; 1991d; 1992; 1993; 1994a; 1994b; 1994c; Kuthubutheen et al. 1992; Nawawi & Kuthubutheen, 1988a; 1988b; 1989). Recently Goh and Hyde (1996a-d) and Hyde et al. (1996) reported several such genera, namely Bactrodesmium, Brachydesmiella, Brachysporiella, Campospoeidium, Cryptophialoidea, Dactylaria, Dendryphiosphaera, Cryptophiale, Canalisporium, Dictyochaeta, Exserticlava, Kionochaeta, Monotosporiella, Nawawia, Phaeoisaria, Sporidesmiella, Sporedesmium, Sporoshisma, Sporoshismopsis, Spadicoides, Trichocladium and Xylomyces. A few taxa such as Diplocladiella appendiculata, D. scalaroides, D. tricladioides, Setosynnema isthmosporum and Triscelosporium venucosum have been considered as examples of submerged-aquatic hyphomycetes (Hyde et al., 1996).

Although the majority of conidia seen in foam samples may be assumed to have an aquatic or stream side-origin some spora might have come from air (Ingold, 1975). Occassionally conidia of such well-known terrestrial hyphomycetes as *Alternaria*, *Beltrania*, *Cladosporium*, *Drechlera*, *Epicoccum* have been encountered in water.

Certain hyphomycetes have been found in aquatic habitats other than streams, lakes or ponds. Fungi such as *Geotrichum candidum*, *Fusarium aquaeductuum*, *Aspergillus* spp. and *Penicillium* spp. have been recorded worldwide in water treatment plants, especially in films of trickling or percolating filters, in sewage and in activated sludge (Cooke,1970; Jones, 1976; Subramanian, 1983).

2.3. Anamorph-Teleomorph connections of freshwater fungi:

Majority of freshwater hyphomycetes is known only in their anamorphic state.

Most of the known teleomorphs, however have been shown to be associated with ascomycetes (Abdullah et al., 1981; Webster 1992, 1993). Studies have shown that many of these connections are heterogeneous, i.e. the ascomycetous teleomorphs include taxa of diverse relationships (Webster 1993; Webster & Descals, 1979, 1981). For example, Flagellospora penicilloides producing sigmoid conidia and Heliscus lugduenensis with branched conidia, have teleomorphic connections with Nectria spp. (Pyrenomycetes). Anguillospora rosea and A. longissima have their teleomorphic connections with Orbilia sp. (Discomycetes) and Massarina sp. (Loculoascomycetes), respectively. Studies from the tropics demonstrated that Tricladium indicum Sati & Tiwari, isolated from South African river, has a teleomorphic connection with the ascomycete (Leotiales), Cudoniella indica Webster et al. (Webster et al.,1995) These discoveries of connections between hyphomycetes and ascomycetes or basidiomycetes provided further evidence of the artificial nature in the taxonomy of anamorphic genera (Webster, 1992; 1993).

2.4. Trapping of aquatic spora by air-bubbles:

Aquatic hyphomycetes form a highly distinctive, abundant and ubiquitous fungus flora on submerged decaying leaves of trees and shrubs of many kinds in well-aerated streams. The branched septate mycelium of the fungi ramifies the dead leaf tissue and the conidiophores project into the water and liberate the spores which are taken away by water currents. Spores are slightly denser than water and do not sink rapidly (Webster, 1959a). In fast flowing well-aerated streams, at barriers formed by twigs, branches and stones, bubbles are formed and accumulated as foam and scum. Spores carried by water currents and brought to the surface, are readily caught and retained by the foam present.

That foam is an effective trap for spores of aquatic hyphomycetes has been pointed out by Ingold (1975).

If foam from a well-aerated stream is collected and examined under a microscope a dense accumulation of spores can be seen (Descals et al. 1977). This is a very quick way of getting an idea of the hyphomycetous fungus flora of a stream (Nilsson, 1964). The increased trapping efficiency of air bubbles for the tetraradiate type of spore also suggests that foam samples may give a somewhat distorted view of the spore content of a stream (Iqbal and Webster, 1973a-b).

Foam samples can be preserved for subsequent examination by the addition of fixative or by smearing on microscope slides and staining with lactophenol cotton-blue. The aquatic hyphomycete spora of different river systems can thus be compared (Nilsson, 1964; Chauvet, 1991, 1992).

2.5. Aquatic fungi in culture

Aquatic hyphomycetes grow well in agar culture on a range of ordinary laboratory media. They appear to have no unusual nutritional requirements, many being able to utilize simple carbohydrates and some can degrade cellulose and other polysaccharides. They also utilize amino acids either as a Carbon or Nitrogen source and can immobilize dissolved nitrogen salts from stream water (Suberkropp and Klug, 1981). Single spore cultures of aquatic hyphomycetes can be established by transferring spores on the tip of a flamed sewing needle or by fine pipette, to malt extract agar. Bacterial growth is minimized by the addition of a mixture of the antibiotics Streptomycin and Penicillin. Bacteria-free hyphal tips can be transferred to fresh malt-extract agar and maintained as stock cultures. In pure

culture, sporulation normally occurs only when culture pieces are immersed in water. Spore production gets increased when culture pieces in water are agitated by compressed air or shaking (Webster and Towfik, 1972). These conditions simulate the turbulence of their normal habitats.

Aquatic hyphomycetes isolated from temperate waters are known to grow better at low to moderate temperature (around 15°C). Prolonged incubation of culture pieces in sterile water under diffuse light at low temperatures resulted in the development of teleomorphs (Webster, 1965).

2.6. Ecological studies:

Decomposition process of organic matter in stream ecosystem has been considered in 3 phases: leaching, microbial colonization and invertebrate feeding (Cummins, 1974). Leaching of soluble materials (carbohydrate, aminoacids and phenolic compounds) was rapid (Suberkropp et al. 1976) and 24-28 h immersion resulted in the loss of up to 25 % of original weight (Webster and Benfield, 1986). If the leaf has been shed in the normal way by abscission after maturity it would contain mycelia of several common terrestrial and phylloplane fungi such as species of *Aureobasidium, Cladosporium, Epicoccum, Alternaria*, etc. In the water, the leaf got rapidly colonized by the conidia of aquatic hyphomycetes which in the autumn reached sgnificantly high concentration of over 1000 spores/L. The original terrestrial leaf inhabitants persisted for a time but gradually replaced by aquatic fungi (Barlocher and Kendrick, 1974; Chamier et al. 1983). Within a few weeks, a mosaic of colonies of aquatic hyphomycetes got established. This early association made up of a relatively small number of species possibly resulted from

competitive interactions. It has been suggested that in the early stage of colonization, a leaf received an imprint from the stream spora that determined the dominant members of the fungal community throughout its decay (Barlocher and Schweizer, 1983). Fungi accounted for 63-95% of microbial biomass in decomposing submerged leaves of *Platanus, Quercus* and *Ulmus* (Findlay and Arsuffi, 1989).

2.6.1. Substrate specificity:

Leaves and twigs of riparian trees and shrubs and to lesser extent grass blades represented major substrates for aquatic hyphomycetes. Conifer needles were less intensively colonized due to the presence of thick waxy cuticles and phenolics (Barlocher and Oertli, 1978; Michaelides and Kendrick, 1978). Macrophytes, when presented, also supported species-poor aquatic hyphomycete assemblages (Kirby et al. 1990).

Differences in dominance patterns and frequencies on different types of leaf litter have been observed (Barlocher and Kendrick, 1974; Suberkropp and Klug, 1976; Chamier and Dixon, 1982; Shearer and Lane, 1983). Barlocher (1992) suggested that most aquatic hyphomycetes (at least those occurring on leaf litter) exploit a ruderal strategy, i.e., rapid colonization of available resources and production of propagules.

In some studies, species composition of conidia in stream water demonstrated little to moderate similarity with fungal species on leaf litter or wood (Sanders and Anderson, 1979; Chamier and Dixon, 1982; Shearer and Lane, 1983) indicating selective colonization. However, when more accurate comparison based on composition of conidia in stream water and distribution of conidia released during the breakdown of the substrate was done, the percentage similarity reached up to 66.8% (Barlocher, 1982).

Colonization by aquatic hyphomycetes started with attachment of conidia to a substrate and was affected by its physical characteristics and specific topographical stimuli (Harrison et al.1988; Read et al. 1992). Colonization also depended on size and shape of the substrate and features inherent to fungal species. A number of external factors such as stream temperature and availability of dissolved nutrients influenced colonization process (Gulis, 2001).

Aquatic fungi can be collected on a wide range of different plant materials. Certain species, however, showed resource preference. Preference is seen in a variety of ways, for example, by more frequent occurrence on particular leaf species, on standard areas of different leaf baits in packs in the same stream, or by different levels of spore output. Inability to colonize a given kind of substratum may result from the presence of barriers to infection, such as a thick cuticle and epidermis, or to other anatomical features such as well-developed sclerenchyma or other lignified tissues, or chemical factors such as nitrogen content of the leaf or the presence of inhibitors such as phenolics. Tetracladium marchalianum showed a higher colonization frequency and importance index on discs cut from leaf packs of Carya glabra than on discs of Quercus alba. Triscelophorus monosporus showed the opposite (Suberkropp, 1984). In comparisons between Fagus sylvatica and Alnus glutinosa, it has been shown that Tetracladium marchalianum is abundant on alder leaves, but absent from beech leaves which were dominated by Tricladium splendens (Bengtsson, 1983; Gonczol, 1989). Thomas, Chilvers and Norris (1992a) compared the frequency of occurrence of aquatic hyphomeetes on discs cut from randomly selected leaves of Eucalyptus viminalis and phyllodes of Acacia melanoxylon in an Australian stream. There was clear evidence of preference. Alatospora acuminata was recorded more frequently on Acacia, whilst *Tetrachaetum elegans* was more common on *Eucalyptus*.

Coniferous leaves were thought to be poor substrata for aquatic hyphomycetes (Ingold, 1966), but later studies showed that they do become colonized especially after prolonged submersion in a stream (Michaelides and Kendrick, 1978; Barlocher and Oertli, 1978; Barlocher et al., 1978).

Woody materials, varying in size from small twigs to large branches or even whole tree trunks, fall into streams and become exposed to colonization by the aquatic hyphomycete spores. The fungal community which develops on woody substrata is similar in part, to the leaf community, but is neverthless distinctive (Revay and Gonczol, 1990). Shearer (1992) has reviewed the role of woody debris in the life cycle of aquatic hyphomycetes. One of the most important features is their longer persistence in a stream, sometimes extending over several years. Another important property of woody materials is that they support the development of the teleomorphs of aquatic hyphomycetes, on which long-distance transport and sexual recombination may depend (Webster, 1992). Compared to the species known to be present in the streams, the number of species which fruit on wood is smaller, i.e., wood selects only fraction of the fungi available (Willoughby and Archer, 1973; Sanders and Anderson, 1979). Experimental studies based on submerged twig packs gave little evidence of host preference (Revay and Gonczol, 1990; Shearer and Webster, 1991).

Fungal colonization patterns on submerged leaves of eight plantation crops were followed over a period of one year in a stream in Western Ghat forests by Sridhar et al. (1992). In all, they recorded 27 fungal species. Coffee (*Coffea arabica* Linn.) and rubber

(Havea brasiliensis M.) leaves were colonized by 22 and 20 species respectively whereas only 4 species colonized jack (Artocarpus heterophyllus Lam.) leaves.

In another study in a stream on the foothills of Western Ghats, submerged leaf litter of natural vegetation and plantation crops was examined by Sridhar & Kaveriappa, (1988a-e). They observed that banyan leaves were colonized by 12 species of fungi, banana (*Musa paradisiaca* Linn.) and jack leaves by only 4 and 3 species. Their subsequent studies confirmed that banyan leaves are among the best substrata for recovering a maximum number of species (Sridhar and Kaveriappa, 1989).

There are a few reports of aquatic hyphomycetes on exotic plant litter such as Acacia, Casuarina, Eucalyptus, Pinus and Rubus (Nilsson, 1962; Cowling and Waid, 1963; Swart, 1986; Thomas et al. 1989). The study of submerged litter from Eucalyptus tereticornis Smith in an Eastern Ghat stream (Manoharachary and Galiah, 1987) and Pinus roxburghii Sarg. in a Himalayan stream (Sati et al. 1989) yielded 2 and 9 species of fungi, respectively. Eleven species were found on submerged litter of Gleichenia pectinata Presl., one of the common ferns in the Western Ghat region (Sridhar and Kaveriappa, 1988).

Aquatic hyphomycetes were also cultured on leaf petioles (Sridhar and Kaveriappa, 1983) and plant latex (Sridhar and Kaveriappa, 1987b). These substrata believed to have stimulated sporulation in pure cultures and helped in inducing the perfect state.

Several studies have been carried out on aquatic ascomycetes colonising submerged wood. Hyde (1994) observed the presence of a new aquatic ascomycete, *Astrosphaeriella aquatica*, on the submerged *Livistona* rachides. Tsui et al. (1999) during

a survey of freshwater fungi on submerged wood in streams of Hong Kong dicovered a new taxon belonging to *Massarina* (*M. proprietunicata*). Shearer (1972) has listed fungi from Patuxent, while Shearer and Bodman (1983) reported ascomycetes from submerged twigs from central Illinois. Revay and Gonczol (1990) have also reported on fungal colonization of twigs in streams in Hungary, and Shearer & Webster (1991) provided details of aquatic hyphomycete communities in the river Teign in Devon, UK. The fungi reported by Hyde and Goh (1999) were different from those reported from the earlier studies which were largely Ingoldian.

Substrate preference and interactions among the species of aquatic hyphomycetes in natural communities have been reproted (Willoughby and Minshall, 1975; Chamier and Dixon, 1982a-b; Bengtsson, 1983; Thomas et al., 1992). In an Australian upland stream, patterns of occurrence of species on different substrates were sufficiently different to predict the correct substrates from which the species came (Thomas et al. 1992). Charcosset and Gardes (1999) studied the intraspecific genitic diversity and substrate preference in *Tetrachaetum elegans*. These studies indicated that substrate preference plays an important role in the success of decomposition.

2.6.2. Succession of aquatic fungi:

The studies reported by Sanders and Anderson (1979) and Barlocher and Schweizer (1983) showed that there is a succession of fungi fruiting on submerged leaves. The detailed picture was dependent on the kind of leaf involved, the time (season) of submersion in the stream, which in turn was related to the range of temperature to which the submerged leaves were subjected, and the abundance and species composition of

spores available in the stream during the period of submergence. On leaves which are rapidly decomposed and consumed by invertebrates, such as those of alder, succession was more difficult to detect than on more persistent leaves such as those of oak (Chamier and Dixon, 1982a). Fungi which fruited early on leaf pack material of oak and hickory were *Flagellospora curvula*, *Lemonniera aquatica* and *Alatospora acuminata* (Suberkropp and Klug, 1976). Similar findings were reported by Gessner et al. (1993) from alder leaf packs submerged in autumn in the River Touyre in the French Pyrenees.

2.6.3. Spore concentration in stream:

Chamier and Dixon (1982a) have estimated that number of spores which can be produced per gram dry weight of leaf tissue are c.140,000 for *Alnus* and *Quercus*. Even higher values have been claimed by Barlocher (1982) who estimated that leaves of *Quercus* and *Larix* could produce 5-6 x 10⁶ conidia g⁻¹ with in a 2-day period. Suberkropp (1991) has estimated that *Lunulospora curvula* allocates 60-80% of its biomass to sporulation and *Anguillospora filiformis* 30-45 %. Such prolific production was said to be a ruderal trait, probably correlated with growth on relatively ephimeral substratum and low chance that an individual spore has of being trapped on a suitable underwater substratum in a rapidly moving water current. It was presumed that rapid and prolific reproduction is an adaptation to minimize the effects of predation by aquatic invertebrates which graze on leaves colonized by aquatic hyphomycetes (Dix and Webster, 1995).

In temperate and cool-temperate latitudes, streams bordered by deciduous trees showed enormous variations in seasonal abundance of conidia of aquatic hyphomycetes directly related to the addition of autumn-shed leaves. In areas where litter input was

spread over a longer period, the seasonal variations in abundance were less pronounced, e. g., in parts of Australia where litter fall in *Eucalyptus* forest may reach a maximum in the late summer to early autumn months, December-March (Thomas et al., 1989; Thomas et al., 1992). In a stream in which needles of spruce (*Picea abies*), which were shed evenly throughout the year, dominated the terrrestrial input, there was no clear peak in conidial concentration during the year (Barlocher and Rossett, 1981).

Spore concentrations between 10³ and 10⁴ I¹ have been reported by Iqbal and Webster (1973b) for a lowland stream in Devon, England, during October-December, and these autumnal values have been matched or exceeded by other workers elsewhere. In the following summer, as the leaf litter was decomposed, consumed by invertebrates, fragmented and swept down stream, the spore concentration fell too low to be detected by filtration. The suggested explanation that these different periods of spore production are controlled by temperature was explored by Moran and Davey (1976) who found differences in temperature optima for growth and sporulation of *Lunulospora curvula*, having higher optima.

2.6.4. Effect of temperature on growth and sporulation of aquatic fungi:

Temperature affects different phases of the life cycle of the fungus, germination, infection, mycelial growth and sporulation (Koske and Duncan, 1974). Experiments designed to separate the temperature and litter availability have been reported by Suberkropp (1984), who immersed leaf packs of oak (*Quercus*) and hickory (*Careya*) for 4-week periods at different points in the stream characterized by different temperature ranges. Laboratory experiments showed that species more abundant in cooler months have

different temperature characteristics from species from the summer assemblage. For example, *Lunulospora curvula, Heliscus tentaculus* and *Flagellospora penicilloides* grew only at temperatures above 50°C, whilst *Flagellospoa curvula* and *Lemonniera aquatica* were able to grow at 1°C (Suberkropp, 1984; Suberkropp and Klug, 1987).

A relatively constant winter temperature of 0.1°C occurs in the Njakajokk stream in the Swedish Arctic (68°N) from the end of October to May. For some of this period, the stream is covered by with ice. Three species dominated the spore output during the year: *Flagellospora curvula*, *Lemonniera aquatica* and *Alatospora acuminata*, all of which are members of the cold water assemblage of Suberkropp (1984).

Temperature is also related to latitudinal and altitudinal distribution. A group of tropical and subtropical species has been recognized (Nilsson, 1964; Webster and Descals, 1981; Sridhar et al., 1992). Nawawi has listed 25 species which are tropical or subtropical in distribution, including *Brachiosphaea tropicalis, Campylospora chaetocladia, Heliscus tentaculus, Flagellospora penicilloides, Lunulopsora curvula, Triscelophorus acuminatus, T. monosporus* and and *Ingoldiella hemata*. All these species occur in Malaysia but some also occur in temperate climates (Dix and Wester, 1995).

2.6.5. Effect of pH and water chemistry on fungal growth and sporulation:

Chauvet (1991) has surveyed the distribution of spores of aquatic hyphomycetes at 27 stations in south-western France based on foam samples collected at intervals throughout the year. The altitude of the stations varied between 9 and 935 m, with a pH range of 5-8.5 and a water temperature range of 2-20°C. Correspondence analysis was used to examine the relationship between distribution patterns and abiotic variables such

as altitude, pH, temperature and season. A group of 5 species associated with lowland streams having high pH and high temperature was identified: *Campylospora chaetocladia, Campylospora* sp., *Heliscus tentaculus, Lunulospora curvula* and *Tricelophorus monosporus*. Two species, *Clavatospora longibrachiata* and *Tetrachaetum elegans*, appeared characteristic of acidic water (pH<6), low altitude and autumn months. Where the gathering grounds for river systems are in mountainous or upland areas, as the streams descend into lowland and the tributaries merge to form larger rivers, there are associated changes in the instream riparian vegetation and chemical and physical characters of the water (Hynes, 1970a-b; Chamier, 1992). These include changes in pH, conductivity, water hardness, dissolved organic matter, flow characteristics, and amount and quality of trapped litter and sediment. Barlocher (1992) has described variations in physical, chemical and biological parameters along river systems, all of which affecting community structure and activity of aquatic fungi.

Shearer & Webster (1985a-c) studied variation in spore concentration in the colonization of alder leaf packs submerged at different points along the River Teign in Devon, England. The upper reaches arise in moorland where the water is acidic (pH 5.4-6.0), with hardly any riparian trees and lower sites characterized by higher pH (7.0-7.2), boarded by various trees, whose detached leaves and branches are trapped amongst boulders. The community of aquatic hyphomycetes at the upland site was distinctly different from the down stream communities and less than 20% of all species were found on leaves at all the sites. This was attributed to local production at the upstream site, and rapid extinction (i.e. loss due to trapping or dilution) as the spores were carried downstream.

Thomas et al. (1991a-b) discussed factors related to changing spore concentrations in a stream and provided a mathematical model to explain the dynamics of spore populations in a body of moving water. The reports on spore concentrations suggested that in a given river system, species number may be correlated with rising pH. When data from streams in Europe and Canada were correlated, however, this simple relationship became less clear (Barlocher and Rosset, 1981; Wood-Eggenschwiler and Barlocher, 1983; Barlocher, 1987). There are also features of water chemistry such as water hardness, alkalinity and the solubility of Ca and Al which are pH-dependent (Chamier, 1992). At low pH (<5.5), ionized forms of Al³⁺ dominated in solution, and some of these were toxic. In acid streams, submerged litter significantly accumulated Al more rapidly than in circumneutral streams (Chamier et al., 1989).

As indicated before, a series of chemical changes followed the immersion of tree leaves in streams, beginning with rapid loss of soluble materials by leaching. Microbial colonization was accompanied by increase in protein content as demonstrated first by Kaushik and Hynes (1971) and later confirmed by many (Barlocher, 1985; Chauvet, 1987). The increase in protein largely resulted from fungal growth. There were also increases in other compounds, e. g., phosphate (Meyer, 1980). Physical changes also occurred, notably the softening of leaf tissues as a result of macerating activity of fungal enzymes (Suberkropp and Klug, 1980, 1981; Suberkropp et al. 1983; Chamier, 1985). These changes in the chemical and physical properties of submerged leaf litter have been termed 'conditioning'. There is much evidence that conditioned leaves are consumed in preference to non-conditioned by leaf-shredding invertebrates such as the larvae of aquatic insects and *Gammarus* spp. Patches of leaf material colonized by fungi are preferred to

uncolonized areas (Arsuffi and Suberkerkropp, 1992).

Aquatic invertebrates thus exert some control over the populations of aquatic hyphomycetes. In experiments in which leaf packs enclosed in fine- or coarse-mesh bags and immersed in streams, Barlocher (1980) showed that leaves in coarse-mesh bags, permitting the entry of invertebrates which fed on the leaves, had a lower cumulative species count of invertebrates. The enrichment of the protein and probably the lipid content of leaf material associated with increase in fungal biomass rendered conditioned leaf material more nutritious, whilst the macerating activity brought about by fungal enzymes made it easier to ingest. For these reasons, animals fed on conditioned leaf material grew more rapidly than those fed on non-conditioned leaves, and expended less energy in foraging for food (Barlocher and Kendrick, 1976, 1981).

2.7. Occurrence of aquatic fungi in terrestrial system

The majority of studies on ecology of freshwater aquatic hyphomycetes have been carried out in the aquatic habitat (Iqbal and Webster, 1973; Suzuki and Nimura, 1962; Triska, 1970) and the continual isolation and description of new species from stream foam and submerged leaves testifies to their abundance in this environment. Aquatic hyphomycetes have however occassionally been reported from non-aquatic environments (Gourley, 1968; Makela, 1972; Park, 1974a; Scourfield 1940; Waid, 1954; Latch and Mc. Kenzie, 1977).

Bandoni (1972) was the first to report these organisms in any number in terrestrial litter in Canada. Later, Koske and Duncan, (1974) recorded them in number in sites away from watercourse. This suggested that these fungi may be more terrestrial than had

previously been supposed and this was supported by Thakur (1977) who found that survival could occur for 2-5 months in air-dried leaves at room temperature. In a year-long study of litter beside an English river, however, Webster (1977) could find only very small numbers of detached conidia and no conidiophores on leaves which were collected more than a few metres above the flood level (Park,1974b). To date, there are 32 species recorded from terrestrial situations.

Sanders and Webster (1978) was carried out a study to determine for how long some of the common species from streams can survive when transported to leaf litter in a terrestrial environment as might occur by flood or animal agencies. They have shown that *V. elodeae* and *Articulospora tetracldia* can extend from colonized leaf discs sandwiched between sterile oak leaves buried in wood land litter to infect the oak leaves. Bandoni (1981) opined that the conidia of some fungi commonly condidered to be aquatic hyphomycetes may be found in terrestrial habitats. Park (1982b) has shown that *Varicosporium elodeae*, found in the litter layers above soil, also colonizes leaves or even filter paper buried in soil.

2.8. Aquatic fungi as root endophytes

A number of studies have dealt with the seasonal dynamics of freshwater hyphomycetes by examining the composition of conidia in freshwater. Iqbal (1992) and Iqbal and Webster (1973, 1977) have shown that at certain times of the year the number of conidia fell to undetectably low levels. Shearer and Webster (1985a-c) reported up to 30,000 conidia/L in the River Teign in November. Much lower numbers of conidia per liter have been reported by Iqbal and Webster (1977) for streams in Dartmoor, England,

by Shearer and Lane (1983) for the Sangamon River, Illionois, and by Iqbal (1992) for streams in Khanspur, Pakistan. In streams of Dartmoor, no conidia were detected during summer months. Low frequency of occurrence or absence of these fungi coincided with the scarcity of leaf debris in stream waters (Iqbal and Webster, 1977). However, some conidia were always present in stream water (Iqbal, 1993), and these served as inoculum when substratum fell into the stream again (Webster and Descals, 1981). It has been reported that freshwater hyphomycetes can also grow on woody litter that has fallen into the stream (Shearer, 1992). This more enduring material may provide an important substratum for survival when leaf litter has become scarce. Submerged living roots of riparian trees provide another perennial source of survival (Iqbal, et al. 1995).

So far, 20 species of freshwater fungi have been reported as root endophytes of submerged roots of riparian tree species. The occurrence of freshwater hyphomycetes of roots has added a new dimension to the ecology of freshwater hyphomycetes. The role of these root-associated freshwater hyphomycetes in plant nutrition, root absorption efficiency, and senescence or susceptibility to disease remains to be investigated.

Fisher and Petrini (1998) reported *Campylospora parvila* Kuzula and *Tricladium splendens* Ingold as endophytes in the root bark and xylem of *Alnus glutinosa* (L) Gaertner. Twelve species of freshwater hyphomyctes were reported as endophytes of the submerged root bark of *A. glutinosa*, three of these species were also present in the xylem (Fisher et al. 1991). Similarly Sridhar and Barlocher (1992a-b) found 16 species of hyphomycetes colonizing the living aquatic roots of riparian trees.

2.9. Fungal dynamics in stream and its importance in litter breakdown

Small streams and rivers would be effectively shielded from direct sunlight by riparian trees. Adjacent terrestrial vegetation would supply the water with substantial amount of dead organic material, mostly in the form of leaves, branches and twigs. What the habitat lacked in primary productivity was thus compensated for by this allochthonous organic matter. These factors favoured development of a stream community largely dependent on allochthonous organic material for its food supply (Hynes, 1970a).

Experiments have confirmed that imported organic material, mostly leaves, is of great significance in the food chain of stream ecosystem (Hynes, 1970b; Kaushik and Hynes, 1971). The contribution of allochthonous organic material to the total energy budget of stream communities has been estimated to range between 50% and 99% (Cummins et al., 1966; Fisher and Likens, 1972; Nelson and Scott, 1962; Teal, 1957).

Only a small fraction of the energy represented by leaf material is directly exploited by animals. Usually more than 60% and often as much as 90% of leaf litter eaten by woodland detritus feeders is returned to the soil in the faecal pellets (Hudson, 1972) and similar conditions predicted in aquatic environments. Hargrave (1970) estimated that *Hyalella azteca*, a freshwater amphipod assimilated only 5% of the material which it ingested when feeding on elm leaves.

For access to the remaining energy, the animal community depended on the intervention of micro-organisms, with their ability to degrade cellulose and lignin which they exploit in increasing their own biomass. Microbial proteins, fats, carbohydrates etc. were then readily digested by many animals. (Fredeen, 1964; Hargrave, 1970; Ivlev, 1945; Newell, 1965). Several authors have recognized and documented that fungi as well as

bacteria are important intermediaries in the detritus food chain of terrestrial environments (Alexander, 1967; Burges and Raw, 1967; Doeksen and Drift, 1963; Graff and Satchell, 1966). But most biologists have assumed that in streams this niche is exhausively or predominantly occupied by bacteria and the potential significance of detritus-decomposing fungi was ignored until Kaushik and Hynes (1968, 1971) and Triska (1970) investigated the degradation of autumn shed leaves in streams. In both cases, fungi were usually found to be more active and successful than bacteria, at least in the early stages of decay. Several detritus-feeders clearly preferred to eat leaves which bore a rich fungal growth rather than freshly fallen or sterile leaves.

Similar to forest floors, many running water systems receive (Webster et al. 1990) and retain (Jones and Smock, 1991, Snaddon et al. 1992) large amount of leaf litter from riparian vegetation, the breakdown of which therefore constitutes a key in the metabolism of such streams (Cummins, 1988). Despite considerable researches on breakdown of leaves (Webster and Benfield, 1986; Boulton and Boon, 1991) and the realization of the critical importance of aquatic microfungi in this process, information on the quantity of fungal biomass developing in decomposing leaves was scarce. Barlocher & Kendrick (1981) and Suberkropp (1992b), in their reviews, published accounts of ATP consumption and hyphal length measurements (Frankland et al. 1990; Newell, 1992). These estimates ranged from 0.004 to 12% of detrital mass. Importance of microfungi in the trophic structure of streams is however currently debated (Suberkropp, 1992b).

While in terrestrial systems the decomposition of leaf litter has been studied in more detail than in streams, current knowledge of the fungal mass developing during decay is incomplete. Recent advances in methodology, in particular the development of

the ergosterol assay, now fecilitate the determination of litter-associated fungal mass with greater accuracy (Newell, 1992, 1994).

Leaves of different plant species are known to lose mass at different rates (Webster & Benfield, 1986). While the absolute breakdown rate of a given leaf species varied with environmental conditions, the sequence of breakdown of a set of species was unpredictable (Petersen and Cummins, 1974; Chauvet et al. 1993). Consequently in woodland soils, attempts have been made to establish relationships between breakdown rates of various types of litter and the chemical nature of these materials. The conclusion of these studies was that, concentration of nutrients such as lignins or a combination of these constituents are critical in determining litter decomposability (Melillo et al. 1984; Taylor et al. 1989, 1991; Geng et al. 1993; Schaefer et al. 1985). Additionally phenolic compounds, particularly tannins have been suggested as major determinants of breakdown rate (Nicolai, 1988; Stout, 1989), physical leaf properties may also be important (Gallardo and Merino, 1993).

In streams, a few notable studies have been compared with regard to the breakdown of more than three types of leaves (Petersen and Cummins, 1974; Hill et al. 1992). None of these attempted to relate leaf litter quality and decomposition rate in a formal manner (Melillo et al. 1983). It is not known if any study attempting to pinpoint the missing organismic link between the chemical nature of leaf litter and the rate at which it is broken down, has been made.

It is now clearer that fungi control leaf breakdown in streams through their growth and production of extracellular enzymes. Fungal activities, in turn, are regulated both by internal characteristics of leaf tissue such as lignin and tannin content (Gessner and Chauvet, 1994) and external environmental factors such as temperature and nutrient concentrations in water (Suberkropp and Chauvet, 1995). Only a few studies has been carried out to see the effects of dissolved nutrients on fungal activities in streams (Chamier, 1992; Suberkropp, 1994), but Mulholland et al. (1984, 1985) provided evidence that the microorganisms colonizing leaf detritus obtains their inorganic nutrition from water. Further more, water chemistry appears to have affected rates of aquatic hyphomycete sporulation (Barlocher, 1982; Rosset et al. 1982; Suberkropp, 1991) and higher rates of breakdown.

One of the major problems in the study of fungi involved in litter decomposition and nutrient cycling has been the lack of proper methodology to measure growth rates and consequently, fungal productivity in the environment. Newell and Fallon (1991) described a method for making such measurements and applied it to the examination of the fungi associated with decomposing grasses and sedges in saline and freshwater marsh habitats. This method is modeled after techniques for determining bacterial growth rates (Moriarity, 1990; Kirchman, 1993) and involved determining the rates of incorporation of radio-labelled acetate into ergosterol, specifically found in higher fungi (Newell, 1992).

Where the rivers flow into the sea, the organisms must be able to tolerate vatiations in salinity. Observations on the distribution of aquatic hyphomycetes on wood blocks showed that they occur, even if they were not abundant in estuaries (Jones and Oliver, 1964). Byre and Jones (1975a-b) have studied spore germination, vegetative growth and sporulation in two species, *Heliscus lugdunensis* and *Tetracladium setigerum*, in sea water at various dilutions and on agar to which sea water was added at varying temperatures.

The abundance of aquatic fungi in rapidly flowing, well-aerated streams suggested that they might be adapted physiologically to water with a high concentration of dissolved O₂. Experimental studies indicated that different species of aquatic hyphomycetes vary in their capacity to survive in anaerobic conditions. The mycelium of *Articulospora tetracladia* failed to survive for 3 months, whilst a small portion of colonies of *Tricladium splendens* and *Anguillospora rosea* survived for 1 year (Field and Webster, 1983). Also, the aquatic hyphomycetes tested were more sensitive to H₂S than species of aero-aquatic fungi which are common in stagnant habitats (Field and Webster, 1985).

2.10. The influence of nutrients and water on aquatic fungi

Decomposition is an important process in ecosystem functioning and recycling of nutrients. In streams and rivers, the major energy source is detritus originating either in the rivers themselves or in the riparian zone (Rodrigues et al.1997). In streams, nutrients dissolved in the water particularly nitrogen (N) and phosphorus (P) are important regulators of biotic processes. Number of studies have examined the effects of dissolved nutrients on rates of primary production and accumulation of fungal biomass, and it is evident that autochthonous production and grazing food chains in streams were affected by such bottom-up controls (Bott, 1983; Grimm and Fisher, 1986; Pringle, 1987; Meyer et al. 1988; Hart and Robinson, 1990; Peterson et al. 1993; Rosemond et al. 1993).

Woodland streams are typically heterotrophic: ie., more energy comes from allochthonous sources than from autochthonous production (Fisher and Likens, 1973; Minshall et al. 1983). A significant portion of the allochthonous energy base enters streams in the form of leaf litter that is used by decomposer microorganisms and

detritivores. Currently, the effects of dissolved nutrients on detrital food chains, ie., on rates of leaf breakdown and the activities of decomposer microorganisms, are not clearly understood.

In laboratory studies, amendments of N, with or without P, have generally been found to stimulate microbial activity, microbial biomass accumulation, and rates of leaf breakdown (Hynes and Kaushik, 1969; Howarth and Fisher, 1976; Fairchild et al. 1984). In streams, the effects of nutrients on leaf breakdown and the microbial communities associated with leaves were variable. The addition of P was shown to stimulate breakdown (Elwood et al. 1981) whereas N enrichment had no effect on leaf breakdown rate (Triska and Sedell, 1976; Newbold et al. 1983). Evidence for stimulation of leaf breakdown by N has been obtained in a comparison of two streams that differed in nitrate concentrations (Meyer and Johnson, 1983). Laboratory studies in contrast to field studies were carried out in closed systems in which nutrient amendments made at the beginning of the study and not replenished as they were utilised by microorganisms. Howarth & Fisher (1976) found that nitrate concentrations of the water in their laboratory set up had decreased to low levels during the study.

Fungi have been demonstrated to be the microbial group responsible for the initial breakdown of leaves in streams (Suberkropp and Klug, 1976; Findlay and Arsuffi, 1989; Gessner and Chauvet, 1994) and for modifying leaf detritus into a more suitable food source of invertebrate detritivores (Barlocher, 1985; Suberkropp, 1992). The activities of stream fungi have been shown to be regulated by substrate quality, that is, by internal chemical factors such as the lignin content of leaves (Gessner and Chauvet, 1994). The effects of external chemical factors such as dissolved nutrients on the activities of stream

fungi were not well understood (Chamier, 1992). Studies, however, have indicated that levels of microbial biomass and rates of sporulation by aquatic hyphomycetes were greater on leaves in hard-water streams (high pH and alkalinity) with elevated concentrations of N and P than on leaves from hard-water streams with lower concentrations of nutrients or from softwater streams (Barlocher, 1982; Rosset et al. 1982; Suberkropp, 1991). These observations suggest that fungal activity can also be controlled by external chemical factors in the water flowing across leaf surfaces.

2.11. Diversity of aquatic fungi in polluted waters:

Aquatic fungi dominate leaf decomposition in streams and play an important role as intermediaries between decaying leaves and leaf-eating invertebrates (Barlocher, 1992; Suberkropp, 1992). Almost exclusively, they have been collected from clean, well-aerated waters (Barlocher, 1992). Due to growing human demand for food, living space, such habitats are becoming increasingly rare. These raised the question of how aquatic fungal communities will be affected by the various types of human interference.

In a study of an organically polluted river in India, Raviraja et al. (1998) found a loss of over 80% of the number of species of aquatic hyphomyctes, but no measurable decline in some ecological functions that are normally associated with this fungal group. Thus decay rates and dynamics of P and C on the leaves did not differ significantly from those studied in a nearby unpolluted river. Krauss et al. (1998) investigated another stream polluted by heavy metals.

In laboratory experiments, low concentrations of Cd, Cu and Zn have been shown to inhibit growth and reproduction of aquatic hypphomycetes (Abel and Barlocher, 1984,

1988) and response of fungi by synthesizing specific stress peptides (Miersch et al. 1997). Little is known, however to which extent aquatic hyphomycetes are able to persist in chronically polluted habitats. In a recent study, Sridhar et al. (2000) observed presence of 17-30 species of aquatic hyphomycetes from six sites of a copper mining site of Central Germany, by collecting foam and naturally occurring plant litter and by following fungal colonization of *Alnus glutinosa* leaf disks.

Pollution by heavy metals has increased over the last few decades through mining, industrial emissions, garbage disposal and by agricultural fertilizers (Merian 1991). Most of these are toxic at low concentrations. Cd, Cu and Zn inhibited both growth and reproduction of aquatic hyphomycetes (Abel and Bärlocher 1984, Miersch et al. 1997). Many fungi, including some aquatic hyphomycetes, have evolved ability to tolerate heavy metals by synthesizing S-rich compounds and peptides derived from glutathione (phytochelatines) (Gadd 1993, Miersch et al. 1997, 2001; Gadd and Sayer 2000).

2.12. Enzymatic analysis of leaf decomposition in freshwater

Aquatic fungi possess an array of enzymes, including pectinases, capable of macerating leaf tissues (Chamier and Dixon, 1982a-b; Chamier, 1985; Zemek et al. 1985; Abdullah and Taj-Aladeen, 1989). Stimulation of pectic enzyme activity of aquatic hyphomycetes by Ca²⁺ may in part explain the more rapid maceration of leaf tissues recorded in streams above pH 6.5 (Chamier and Dixon, 1983).

In the early stages of decomposition, microbial activity was responsible for the breakdown of the complex polysaccharides of plant cell walls. As a consequence, leaves became softer and their N content increased (Kaushik and Hynes, 1968; Suberkropp and

Arsuffi, 1984; Graca et al. 1993). There is now a general concensus that fungi are the dominant agents involved in the degradation of leaf tissue. In the final stages of decomposition, however, bacteria may become more important (Suberkropp and Klug, 1976). Detritivores preferentially feed on microbially colonized leaves (Suberkropp, 1992; Graca, 1993) promoting leaf fragmentation and thereby accelerating decomposition.

Aquatic hyphomycetes dominate the fungal community of submerged leaves (Barlocher and Kendrick, 1974). Their ability to depolymerize complex polysaccharides has been confirmed (Suberkropp and Klug, 1980; Chamier and Dixon, 1982a, b; Suberkropp et al., 1983; Chandrashekhar and Kaveriappa, 1988). In spite of the apparent importance of aquatic hyphomycetes in freshwaters, terrestrial fungi have been isolated from submerged leaf substrates (Kaushik and Hynes, 1968; Godfrey, 1983; Rossi et al., 1983). Little is known of submerged plant detritus. It has been shown that leaves entering streams were already colonized by terrestrial fungi that remained alive under submerged conditions, but were rapidly overgrown by the aquatic hyphomycetes. The advantage of aquatics over terrestrial fungi in freshwater has been related to their more effective dispersal due to the tetraradiate and sigmoid spores, high sporulation rate stimulated by water turbulance (Webster & Descals, 1981) and growth at lower temperatures (Koske and Duncan, 1974). Graca and Ferreira (1995) showed that at both low (4°C) and high (20°C) temperatures, aquatic hyphomycetes had a higher capability to degrade leaf tissue than terrestrial fungi isolated from streams.

Rodrogues and Grace (1997) examined the role of terrestrial fungi in lotic environments by comparing the enzymatic capabilities of selected aquatic and terrestrial fungi isolated from streams. They showed that aquatic hyphomycetes and terrestrial fungi

differed in their ability to macerate leaves in the water and leaf softening was highly correlated with xylanase activity. Previously published results attributed the role of pectinases in leaf softening. Pectic polysaccharides are the major component of primary cell walls (about 34% w/w) and are the most immediately available polymers in non-lignified plant tissues. Their degradation exposed the other components (the hemicelluloses and celluloses, 24% and 19% respectively) to enzymatic attack (Chamier & Dixon, 1982b). The attack of pectolytic enzymes caused the release of epidermal and parenchymal cells into the water column (Suberkropp and Klug, 1980). Jenkins and Suberkropp (1995) showed that pectin lyase was closely associated with the softening and maceration of leaf detritus, confirming that pectin degradation is a key process in the initial stage of breakdown.

According to Chamier and Dixon (1982b), while pectin lyase and esterase have minimum activity at high pH (>8.0), polygalacturonases were most active at lower pH values (5.0-6.0). Other studies confirmed the production of this enzyme (Zemek et al. 1985; Suberkropp et al. 1983; Suberkropp and Arsuffi, 1984). Suberkropp et al. (1993) reported high activity for *Heliscus lugdunensis* and medium activity for *Lemonniera aquatica*. Suberkropp and Klug (1980) and Zemek et al. (1985) reported Xylanase production in aquatic hyphomycetes. (1985). The low activity of *H. lugdunensis* towards pectin, cellulose and xylans was consistent with its low ability to macerate alder and chestnut leaves (Graca and Ferriera, 1995). Xylanase production in terrestrial fungi, *Cladosporium cladosporoides* and *Epicoccum nigrum* in aquatic conditions had already been reported (Godfrey, 1983).

Cellulose decomposition by aquatic hyphomycetes has been investigated by various

methods. There are reports suggesting that cellulose activity is widespread in aquatic hyphomycetes (Suberkropp and Klug, 1980; Singh, 1982; Suberkropp et al. 1983; Zemek et al. 1985; Chandrashekar and Kaveriappa, 1988; Abdullah and Taj-Aldeen, 1989). For terrestrial species, Godfrey (1983) and Park (1982) reported the cellulase activity in cultures of C. cladosporoides and E. nigrum isolated from aquatic habitats.

Fungi are able to degrade lignin and use it as a C source (Abdullah and Taj-Aldeen, 1989; Zemek et al. 1985; Gessner and Chauvet, 1994). The oxidation of tannic acid was considered as an indicator of polyphenol oxidase activity, an enzyme presumably involved in the degradation of lignin (Davidson et al. 1938; Abdullah and Taj-Aldeen, 1989). Zemek et al. (1985) noticed lignin degradation in all aquatic hyphomycetes tested. Fisher et al. (1983) found that only 6 out of 16 species of aquatics were able to produce phenol oxidising enzymes. Rodrigues and Graca (1997) detected lipolytic activity of 9 aquatic hyphomycetes and 7 terrestrial fungi sampled from submerged leaf leaves in the same stream.

2.13. Aquatic fungi in the Indian sub-continent

The Indian sub-continent encompasses a wide range of climatic conditions. Tropical conditions prevail in majority of places; some regions have subtropical/ temperate climates. Extensive forested areas with numerous permanent and temporary streams are present in the Himalayan range, the Western Ghats and the Eastern Ghats. These include alpine/subalpine, moist deciduous, evergreen and tropical rain forests. The other major biomes represented are semi-arid zones in the southern and northern plains, Thar desert in the northwest and vast estuaries on both west- and east-coasts.

The Western Ghats consist of a wide range of mountains, about 1600 km long. They run parallel to the west coast from the mouth of the River Tapti (21°N) to the tip of South India (8°N). They extend through the states of Maharashtra, Karnataka, Kerala and Tamil Nadu. This range is rich in tropical evergreen and moist deciduous forests. Average elevation is 1220m and average yearly rainfall 250-450 cm. The mean annual temperature varied between 28 and 30°C and relative humidity about 80%. Several fast flowing perennial streams and rivers originated from the midst of the Ghats and flew east into the Bay of Bengal or west into the Arabian sea (Pascal, 1987).

2.13.1. Researches on Aquatic hyphomycetes

A total of 32 water bodies were surveyed for aquatic hyphomycetes. In addition to streams and rivers, thermal spring, estuary, freshwater ponds and lake were examined. Sidhar et al. (1992) noted the presence of 78 species belonging to 45 genera from aquatic habitats of the Indian subcontinent. Of these, 20 have been recorded only once; 24 found in ten or more independent surveys. Eight speices have been found only in foam and two only in water samples, another six occurred predominantly in foam.

Incubation of leaf litter or examination of naturally occurring foam (Ingold, 1942), foam induction (Sridhar and Kaveriappa, 1984a) and water filtration (Iqbal and Webster, 1973) were common methods employed to characterize aquatic hyphomycete communities of streams. Most records from India were based on the examination of foam samples and these yielded fairly a high number of species. In some studies, leaf litter of natural vegetation, plantation crops and fern were also scanned (Mer and Khulbe 1981; Sridhar 1984; Sridhar and Kaveriappa, 1986, 1988a-c, 1989e, Manoharachary and Galiah 1987).

In a comparitive study of seeds, dried leaf litter and newspaper pieces, the seeds of *Tagetus erecta* Linn. (Mer and Khulbe 1981) and leaf midribs of banyan (*Ficus bengalensis* Linn.) (Sridhar and Kaveriappa, 1989 b) were found to be the best baits.

The aquatic hyphomycete community of Indian waters showed a marked seasonal periodicity (Sridhar et al., 1992) The main factors that appeared to be correlated with these fluctuations were water temperature, timing of litter fall and precipitation, changes between wet and dry seasons and the types of substrata. To analyze the factors influencing aquatic aquatic hyphomycetes communities, the studies in three different geographical areas viz., the Central Himalayas, the Western Ghats and the Eastern Ghats were considered. In the central Himalayan region, (Mer and Sati, 1989) the highest number of fungal species was recorded during monsoon season (September-November) and during spring (March-April). The situation in the Western Ghats seemed to be similar as far as maxima of conidial numbers and types were concerned. They were generally highest during the later part of the monsoon (August-November), when the water temperature was 17-22°C. Leaves were shed from March through June; rainfall began in June/July and continued through November (Manoharachary and Galiah 1987; Sridhar and Kaveriappa, 1989a, d).

In coastal streams, the number of species and conidia transported in water column was greater from July to December than from January to June (Sridhar and Kaveriappa, 1984b). The number of species per sample varied between 6-20 in coastal streams compared to 24-36 in the ghat streams (Sridhar and Kaveriappa, 1984, 1989, Chandrashekar et al. 1990). The lower numbers in coastal streams might be due to higher water temperatures (23-30°C), a longer dry season (mid December to end May) and

comparatively sparce riparian vegetation (Sridhar and Kaveriappa, 1984, 1989a-d). A dense canopy of forests, plantation crops and higher altitude were reasoned for the coolness of forest floor and the streams.

2.13.2. Distribution of aquatic fungi in India

Systematic studies on aquatic fungi of India began about 45 years ago, with report of *Articulospora tetracladia* Ingold by Patil & Rao (1972) from the forests of Western Ghats. Ingold & Webster (1973) reported four species, namely *Condylospora spumigena*, *Ingoldiella hamata*, *Lunulospora curvula* and *Triscelophorus monosporus* from a mountain stream near Madras, Tamil Nadu. Subsequently a large number of aquatic fungi were reported from several parts of India including the Himalayan region (Uttar Pradesh), the Western Ghats (Maharashtra, Karnataka, Kerala and Tamilnadu), the Eastern Ghats (Tamil Nadu and Andhra Pradesh) and Central India (Madhya Pradesh).

Sridhar et al. (1992) gave an exhaustive and comprehensive review of hitherto work carried out on aquatic fungi in India along with a complete list of so far reported freshwater mycota from the sub-continent. A list of 65 species belonging to 40 genera of fungi so far reported from the streams of Western Ghats in southern India alone is given below (Pinto and Bhat, 1992; Sreekala and Bhat, 1998; Sridhar et al. 1992; Subramanian and Bhat, 1981).

2.13.3: Aquatic fungi from Goa:

So far, there are no major published work on aquatic fungi from the streams of Goa, except a brief reference of conidia of three hyphomycetes reported from Dudsagar

Falls near Anmod (Subramanian and Bhat, 1981. In a preliminary floristic survey, Pinto and Bhat (1992) recorded spora of 17 species of aquatic hyphomycetes from a few streams in and around Goa. Sreekala and Bhat (1998) documented 29 species of aquatic fungi from four streams of Goa. However, no significant ecological or taxonomic studies were done on aquatic fungi of Goa.

Table 2.1. Aquatic fungi from the streams and rivers of Western Ghat forests (Pinto and Bhat, 1992; Sreekala and Bhat, 1998; Sridhar et al. 1992; Subramanian and Bhat, 1981)

Fungi	MH	GOA	KA	KL	TN
Actinospora megalospora Ingold		F	F	F	-
Alatospora acuminata Ingold	-	F	F,L	F	-
Anguillospora crassa Ingold	L	F	F,L	-	-
A. longissima Ingold	L	F	F,L	F,L	F
Articulospora angulata Tubaki	L	-	-	-	-
A. inflata Ingold	T -	-	F	-	-
A. tetracladia	-	-	F,L	F	F
Brachiosphaera tropicalis Nawawi & Webster	-	-	F	-	-
Camposporium pellucidum	-	F	-	-	•
Camposporium sp.	—	F	-	-	-
Campylospora chaetocladia Ranzoni	-	F	F,L	F	-
C. filicladia Nawawi	-	F	F,L	F	F
C. parvula Kuzuha	1 -	-	F	-	•
Clavariana aquatica Nawawi	1 -	-	F,L	-	-
Clavariopsis aquatica de Wildeman	_	_	F,L	F	F
C. ozlanii Nawawi	-	_	F,L	-	-
C. brachycladia Tubaki	-	-	F	-	
C. bulbosa Anastasiou	-	_	L	-	-
Clavatospora tentacula (Umphlett) Nilsson	-	_	F,L	F,L	-
Condylospora spumigena Nawawi	_	-	F,L	F	F
Dactylella oviparasitica Stirling & Mankau	_	-	F	_	-
D. submersa (Ingold)Nilsson	_	-	F,L	_	-
Dendrospora erecta Ingold	-	F	L	_	-
D. juncicola Iqbal	-	_	F,L	_	
Dendrosporomyces prolifer Nawawi, Webster &	-	-	L	-	-
Davey					
Diplocladiella scalaroides Arnaud	-	F	-	-	
Erynia conica (Nawakowski) Rem & Henn	-	-	F	-	-
E. rhizospora (Thaxter) Rem.& Henn	-	-	F	-	•
Flabellocladia tetracladia (Nawawi) Nawawi	-	_	F	F,L	
Flabellospora crassa Alasoadura	-	-	F,L	-	•
F. multiradiata Nawawi	-	F	F	F,L	-
F. verticillata Alasoudura	-	F	F,L	F,L	F
Flagellospora curvula Ingold	-	F	F,L	F,L	-

F. penicilloides Ingold	1 - 1	_	F,L	F	
Heliscella stellata (Ingold & Cox) Marvanova	+ 1	-	F,L	-	<u>-</u>
Ingoldiella hamata Shaw			F,L		F
Isthmotricladia gombakiensis Nawawi	 -		F,L	-	T.
I. Iaeenis Mathsushima	 	- F	F,L	-	
		Г		F	F
Laridospora appendiculata (Anastasiou) Nawawi	- F	-	F,L	F	
Lateriramulosa uni-inflata Matsushima			F,L	F	F
Lemonniera aquatica de Wildeman	L		- -	-	-
Lunulospora curvula Ingold		<u> </u>	F,L	F,L	L
L. cymbiformis Miura	-	-	F,L	F,L	-
Mycoleptodiscus sp.	1 - 1	F		-	
Nawawia filiformis (Nawawia) Marvanova	-	F	F,L	F	F
Phalangispora constricta Nawawi & Webster	-	F	F,L	F,L	-
P. nawawii Kuthubutheen		_	F	-	-
Pyramidospora casuarinae Nilsson		-	F,L		-
P. constricta Singh	-	-	F,L	-	-
Speiropsis hyalospora Subramanian & Lodha		-	F,L	F	-
S. irregularis Petersen		-	F	-	
S. pedatospora Tubaki	F	F	F,L	-	•
Tetracladium marchalianum de Wildeman	F	•	F,L	-	-
T. setigerum (Grove) Ingold	F,L	F	F	F	F
Tricladiomyces malaysianum Nawawi	-	-	L	-	-
Tricladiospora brunnea Nawawi &Kuthubutheen		-	F,L	_	-
Tricladium angulatum Ingold	F	-	F,L	-	-
T. fuscum Nawawi	-	-	F	-	-
T. splendens Ingold		-	-	-	-
Tripospermum camelopardus Ingold et al.	-	-	-	-	F
T. myrti (Lind.) Hughes	-	F	F,L	F	F
Triscelophorus acuminatus Nawawi	1 -	_	F,L	F,L	-
T. konajensis Sridhar &Kaveriappa	-	_	F,L		_
T. monosporus Ingold	F	F	F,L	F,L	F
Tumularia aquatica (Ingold) Descals &Marvanova	_	-	F	-	-
Varicosporium elodeae Kegel		-	F,L	_	-
					
Total number of species reported from each State	13	21	58	25	15

F-Foam; L -Litter; MH - Maharastra; GOA-Goa; KA-Karnataka; KL-Kerala; TN-Tamil Nadu

CHAPTER 3: MATERIALS AND METHODS

This investigation on the diversity, ecology and biology of aquatic fungi occurring in the streams and rivers of forests of Western Ghats in Goa was carried out for a period of two and a half years, from January 1999 to June 2001. In this Chapter, the materials used and methods followed during the study are described.

3.1. Sampling sites:

(A) For studies on seasonal occurrence of fungi:

For study of aquatic fungi occurring in different seasons, sampling was done at regular intervals in one stream each in three wildlife sanctuaries, Bondla, Cotigao and Mollem. Assuming that tree canopy may play a decisive role on the ecology of aquatic fungi by providing organic matter input, only those streams in the sanctuaries with dense riparian tree cover were chosen for the study (Plate.1.1). Besides abundance and frequency of occurrence, diversity of aquatic fungi was also studied from these sites. The sampling sites are described below.

Stream 1: A seasonal moderately fast flowing stream originated at a slightly high altitude in the ghats and flown down through Bondla wildlife sanctuary (120 MSL), in Ponda Taluka, was considered as 'stream 1'. The collecting site was about 2 km before the entry gate to the sanctuary and 60 km north-west of Goa University campus. The stream bed at the site was rocky. Banks of the stream on either sides were lined mainly by cane, Calamus thwaitesii Becc. and Dendrocalamus strictus (Roxb.) Hook and bamboo, Bambusa arundinaceae (Retz.) Willd, besides tree species such as Adina cordifolia (Roxb.) Hook, Bauhinia tomentosa Linn., Dillenia indica Linn., Grewia hirsuta Vahl.,



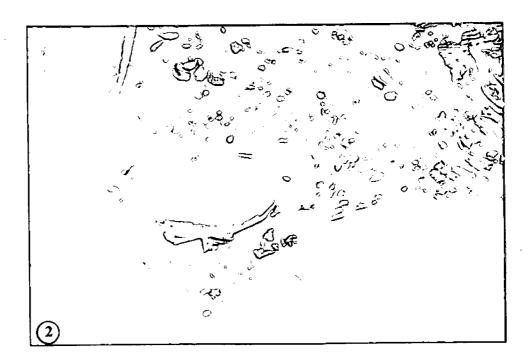


Plate. 1

Hydnocarpus laurifolia (Dennst.) Sleumer, Indigofera dalzelii Cooke, Stephania japonica (Thunb.) Miers. and Terminalia tomentosa Wt. & Arn.

Stream 2: A fast flowing seasonal stream originated at Anmod Ghat and flown down into Bhagvan Mahavir wildlife sanctuary (230 MSL) was chosen as 'stream 2'. The collection site was located about 4 km west of Mollem in Sanguem Taluka, and 65 km west of Goa University campus. The stream bed had soft soil at the collecting site. Even though a variety of tree species was present in the sanctuary, riparian vegetation along the stream mainly composed of *Hopea ponga* (Dennst.) Mabb, the roots of which extended into flowing waters along the sides of the stream. The other dominant tree species in the catchment region included evergreen types such as *Careya arborea* Roxb., *Cyclea peltata* (Lamk.) Hook, *Dillenia indica* Linn., *Flacaurtia montana* Grah., *Lophopetalum wightianum* Arn., *Osbekia truncata* Don ex Wt. & Arn., *Saraca asoca* (Roxb.) De Wilde and *Tinospora cordifolia* Miers.

Stream 3: A moderately fast flowing seasonal stream, originated above in the ghat and flown down near 'Tree-top' view point of Cotigao wildlife sanctuary (280 MSL) in Canacona Taluka, was considered as 'stream 3' for the study. The stream had lateritic soft soil in the bottom and good run off in monsoon season but gradually dried up during late post-monsoon and summer months. The site was about 75 km south-east of Goa University campus. The catchment area was covered by dense vegetation of semi-evergreen and evergreen tree species and some of the dominant riverine trees found in the area included *Careya arborea* Roxb., *Calycopteris floribunda* (Roxb.) Lamk., *Dillenia indica* Linn., *Grewia hirsuta* Vahl., *Kandelia candel* (Linn.) Druce., *Lagerstroemia lanceolata* Wall. ex Wt. & Arn., *Terminalia paniculata* Roth. and *Xylia xylocarpa* Taub.

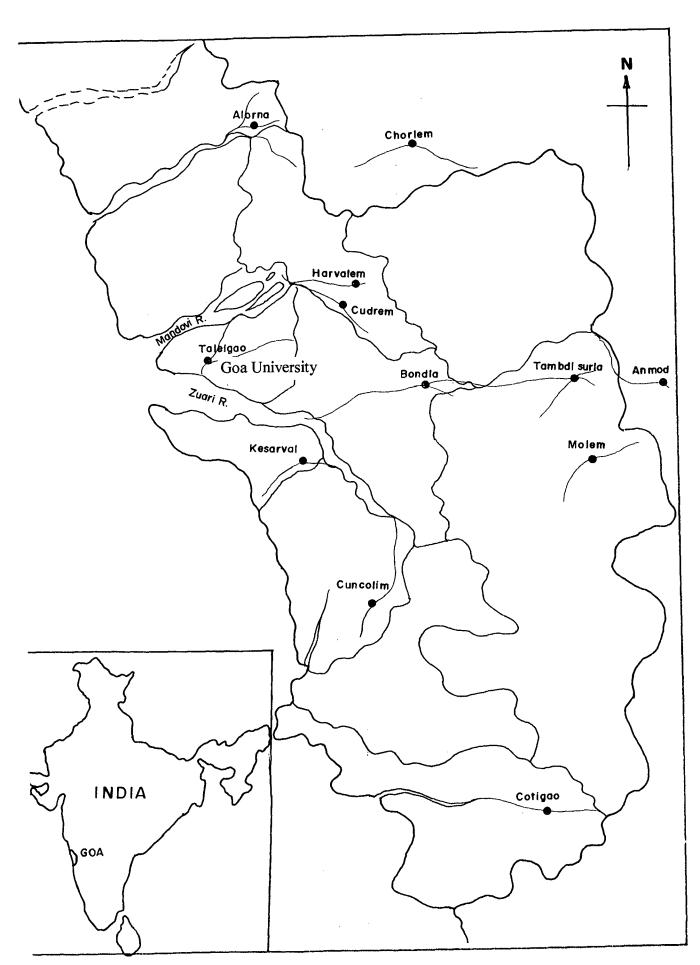


Fig.3.1. Map of Goa showing the sampling sites

B. For studies on diversity and taxonomy of fungi:

For floristic studies of aquatic fungi, samples - accumulated surface foam and submerged leaves, were collected at different sites from several seasonal streams and perennial rivers in the forests of Western Ghats in Goa region. Besides the three streams described above, other collection sites included - Alorna, Anmod Ghat, Chorlem Ghat, Cudnem, Cuncholim, Harvalem, Kesarval, Taleigao Plateau (Goa University campus) and Tamdisurla (Table 3.1; Fig. 3.1). These are briefly described below.

Table 3.1: Sampling sites for diversity study:

Site	Locality and type	Altitude	Distance from	*Dominant tree species.
ļ	of stream	(MSL)	GU Campus	,
j			(in km)	
1	Alorna:	180	60	Acacia pennata, Terminalia tomentosa
	seasonal stream			and Zyziphus glaberrima
2	Anmod Ghat:	420	70	Artocarpus heterophyllus, Dillenina
ļ	perennial stream			indica and Terminalia paniculata
3	Chorlem Ghat:	360	75	Ensete superbum, Ficus benghalensis,
}	seasonal stream			Grewia hirsuta and Tectona grandis
4.	Cudnem:	240	65	Acacia pennata, Alstonia scholaris and
	seasonal stream			Glochidion hohenakeri and ferns, grasses.
5	Cuncolim:	170	40	Ficus benghalensis, mangifera indica
	seasonal stream			and several grasses
6	Harvalem:	230	60	Anacardium occidentale, Grewia hirsuta,
	perennial stream			Havea brasiliensis and Mangifera indica
7	Kesarval spring:	120	20	Areca catecheu, Musa paradisica and
ļ	perennial stream			Cocus nucifera
8	Taleigao:	100	-	Eriocaulon cuspidatum, Cynodon
	seasonal stream			dactylon and Digitaria bicornis
9	Tambdisurla:	200	75	Ficus benghalensis, Dillenina indica and
	perennial stream			Terminalia tomentosa.

^{*}Presence of a tree species more than 20 % in number at the collecting site is recorded as 'dominant'.

1. <u>Alorna</u>: The collection site, a monsoon seasonal stream at Alorna (180 MSL) in Pernem Taluka, was about 60 km north-east of Goa University Campus. Dominant

riparian tree species of the region included Acacia pennata (Linn.) Willd., Erinocarpus nimmonii Garh., Melastoma malabathricum Linn., Terminalia tomentosa Wt. & Arn. and Zyzipus glaberrima Sant.

- 2. <u>Anmod Ghat:</u> A perennial stream, originated upland and flown down through the slopes of Anmod ghats (420 MSL), was located about 70 km from the University Campus. Major tree species found in this region included *Artocarpus heterophyllus* Lam., Celastrus paniculata Willd., Dillenina indica Linn., Naravelia zeylanica Linn. and Terminalia paniculata Roth.
- 3. <u>Chorlem Ghat</u>: A monsoon seasonal stream originated upland in the ghat and flown down at Chorlem (360 MSL) near Anjuna dam in Bicholim Taluka, the site was about 75 km north-east of Goa University Campus. Dominenat plant species in the catchment area included *Calycopteris floribunda* (Roxb.) Lamk., *Centratherum courtallense* (Wt.) Benth., *Ensete superbum* (Roxb.) Cheesman., *Grewia hirsuta* Vahl., *Ficus benghalensis* Linn., *Polyalthia cerasoides* (Roxb.) Bedd. and *Tectona grandis* L.
- 4. <u>Cudnem</u>: An open-cast iron-ore mining belt in Bicholim Taluka with a sizable coverage of cashew and mango plantations. Abandoned mines had several spring water reservoirs of varied sizes and from these a few small seasonal creeks originated. About 5 M cubic meters of annual discharge has been recorded from these ponds. The creeks were fast flowing during monsoon but became weak in summer months. Plants such as *Acacia pennata* (Linn.) Willd., *Alstonia scholaris* (Linn.) R. Br., *Anacardium oxidentale* L., *Carissa congesta* Wt., *Cassia tora* Linn., *Glochidion hohenakeri* Bedd and *Mangifera indica* L. and a few ferns and grasses were present in the area.

[Note: Besides foam and litter, soil and water samples were collected from this site to study the effect of mining rejects on growth and sporulation of aquatic fungi].

- 5. <u>Cuncolim</u>: The stream was a slow flowing seasonal creek at 'Seven Temple Hill' in Cuncolim (170 MSL), in Salcete Taluka, about 40 km south of the University Campus. Upstream region was lateritic and not many tree species were found here. Besides a few species of grasses, *Bombax ceiba* Linn., *Ficus benghalensis* Linn. and *Ixora arborea* Roxb, were found at the site.
- 6. <u>Harvalem</u>: The stream was seasonal and with a sharp waterfall during monsoon months into which rejects were let out from an adjoining iron ore mine at Harvalem (230 MSL) in Bicholim Taluka, 60 km away from the University Campus. The stream-bed was mostly rocky and banks were barren. Major plant representatives included *Anacardium occidentale* L., *Grewia hirsuta* Vahl., *Havea brasiliensis* M. and *Mangifera indica* L.
- 7. <u>Kesarval spring</u>: A freshwater stream at Kesarval in Tiswadi Taluka, was located about 20 km south-west of Goa University Campus. The stream bed was a thick lateritic sheet and on both sides had *Areca catecheu* L. and *Cocus nucifera* L., besides *Caryota urens* L., *Crotalaria retsa* (Roxb.), *Naregamia alata* Wt. & Arn. and *Memycylon umbellatum* Burm.
- 8. <u>Taleigao</u>: A seasonal stream at Taliegao Plateau, located behind the main administrative building of the University. The rainwater accumulated on the plateau flown down in the form of a narrow stream during monsoon. In the initial 500 m, the catchment area is flat and water flowed slowly and thereafter the stream formed a sharp waterfall. Sampling was done at this point. The plants present in the cathment area above the waterfall included *Carissa congesta* Wt., *Cynodon dactylon* (Linn.) Pers., *Digitaria bicornis* Loud., *Drosera indica* Linn., *Eragrostis ciliaris* (Linn.) R.Br., *Eriocaulon*

cuspidatum Dalz.., Glyophocloa acuminata Clayton., Ischaemum angustifolium Hack., Rotala indica (Wild.) Koehne. and Utricularia reticulata Smith. besides a few genera of grasses and ferns.

9. <u>Tambdisurla</u>: Tambdisurla (200 MSL) in Sanguem Taluka, about 75 km away from the University Campus, is a dense evergreen forest. Samples were collected from a fast flowing perennial stream. The banks of the stream were lined by grasses and several trees species such as *Bauhinia racemosa* Lamk., *Ficus benghalensis* Linn., *Saraca asoca* (Roxb.) Dc. Wilde., *Sesbania grandiflora* (Linn.) Pers. and *Terminalia tomentosa* Wt. & Arn.

3. 2. Sampling season and intervals:

During monsoon, from June to September every year, the windward western slopes of the Western Ghats receive a total rainfall of 250-350 cm. The mean annual temperature varies between 22-36°C, the minimum seldom falling below 18°C. Humidity ranges between 60-90%. All along the Western Ghats, in monsoon the streams are usually gorgeous with flowing water. During post-monsoon, the streams either have little flowing water or mostly at many places exhibit pools of stagnant water bodies. In the summer months, the streams are practically dry and without water, except those perennial ones where flow of water is very slow.

While raining, fallen leaves from the trees lining the stream and river banks and adjoining forests get washed into the streams. The leaf litter in the stream either remains submerged or gets parked against rock crevices, fallen logs or any obstacles.

(i) For studies on occurrence and quantitative analysis of aquatic fungi, sampling was done during every 'monsoon' (June-September), 'post-monsoon' (October-January) and 'summer' (February- May) in 1999 and 2001. The dates on which collections were made from different sites are given in Table 3.2.

Table 3. 2: Collection of samples for quantitative analysis:

Place of collection	Monsoon (June-September)	Post-Monsoon (October-January)	Summer (February-May)
Bondla	9-8-1999	25-10-1999	15-2-2000
Cotigao	19-8-1999	23-12-1999	11-4-2000
Mollem	18-9-1999	29-12-1999	17-5-2000
Bondla	12-8-2000	21-10-2000	14-2-2001
Cotigao	24-8-2000	17-12-2000	20-4-2001
Mollem	20-9-2000	25-12-2000	11-5-2001

(ii) For studies on diversity and taxonomy of freshwater fungi, the samples were randomly gathered several times but mostly in monsoon and post-monsoon seasons of 1999 and 2000. The collections sites and dates are given in Table 3.3.

Table 3. 3: Collection of samples for diversity study:

Place of collection	1999	2000
Alorna	23 Sept.	20 July.
Anmod Ghat	16 July, 25 Nov.	30 Sept.
Bondla	9 Aug, 25 Oct.	12 Aug, 21 Oct.
Chorlem Ghat	2 July, 20 June	5 Sept., 4 Oct.
Cotigao	19 Aug, 23 Dec.	24 Aug, 17 Dec.
Cudnem	5 Oct	4 Oct, 11 Nov.
Cuncolim	27 Sept	23 Nov.
Harvalem	23 June	28 June
Kesarval spring	9 Oct., 2 Nov.	10 Aug., 7 June.
Mollem	18 Sept., 29 Dec.	20 Sept., 25 Dec.
Taleigao Plateau	2 Aug., 12 Oct.	8 June, 10 Sept
Tambdisurla	15 Nov.	8 July

3. 3. Field gear:

The materials used for collection of aquatic samples (Plate.2.1) in the field and transportation to the laboratory included the following:

Fresh polythene bags, clean plastic vials of varied sizes, portable ice-pack container with ice blocks, FAA fixative [mixture of 40% Formaldehyde (5 ml), Glacial Acetic Acid (5 ml) and 70% Ethyl alcohol (90 ml)], laboratory thermometer, pH paper, amber-coloured bottles for collection of water samples, glass jars with wide mouth, microslides and Petri plates with malt extract agar (MEA) medium.

3. 4. Culture medium:

In this study, malt extract agar (MEA) medium was used for isolation, study and maintanance of fungi. The medium was prepared by dissolving malt extract (5 g) and psi agar-agar (20 g) in distilled water (1 L) and sterlizing in an autoclave at 15. for 15 min. A mixture of antibiotics consisting of bacitracin (0.02 g), neomycin (0.02 g), penicillin G (0.02 g), streptomycin (0.02g) and terramycin (0.04 g) dissolved in 10 ml of distilled water was added to 1 L of MEA isolation medium. The medium for maintenance of isolated cultures in test tubes consisted of 2% MEA without the antibiotics.

3. 5. Samples for isolation of fungi:

The freshwater foam entrapped with propagules of aquatic fungi such as hyphal fragments, fructification, mitospores, ascospores, basidiospores, etc., fungi-colonized submerged leaves, twigs and live or dead roots from freshwater streams or rivers constituted one of the 'samples' used in the present study. Besides the spores of aquatic





Plate. 2

fungi, small-sized particles such as dead insects, pollen, phyto- and zooplankton, etc, get trapped in the foam or water bubbles accumulated in rock crevices or any wooden logs in running waters (Plate.1.2). Fresh foam was white whereas more than 2 day old foam cakes were yellow to brown in colour. The other samples included submerged leaves, twigs and roots on which these fungi often colonise and sporulate.

The types of 'samples' collected and subjected to various fungal recovery processes are outlined below.

3.5.1. Foam:

(a) Collection for floristic study:

Spores of Deuteromycotina, Ascomycotina and Basidiomycotina get trapped in foam of freshwater stream. Besides, the foam also contains bacteria, algae, insects, pollen and other microscopic particles. If a drop of foam is examined under a light microscope, spores and hyphal fragments of a variety of aquatic fungi can be seen.

- (i) <u>Fixed foam</u>: Foam accumulated on the surface of stream water was gathered by gently and repeatedly scooping a wide-mouth glass jar or glass petri-plate lid over the foam (Plate 2.2). The collected foam was fixed by adding a few drops of FAA mixture into the jar. The foam bubbles break into a cream-coloured or slightly turbid liquid at the bottom of the container. The sample was maintained in 10 ml size screw-capped vials. The vials were appropriately labeled in the field indicating the sample number, location and date of collection.
- (ii) <u>Dried foam</u>: A drop of fresh foam was directly placed and spread over a clean slide and air-dried at the collection site. The slides were appropriately labeled indicating the

sample number, location and date of collection and brought to the laboratory by arranging them vertically and in rows in a slide box. These slides were observed under a microscope by placing a drop of lactophenol-cotton blue mountant over the foam dried area.

Examination of FAA-fixed or air-dried foam under a microscope revealed the floristics of aquatic fungi of the catchment region (Plate 5.1, 5.2., 5.3). The slides examined were deposited as specimens at the Herbarium of Botany Department, Goa University (GUBH).

(b) Collection for isolation and cultural study:

Pre-sterilized petri plates containing 10 ml of MEA medium with antibiotics were carried to the collection site aseptically in an icepack container. Fresh foam from the stream was scooped out (Plate.2.2) and directly poured onto the MEA plates (Plate 3.1). The plates were horizontally rotated so as to have uniform spread of particles present in the foam. Excess foam was decanted and edges of the plates were sealed with parafilm and brought to the laboratory in the icepack container.

Maintenance of poured plates in low temperature during transport delayed germination of spores of aquatic fungi present in the foam.

(c) <u>Isolation of fungi from foam</u>:

The foam-poured plates were scanned under a stereoscope with incident light and germinating spores were picked up individually with the help of a sterile needle and aseptically transferred to fresh 2% MEA plates marked on the lower side into nine equal segments and appropriately labelled. The spores thus transferred to and recorded in MEA plates were allowed to grow for 24 h at 20°C. The hypha emerging out from germinating

spore was aseptically transferred to MEA slants. The culture tubes were labelled and deposited in the Goa University Fungal Culture Collection (GUFCC).

3.5.2. Submerged plant parts:

(a) Collection of submerged leaf and roots:

Leaves of trees lining the stream and rivers on senescence fall into water. The fallen leaves act as substrate for growth and colonisation of aquatic fungi (Ingold, 1975). The submerged and partially to fully decayed leaf litter and twigs thus form excellent source for isolation of aquatic fungi. Similarly, riparian tree roots were also considered as substrates of harbouring aquatic fungi (Fisher, 1993). The submerged leaves were hand-picked, thoroughly washed in water and placed in clean polythene bags. These were transported to the laboratory in an ice-pack container.

(b) Isolation and quantification of fungi from submerged leaves in the laboratory:

(i) Through aerated water: Aquatic fungi sporulate under water, when conditions are favourable (Webster & Dix, 1995). A slightly modified 'water filtration method' of Iqbal & Webster (1973) was followed. Decayed and well-skeletonised 2-3 leaves were washed thoroughly in deionized tap water and placed in large specimen jars containing 500 ml sterile distilled water. The water was aerated continuously using a fish-tank aerator for 3-7 days (Plate, 3.2). The aerated water was filtered through a millipore filter (8 μm pore size) and the spores got collected on the filter were counted under a stereoscope. Foam accumulated on the surface was also examined under an inverted microscope for spores of aquatic fungi.

Plate:3

- 1. Foam sample
- a) collected in glass petriplate.
- b) poured onto MEA plates.
- 2. Leaf litter kept for incubation in sterile distilled water in glass jar connected to a fish-tank aerator.
- 3. Leaf litter kept for still water incubation.
- 4. Twigs kept for moist chamber incubation.
- 5. Root bits kept in MEA plates to see the appearance of endophytic fungi.

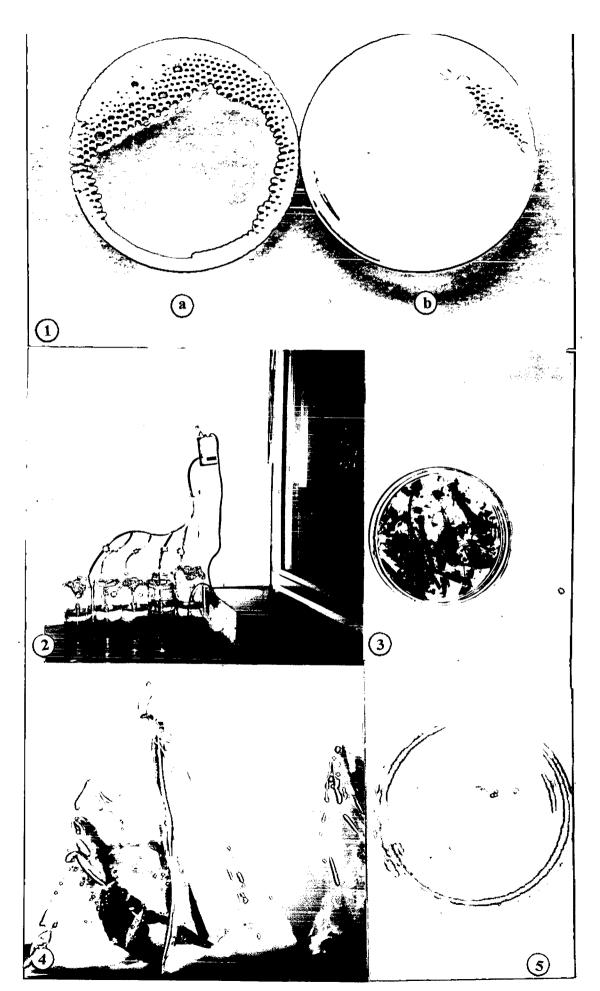


Plate. 3

(ii) <u>Through still water and moist chamber</u>: The leaf samples were kept in petriplates containing sterile distilled water and incubated for 3-7 days (Plate, 3.3). The leaf samples were examined under inverted microscope.

The submerged twigs collected from the sites were washed in sterile distilled water and incubated in polythene bags lined with sterile moist cotton for about 3-7 days (Plate, 3.4). The twigs were observed under stereo-microscope with incident light. Both these set up yeilded number of fungi.

(c) <u>Isolation of fungi from live roots:</u>

Riparian trees extend their roots into water and several aquatic fungi are believed to be living as root endophytes. For isolation of these fungi, three catagories of root pieces, namely primary, seconday and tertiary, of 7 plant species, Alstonia scolaris, Calamus thwaitesii, Dellinia indica, Glaberus sp., Glochidion hohenackeri, Hopea ponga and Saraca asoca, were considered. The three types of roots were the following:

- i) The primary roots were those extended from the main stem,
- ii) the secondary roots generally arose from the primary roots and
- iii) the tertiary ones, thin root hairs, grew from secondary roots.

Using sharp knife, submerged roots of varied length and thickness were cut below the water surface into pieces of 6-10 cm and transported to the laboratory in fresh polythene bags in an ice-pack container. The root pieces were thoroughly washed in tap water and de-ionized water to remove adhered debris and soil particles. These were further surface-sterilised first in 70% ethanol (1 min.), second in 4% sodium hypochlorite (3 min.) and third in 70% ethanol (30 sec) (Petrini, 1986).

The surface-sterilized roots were thoroughly rinsed in sterile distilled water for 5

m and cut into 3-5 mm long bits using a sterile razor blade. Nine to 12 root bits were placed equidistant in parallel rows in antibiotic incorporated MEA plates (Plate, 3.5). The plates were incubated at 20°C. The fungal colonies emerged out from cut edges of the root bits and extended into the agar medium were aseptically transferred into MEA slants without antibiotics for storage and further study.

Each such isolated distinct fungal colony was considered as a 'morphotype'. The isolates were grouped into 'sporulating' and 'nonsporulating' forms.

(d) Observation and recognition of diagnostic features of fungi:

Fungal colonies become visible on segment-marked petridishes from 3rd day of incubation. A small portion of the colony was aseptically transferred to MEA slants for preservation and further study.

The isolates were examined at periodical intervals for growth and sporulation. Semipermanant slides were prepared using lactophenol mountant (Hawksworth, 1974). Based on criteria recommended by Hawksworth et al. (1995) in the Eigth Edition of 'Ainsworth & Bisby's Dictionary of The Fungi', cultural characters of the fungi such as colony morphology, colour and size and microscopic features such as conidiomata, conidiophores, conidia or ascocarp, asci and ascospores were recorded in a specially prepared comprehensive 'Fungus data-sheet' (Table3.4). Only those features relevant with the fungus or isolate under consideration were retained in the sheet. Illustrations of the fungi were made using a camera-lucida drawing tube attached to a binocular research microscope (Olympus Make). Photomicrographs were taken using an automatic camera fitted to a bright-field research microscope (Nikon Make).

Table 3.4: 'DATA-BASE FOR AQUATIC FUNGI COLLECTED, ISOLATED AND DOCUMENTED FROM FORESTS OF WESTERN GHATS':

1. Name of Fungus Taxon:	: Common name (if any):
2.Collection details: Coll	ection No;Coll. by
	; Season (Weather) Rainy /Dry /Windy /Foggy /
	(Site); (taluka); (Dist); (State); (Altitude)
	be specified in the dotted or continuous line/space given at the end of each item)
	shwater: ; Lotic: (Rivulet/Stream/River); Lentic: (Pond/Lake/Dam)
Marine: : Mang	rove:; Extraterrestrial: (Above ground /Tree-hole/Bird nest/)
	Decidous /Moist decidous /Mixed deciduous /Shola /Riverine Forest
	nt /Grassland /Scrub /Dry /Jungle /Plantation /Epiphytic /
	lack /Mudflat /Laterite /Granite /Humus /Fertilized /Nonfertilized /pH
	esh leaf /Twig /Log /Root /Fruit /Flower /Seed /Grain /Latex /Gum /Fiber;
	bit /Monkey /Deer /Antelope /Elephant /Bison /Bird /Lizard /Frog /
	Hair /Feather /Paper /Cloth /Lipstick /Contact Lens /Glass /Leather
	Plastic /Metal /Paint(Type) /Rubber /Cement /Jute /
	(Range recorded at the sample collecting site)
	of material/substrate); Size; Status/Condition(dry/wet/fixed)
	to Lab: (In paper bag /polythene bag /Bottle /Culture Plate;
40 3 6 4 1 CT 1 1	(Direct /Moist Chamber Incubation /Particle Plating)
11. Cultural characters:	
	(hypha /spore /Conidium); Germinated: Yes/No
	; Height: ; Colour: ; Size: (diam. range in cm/7 days.)
	(Smooth /Serrated /Wavy /Rhizoidal /)
Reverse:	(white /brown /colourless /black /etc.)
Growth:	(Slow: 0.2 -1.0cm; Median: 1.1- 4.9 cm; Fast:5.0- 9.0 cm diam.)
Teyture:	(Cottony /Slimy /Dry /Powdery /Wet /Floppy /Flat /Concentric rings
	Convex /Dome /Wavy /Parched /Stubs /Exudates /Crystals /Fruit-bodies);
12. Microscopic:	Soliver 150me 1 wavy 11 at ched 15tubs 12 reductes 1 el jotals 11 tute 60 dies);
Mycelium/Hyphae:	(Smooth /Verrucose /Septate /Branched /Hyaline /Dark or light
iviyeenanviiypnae.	Brown / Guttulate /Thick or Thin-walled /um wide)
Conidiomata:	(Synnematous /Mononematous /Sporodochial /Pycnidial)
Conidiophore:	(Smooth /Verrucose /Septate /Branched /Hyaline /Dark or light
Comarophore.	/Brown /Thick or Thin-walled / lum long and wide)
Conidiogenous cell:	(Poly or Monoblastic /Phialidic /Annelidic /Percurrent /Tretic
Comarogenous cen.	/Basauxic /Gangliar /intergrated /discrete)
Conidia	(Dry /Wet /Catenate /Solitary /Globose /Clavate /Pyriform /Fusiform
Conidia:	/helicoid /Septate /Branched /Smooth /Verrucose /Velvety)
	(Stromatic /Solitary /Gregarious /Superficial /Submerged /Perithecial
Ascocarp:	/Apothecial /Cleistothecial /Stalked /Sessile /Ostiolate /)
Assessme Walls	(Textura angularis /Pseudo-parenchymatous /Plectnchymatus
Ascocarp Wall:	
Contract	Mycelial /Carbonaceous /Colourless /.coloured)
Centrum:	(Paraphysoid /Pseudo-/Apical paraphysoid /Loculate /)
Ascus:	(Bi-/Uni-tunicate /Stalked /Sessile /With-/Without Apical Apparatus
A	Operculate /Inoperculate /Clavate /Cylindrical /Curved /
Ascospore:	(Shape, colour, size, appendaged, septate)
Pycnidium:	(Stromata/sclerotia /Wall: /Colour /)
	terial from field (Leaf, Twig, Bark, Fruit, Seed)
	ure derived 2.1 Live culture No. 2.2. Dried culture mat No.
3 Slid	e ino.

(e) Identification:

Taxonomic identification of the fungi was done by observing the habitat, colony morphology and sporulating structures as diagnostic characters. The identified isolates were assigned to respective genera and species using standard monographs and keys (Carmichael *et al.*, 1980; Descals, 1995; Ingold, 1975; Matsushima, 1971, 1975; Webster & Descals, 1981). The description of the fungi was written in a diagnostic form. Details of specimens examined for each fungus were given.

(f) Ex situ Conservation of fungi:

The description of species was based on specimens or cultures of fungi, as prescribed in the International Code of Botanical Nomenclature (Hawksworth, 1974). Pure cultures of aquatic fungi recovered were properly labelled, numbered and maintained in the collections of the Goa University Fungus Culture Collection (GUFCC). The microslides containing fungal diagnostic structures were made, properly sealed, labelled and maintained in the Goa University Botany Herbarium (GUBH).

Holotypes of all new taxa described in this thesis were maintained at the International Mycological Institute, Cabiscience, U.K. Where the new taxon is based on a live fungus, dried and dead culture mats in herbarium sheets were maintained in the Herbarium of the IMI, U.K. and/or the GUBH, Goa University.

3.5.3. Water:

Temperature and pH of the water were recorded on the spot using a laboratory thermometer and pH paper respectively. The canopy cover of each sampling site was measured using a spherical densiometer (Model-C, Forest densiometers, Cornell).

Following the methods outlined by Trivedy *et al.* (1998), for analysis of chemical properties, samples were collected from the sites in sampling bottles and transferred to the laboratory in an ice-pack container.

- a) <u>Dissolved oxygen</u>: For analysis of dissolved Oxygen, water was transported in ambercoloured bottles. The reagents used were as follows:
- i) Sodium thiosulphate, 0.025 N: 24.82 g of Na₂S₂O_{3.5}H₂O was dissolved in pre-boiled distilled water and the volume was made to 1 L. One pellet of NaOH was added as a stabilizer. This was 0.1N stock solution. It was diluted to 4 times to prepare 0.025 N solution and manitained in a brown glass bottle.
- ii) Alkaline potassium iodide solution: 100g of KOH and 50g of KI were dossolved in 200ml of preboiled distilled water.
- iii) Manganous sulphate solution: 100g of MnSO₄.4H₂O was dissolved in 200ml of distilled water and warmed to dissolve maximum salt. The content was filtered after cooling.
- iv) Starch solution: 1 g of starch was dissolved in 100ml of distilled water.
- v) Sulphuric acid, conc.

Method: The water sample was filled in a 250 ml glass-stoppered bottle and air bubble getting trapped while placing the stopper was avoided. One ml of each MnSO₄ and alkaline KI solutions were added well below the surface through the wall. A brown precipitate appeared indicated the presence of Oxygen. The contents were well-shaken by repeatedly inverting the bottle and the precipitate was allowed to settle. Two ml of conc. H₂SO₄ was added and shaken well to dissolve the precipitate. About 100 ml of the content was trasferred to a conical flask and titrated against sodium thiosulphate solution

using starch as an indicator. The initial dark blue-black colour changes to colourless.

Calculation: Dissolved oxygen, $mg/L = (ml \times N)$ of sodium thiosulphate $\times 8 \times 1000$

V2 (<u>V1-v</u>)

Where V1 = volume of sample bottle

V2 = volume of contents titrated

v = volume of MnSO4 and KI added (2ml)

b) Dissolved Carbon Dioxide: Free CO2 in water was determined by titrating the sample

using a strong alkali to pH 8.3. The following reagents were used:

i) Sodium hydroxide, 0.05 N: 40 g of NaOH was dossolved in boiled CO₂ free distilled

water and the volume was made up to 1 L. The solution was filtered through a sintered

glass filter to remove any NaCO₃ and to get 1N NaOH solution. This solution was diluted

to 20 times to prepare 0.05 N solution.

ii) Phenolphthalein indicator: 0.5 g of phenolphthalein was dissolved in 50ml of 95%

ethanol and mixed with 50ml of distilled water. 0.05 N CO₂ free NaOH solution was

added dropwise, until the solution turns to faintly pink colour.

Method: 100ml of sample was taken in a conical flask and a few drops of

phenolphthalein indicator added. The change of colour to pink indicated the absence of

free CO₂. In case the sample remained colourless, titration was done with 0.06 N NaOH,

in which a pink colour appears as the end point.

Calculation: Free CO₂, mg/L = $(ml \times N)$ of NaOH $\times 1000 \times 44$ ml sample

c) <u>Total alkalinity</u>: The following were the reagents used in detecting total alkalinity of

the water sample:

i) Hydrochloric acid, 0.1 N: 12 N conc. HCl was diluted 12 times (8.34 in 100ml) to get

10 N HCl. This was further diluted to 10 times to prepare 0.1 N HCl (100 in 1000ml).

- ii) Sodium carbonate, 0.1 N: 5.3 g of Na₂CO₃ (pre-dried at 250°C for 4 h) was dissolved in distilled water to prepare 1 L of solution.
- iii) Methyl orange indicator: 0.5 g of methyl orange was dissolved in 100ml of distilled water.
- iv) Phenolphthalein indicator: 0.5 g of phenolphthalein was dissolved in 50 ml of 95% ethanol and mixed with 50ml of distilled water. 0.05 N CO₂ free NaOH solution was added dropwise, until the solution turned faintly pink.

Method: 100ml of the sample was taken in a conical flask and a few drops of phenolphthalein was added. When the colour of the sample changed to pink, it was titrated with 0.1 N HCl until the colour disappeared at the end point. This is 'phenolphthalein alkalinity' (PA). Two to 3 drops of methyl orange was added to the same sample and the titration was continued further, until the yellow colour changed to pink at the end point. This is total alkalinity (TA).

Calculation: PA as $CaCO_3$ mg/l = $(A \times N)$ of $HCl \times 1000 \times 50$ ml/sample

TA as $CaCO_3$ mg/l = (B x N) of HCl x 1000 x 50 ml/sample

where A= ml of HCl used only with phenolphthalein
B= ml of HCl used with phenolphthalein and methyl orange, ie. total HCl
used with both the indicators

- f) <u>Sodium (Flame Photometric method)</u>: The following were the reagents used in the estimation of sodium in water sample:
- i) Stock sodium solution, 1000mg/L: 2.542g NaCl (pre-dried at 140°C) was dissolved in distilled water and made to 1 L of solution.
- ii) Intermediate sodium solution, 100mg/L: The above solution was diluted to 10 times.

- iii) Standard sodium solution (10 mg/L): The intermediate solution was further diluted 10 times.
- iv) Nitric acid: HNO3, conc.
- v) Hydrochloric acid: HCl, conc.
- vi) Hydrogen peroxide: H₂O₂, 30%
- vii) Ammonium hydroxide: NH4OH, conc.

Pretreatment of the sample: The sample was taken in a 250 ml conical flask and acidified using nitric acid. This was evaporated to dryness in a water bath. 25ml of conc. HNO₃ was added and heated near to boil until the acid evaporated to small volume. The presence of brown fumes indicated the unoxidized organic matter. Additional conc. HNO₃ was added and small quantities of H₂O₂ for complete ashing of organic matter. The final residue was colourless on drying. The residue was dissolved in small amount of HCl and distilled water. The contents were filtered and neutralized with conc. NH₄OH (liquid ammonia) and diluted to suitable volume.

<u>Method</u>: The flame photometer was set for a particular range of sodium concentration, 0-100 mg/L depending upon the amount of sodium present in the sample. A 'calibration curve' was prepared using the range of 0-100 mg/L of sodium.

Calculation: Na, $mg/L = (mg/L \text{ Na in diluted aliquot}) \times \text{dilution factor}$

- g) <u>Potassium (Flame Photometric method):</u> The following were the reagents used for estimation of potassium:
- i) Stock potassium solution (1000 mg/L): 1.907 g of KCl (pre-dried at 110°C) was dissolved in deionized distilled water to prepare 1 L solution.
- ii) Intermediate potassium solution (100 mg/L): The stock solution was diluted to 10

times.

iii) Standard potassium solution (10 mg/L): The intermediate solution was diluted further 10 times.

Method: The same procedure as described for determination of sodium was followed except that 768 nm filter was used for potassium.

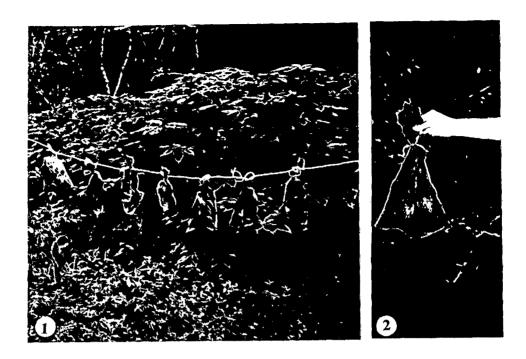
3. 6. Litter bag experiment:

An *in situ* exercise, 'litter-bag experiment', was conducted in one of the sampling sites of seasonal studies, the Molem wildlife sanctuary, using two riparian plant species, i.e. *Hopea ponga* and *Dillenia indica*, for a duration of 8 months, from September 2000 to April 2001, to study the colonisation of aquatic fungi in nature (Plate.4.1, 4.2, 4.3).

The sampling units were A4 size nylon mesh bags (mesh size # 3 mm), each containing a hand full of naturally shed senescised and decayed leaves of *Hopea ponga* and *Dillenia indica*, collected from the vicinity of stream at Molem. The littler bags were tied together to a nylon rope and suspended in stream water at the same locality in the month of August 2000. The rope was tied to roots of riparian trees so as to prevent washing away in water current. The bags were examined at monthly intervals for presence of aquatic fungi using diffreent isolation techniques outlined above. Water samples were analysed for physical and chemical properties.

3.7. Effect of mining on growth and sporulation of aquatic fungi:

To see the effect of mining rejects on growth and sporulation of aquatic fungi, foam and submerged leaf samples were collected from two perennial streams at Cudnem



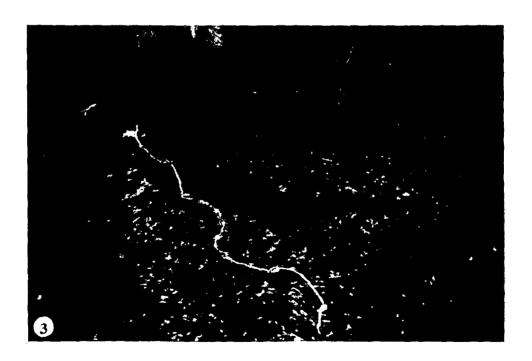


Plate. 4

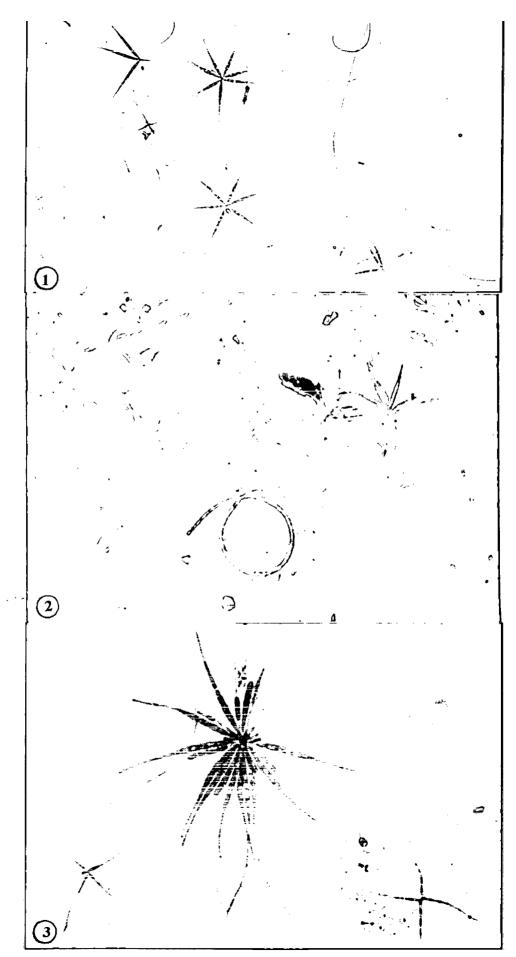


Plate. 5

Iron Ore Mines, Goa. The samples were subjected to various fungal recovery processes such as water aeration, still water incubation and single spore isolation from foam.

Mining rejects were also brought to the laboratory in fresh polythene bags. Rejects in different quantities (5g, 10g and 15 g) were aerated along with submerged leaves placed in glass jars in sterile distilled water. Control was run using natural soil gathered from away from mining site. The floristic analysis for aquatic fungi appearing in each jar was conducted by observing the spora under an inverted microscope.

3. 8. Enzyme assays:

Of the nearly 800 isolates obtained by single spore isolation method from foam samples, 60 isolates of 'strict aquatics' were subjected for screening experiments in order to detect the presence of degradative enzymes such as amylase, cellulase, esterase and pectinase. The methods prescribed by Carder (1986)) for amylase and Hankin & Anagnostakis (1975) for cellulase and pectinase were followed.

Discs of 5mm, cut from 7-10 day old colonies grown at room temperature (22-25°C) on MEA were used as the source of inoculum for enzyme assays.

a) <u>Amylolytic activity</u>: The ability to degrade starch was used as the criterion for determination of amylolytic activity of fungi. The composition of 'starch agar' medium used was as follows: malt extract 5g/l, agar 20g/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, 10 μg; MnSO₄, 10 μg; ZnSO₄, 70 μg; CuSO₄ 50 μg; MoO₃, 10 μg; pH 6. The test fungus was point-innoculated in the centre of the medium and allowed to grow for 7 days. Amylolytic activity was observed by flooding the plates with 1%

iodine solution (Hankin & Anagnostakis, 1975). A clear yellow zone around the colony in an otherwise blue medium indicated the production of amylase (Plate.13.2).

- b) <u>Cellulolytic activity</u>: The composition of the medium used was as follows: malt extract agar 5g/l, agar 20g/l, carboxymethyl cellulose 10g/l and mineral salt solution as described above in (a). The test fungus was point-innoculated and after 7 days 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 allow the clearance zone to stay for a longer period (Plate.14.1).
- c) Esterase activity: The medium used is for 'presumptive fatty acid esterase activity' (Sierra, 1957). It contains Tween 80 as C source, bromocresol purple as pH indicator and a Calcium salt. Tween 80 is a mixture of fatty acids predominantly elaidic, linoleic and palmitic, and the final pH of the medium is acidic. Growth of the fungus on fatty acids result in rise of pH, turning the medium blue-purple and formation of insoluble calcium salts such as calcium stearate, which appear as a white ring in the medium around the colony (Plate.14.2).

Composition of the medium: Peptone 10 g, NaCl 5 g, CaCl₂.2H₂O 0.1 g, bromocresol purple 25 mg, agar 15 g in 1litre of distilled water. The pH of the medium was adjusted to 5.4. The medium were sterilized in an autoclave at 15 psi for 20 min. 10 ml of aqueous solution of Tween 80 (10%) dissolved in 80-90 ml warm distilled water (65-70°C) was separately sterilized. On cooling to 65-70°C, 10ml Tween solution was mixed with 90 ml basal medium and poured into 5 cm diam. petridishes (10 ml/plate). The plates were point-inoculated with test fungus and incubated for 8-10 days.

d) <u>Pectinolytic activity</u>: Composition of the medium: mineral salt solution 500 ml, yeast extract 1 g, agar 15 g, pectin 5 g and distilled water 500 ml. The plates were point-inoculated with test fungus and incubated for 8-10 days and then flooded with Centrimide (hexadecyltrimethyl ammonium bromide). The reagent precipitates intact pectin in the medium and presence of clear zone around the colony in an opaque medium indicated degradation of pectin (Hankin & Anagnostakis, 1975) (Plate.13.1).

3. 9. Statistical analysis:

The data collected during the 24 month study period was subjected to statistical analysis. 'Margaliff's' and 'Shanon's' index were used for analysis of diversity of fungi. 'Analysis of Variance (Anova) Test' was carried out to analyse the variations in occurrence of fungi in different seasons. The abundance of fungi in monsoon and frequency of occurrence of diverse fungi in different localities were correlated using 'rare-faction curves'. The following formula were used for statistically analysing the data obtained during the seasonal and diversity study of aquatic fungi.

Percentage Frequency = <u>Total no. of quadrats in which a species occurred</u> X 100 (%F)

Total no. of quadrats sampled

Relative Frequency = No. of occurrence of a species X 100 (R. F) No. of occurrence of all species

Density (D) = <u>Total no. of individuals of the species</u> Total no. of quadrats taken

Abundance (A) = <u>Total no. of individuals of a species in all quadrats</u>
Total no. of quadrats in which the species occurred

The number of species that can be expected in a sample of n individuals [denoted by $E(S_n)$] drawn from a population of N total individuals distributed among S species was

calculated using the formula:

$$E(S_n) = \sum_{i=1}^{S} \sum_{n=1}^{N} [1 - \{(N-n_i) / (N)\}]$$

whereas n_i is the number of individuals of the ith species (Ludwig & Reynolds, 1988).

CHAPTER 4: RESULTS AND DISCUSSION

RESULTS

This investigation on aquatic fungi occurring in the streams and rivers of forests of Western Ghats in Goa State was carried out for a period of more than two years, from January 1999 to May 2001. For study of seasonal occurrence and abundance of fungi, sampling of aquatic fungi was done during pre-monsoon, monsoon and post-monsoon periods in the streams of Bondla, Molem and Cotigao wildlife sanctuaries of Goa State. All the streams had dense riparian canopy.

Studies on the diversity and floristics of fungi were carried out based on samples gathered from several streams and rivers of Goa in monsoon and post-monsoon seasons during which the flow of water was continuous. The sampling sites exhibited varying degrees of density in moist deciduous tree canopy. Submerged leaf-litter (SL), floating foam (FF) and submerged roots (SR) constituted the primary source of aquatic fungi in this study.

Interiors of submerged roots and litter of riparian trees maintained in nylon bags were scanned at regular and periodical intervals for aquatic fungi and their presence was analysed confirm the fate of aquatic fungi in dry seasons. The activity of aquatic fungi was studied with reference to their ability to secrete enzymes and to grow in waters of mining rejects.

The results are presented below in four separate parts.

<u>PART I</u>: TAXONOMIC DIVERSITY AND HABITAT AFFINITY OF AQUATIC FUNGI

In all, 102 species of fungi belonging to 80 genera were recoverd. These belonged to various taxonomic groups, namely Hyphomycetes, Coelomycetes and Ascomycetes (Table 4.1.1), and of which majority were of Hyphomycetes (Table 4.1.2). The description and illustration of aquatic conidial fungi are given below in the taxonomic part. The ascomycetous fungi are listed in Table 4.1.3.

Standard diagnostic characters followed in the taxonomy of conidial fungi such as structure and size of conidiomata, shape, size, septation and branching pattern of conidiophores, type of conidiogenesis, shape and size of conidiogenous cells and conidia were used while describing the imperfect fungi (Table 3.4). Camera Lucida drawing and photomicrographic illustration of diagnostic features such as conidiophores, conidiogenous cells and conidia of described taxa are given for all described species.

The description of species is based on the fungi isolated from natural foam, submerged leaf litter and those recovered in pure culture form. The microslides and specimens containing fungal diagnostic structure on which the descriptions based were maintained at the Herbarium of Goa University Botany Department (GUBH). The pure cultures of fungi recovered in this study were housed at the Goa University Fungus Culture Collection Unit (GUFCC).

Latin is required to be provided for the diagnosis of all new taxa and in the absence of this, as per article 36 of the International Code of Botanical Nomenclature

(Hawksworth, 1974), the novelty of these fungi remains only provisional. As and when the new taxa are published, latin will be provided for the diagnosis.

Table 4.1.1: Fungi recovered from different streams in Goa:

Stream	Hyphomycetes			<u>Coelomycetes</u>				<u>Ascomycetes</u>	
Locality	FF	SL	SR	FF	SL	SR	FF	SL	SR
Alorna	23	13	-	2	-		-	-	-
Anmod	2 7	17	-	3	1	-	-	-	_
Bondla	61	47	8	5	3	2	-	-	2
Chorlem	20	6	-	1	-	-	-	-	-
Cotigao	67	48	9	4	2	2	-	1	3
Cudnem	13	8	4	-	-	-	-	-	-
Cuncolim	18	9	-	-	-	-	-	-	-
Harvalem	16	6	-	1	1	-	-	-	-
Kesarval	14	5	-	1	1	-	-	-	-
Molem	53	32	6	3	-	3	-	-	2
Taleigao	10	2	-	1	-	-	-	-	-
Tambdisurla	26	10	-	2	-	-	-	-	-

TAXONOMIC PART:

1. HYPHOMYCETES

1. Actinospora megalospora Ingold, 1952. Trans. Br. mycol. Soc. 35: 68-69.

(Fig. 1)

Conidial fungus, Hyphomycete. *Colonies* on MEA effuse, with a circular margin, white to pale cream, regular, attaining a diam. of 4.5 cm in 14 days. *Mycelium* dense, partly immersed in the medium, composed of smooth, septate, branched, hyaline, thinwalled hyphae 2-3 µm wide. The fungus does not sporulate on agar medium; but sporulates when an agar block with mycelia composed of indistinct conidiophores and conidiogenous cells immersed in sterile distilled water. *Conidiophores* mycelial, thickwalled. *Conidiogenous cells* enteroblastic. *Conidia* large, with a spherical central region

composed of dense cytoplasm, 50-60 µm diam, hyaline, with 3-6 (mostly 4) divergent, 6-12-septate, smooth, thick-walled, 120-200 µm long, 2-5 µm wide arms. Conidia isolated from foam are similar to those produced in submerged agar blocks.

Specimen examined: (i) Dried foam slide, monsoon seasonal stream, Molem Wildlife Sanctuary, Goa, India, Herb. No. GUBH-140, Sreekala K. Nair, 9-8-1999. (ii) Dried culture mat derived from foam conidium, monsoon seasonal stream, Molem Wildlife Sanctuary, Goa, India. Herb. No. GUFCC-140-a, Sreekala K. Nair, 9-8-1999. (iii) Dried foam slide, post-monsoon seasonal stream, Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUBH-141, Sreekala K.Nair, 17-12-2000.

Although this is the first record for Goa State, the fungus was first described from Silent Valley in Kerala State, India (Subramanian and Bhat, 1981) and subsequently from the forests of Western Ghats in Karnataka State (Sridhar et al., 1992).

2. Alatospora acuminata Ingold, 1942. Trans. Br. mycol. Soc. 25: 381-384. (Fig. 2)

Conidial fungus, Hyphomycete. *Colonies* on MEA effuse, regular, white, circular, composed of sparcely immersed mycelium, attaining a diam. of 2.2 mm after 7 days. *Mycelium* composed of smooth, septate, branched, hayline hyphae 2.2 µm wide. *Conidiophores* micronematous, indistinct. *Phialoconidium* alate-shaped, hyaline, tetraradiate, with arms 15-25 x 1.5-2.5 µm. Culture on agar medium did not sporulate, but produced conidia when agar blocks containing fungal mycelium were submerged in sterile distilled water.

Specimen examined: (i) Dried culture mat derived from single conidium from foam, seasonal stream, Bondla Wildlife Sanctuary, Goa, India, Herb. No. GUFCC-115, Sreekala K. Nair, 9-8-1999. (ii) Dried culture mat drived from single conidium isolated from foam samples, seasonal stream, Cotigao Wildlife Sanctuary, Goa, India, Herb. No.

GUFCC-116, Sreekala K. Nair, 24-8-2000. (iii) Dried culture mat derived from single conidium from submerged leaf litter, seasonal stream, Molem Wildlife Sanctuary, Goa, India, Herb. No. GUFCC-117, Sreekala K. Nair, 20-9-2000.

The fungus was previously reported from the Western Ghat forest streams of India (Sridhar et al,1992). However, this is the first report from Goa State.

3. Anguillospora crassa Ingold, 1958. Trans. Br. mycol. Soc. 41: 365-72. (Fig. 3)

Conidial fungus, Hyphomycete. *Colonies* in MEA effuse, with circular margin, regular, greyish blue, attaining 2.5 cm diam. in 14 days. *Mycelium* sparcely superficial, composed of branched, light brown, septate, thin-walled hyphae 2-4 µm wide. *Conidiophores* micronematous, indistinct. *Conidiophores* cells monoblastic, integrated, terminal, 5-10 µm long, 2.2 µm wide. *Conidia* elongated, cylindrical, 6-8-septate, hyaline, up to 180 µm long, curved at the tip forming a broad hook. The fungus sporulates moderately well in agar medium.

Specimen examined: (i) Dried foam slide, seasonal stream, Chorlem Ghat, Goa, India, Herb. No. GUBH-139, Sreekala K. Nair. 2-7-1999. (ii) Dried foam in slide, seasonal stream, Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUBH-142, Sreekala K. Nair. 19-8-1999. (iii) Dried culture mat derived from single conidium, seasonal stream, Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUFCC-142-a, Sreekala K.Nair. 19-8-1999.

The fungus was found to be a common inhabitatnt in the streams of forests of Western Ghats of Kerala and Karnataka. Besides other parts of the world, in India it was earlier reported from Uttar Pradesh (Sridhar et al., 1992).

4. Anguillospora furtiva Descals, 1997. Mycotaxon. 68: 433-435 (Fig.4)

Conidial fungus, Hyphomycete. Colonies with alternating grey and white

concentric rings, with diffused margin, with sparse aerial mycelium, attaining a diam. of 2 cm in 15 days. *Conidiophores* hyaline, solitary, semi-macronematous, simple, septate, sparsely branched. *Conidiogenous cells* monoblastic, terminal, integrated, with a broad scar on sessesion of conidia, with 1-4 percurrent proliferations. *Conidia* hyaline, long-fusoid, curved, with one to two intercalary constrictions, multiseptate, with truncate to conical base, 150-300 x 6.5-10 µm.

Specimen examined: Dried foam slide, seasonal stream, Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUBH-103, Sreekala K.Nair, 19-8-1999.

Descals (1997) found this fungus in scums of various streams of Great Britain. as an anamorphic phase of *Pezoloma* (Leotiales). This is the first report of the species from tropical waters.

5. Anguillospora longissima (Sacc. and Sydow)Ingold, 1942. Trans. Br. mycol. Soc. 25: 402. (Fig.5)

Conidial fungus, Hyphomycete. *Colonies* effuse, regular, grey to brown, attaining a diam. of 3.2 cm. in 14 days; reverse of the colony pale to medium brown. *Mycelium* sparcely superficial, composed of smooth, septate, branched, pale brown, thin-walled hyphae 2-2.5 µm wide. *Conidiogenous cells* monoblastic, integrated and terminal, truncate at the tip on succession of conidia. *Conidia* elongated, cylindrical, sigmoid with two curvature in more than one plane, hyaline, 8-12-septate, 200-280 x 2.5-3.5 µm, often carrying ramains of separating cell as a basal frill. The colony derived from single spore isolate readily but moderately sporulated in MEA medium after 15 days of incubation at 20°C.

Specimen examined: (i) Dried culture mat derived from single spore isolate of the fungus, Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUFCC-158, Sreekala K. Nair,

19-8-1999. (ii) Dried foam slide, seasonal stream, Alorna, Goa, India, Herb. No. GUBH-160, Sreekala K. Nair, 20-7--2000. (iii) Dried foam in slide, seasonal stream, Bondla, Wildlife sanctuary, Goa, India, Herb. No. GUBH-164, Sreekala K.Nair, 21-10-2000.

The fungus has earlier been reported from the streams of Western Ghat forests (Sridhar et al., 1992).

6. Ardhachandra selenoides Subram. and Sudha, 1978. Can. J. Bot. 56: 729-731. (Fig. 6)

Conidial fungus, Hyphomycete. *Colonies* on MEA effuse, pale brown, slow growing, attaining a diam. of 4.5 cm in 10 days. *Mycelium* partly superficial, composed of pale brown, branched, septate hyphae 2-3 µm wide. *Conidiophores* solitary, simple, erect, 0-3-septate, rarely branched, pale brown, 30-55 µm long, 3.5-4.5 µm wide. *Conidiogenous cells* polyblastic, denticulate, with 4-15 conspicuous denticles in the above half; denticles peg-like, cylindrical to broadly conical, 1-1.5 x 0.5-0.7 µm. *Conidia* broadly selenoid in side view, obovoid to clavate in face view, guttulate, 19-22 x 6.5-7.5 µm. The conidia readily germinate in sterile distilled water within 20 h. On MEA, the fungus sporulated after 10 days.

Specimen examined: (i) Dried foam in slide, seasonal stream, Molem Wildlife Sanctuary, Goa India, Herb. No. GUBH-155, Sreekala K. Nair, 18-9-1999. (ii) Dried culture mat derived from single spore isolate of the fungus, Molem, Goa, India, Herb. No. GUFCC-155-a, Sreekala K. Nair, 18-9-1999. (iii) Dried foam slide, perennial stream, Kesarval, Goa, India, Herb. No. GUBH-156, Sreekala K. Nair, 10-8-2000.

The fungus is a well known species found inhabiting terrestrial leaf litter but often encountered in stream waters, both in submerged leaf litter and foam (Matsushima, 1975).

7. Ardhachandra sp. (Fig. 7)

Conidial fungus, Hyphomycete. Colonies on MEA effuse, circular, pale to medium

brown, attaining a diam. of 4.2 cm in 10 days; reverse of the colony dark brown. *Mycelium* partly superficial, composed of pale to mid-brown, septate, branched, hyphae 2.5-3.5 μm wide. *Conidiophores* solitary, simple, erect, 0-3-septate, pale brown, 30-60 μm long, 3.5-4.5 μm wide. *Conidiogenous cells* polyblastic, denticulate, with 5-10 denticles per cell; denticles small, 1-1.5 μm long, 0.5-0.75 μm wide. *Conidia* obovoid to elliptical,non-selenoid, 15-25 x 6.5-8 μm. Colony derived from single spore isolation readily growing and sporulating in MEA after 10 days of incubation at 20°C.

Specimen examined: (i) Dried foam slide, seasonal stream, Bondla Wildlife Sanctuary, Goa, India, Herb. No. GUBH-154, Sreekala K. Nair, 9-8-1999. (ii) Dried culture mat derived from single spore isolate of the fungus, Bondla Wildlife Sanctuary, Goa, India, Herb. No. GUFCC-157, Sreekala K. Nair, 25-10-1999.

8. Articulospora tetracladia Ingold, 1942. Trans. Br. mycol. Soc. 25: 376. (Fig. 8)

Conidial fungus, Hyphomycete. *Colonies* effuse, circular, regular, off white to pale brown; reverse of the colony pale brown. *Mycelium* composed of smooth, septate, hyaline, thin-walled hyphae 2-2.3 µm wide. *Conidia* tetraradiate, hyaline, with four, 1-3-septate divergent arms, 25-45 x 3 µm, show considerable variation in size. The fungus remained sterile in agar medium but readily sporulated when an agar block containing fungal mycelium was immersed in sterile distilled water and incubated for 7 days.

Specimen examined: Dried foam slide, seasonal stream, Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUBH-130, Sreekala K. Nair, 24-8-2000.

The fungus was reported earlier from stream waters of Western Ghats of Karnataka and Kerala and eastern Ghats of Tamil Nadu and Andhra Pradesh (Sridhar et al., 1992). However, here it is recorded for the first time from Goa.

Conidial fungus hyphomycete. *Colonies* effuse, white to pale brown. *Mycelium* consists of smooth, septate, hyaline and thin-walled hyphae, 2-3 µm wide. Conidiophores are hyaline, thinwalled, aseptate, arising from the vegetative hyphae, 10-10 µm long and 3-5 µm wide. *Conidia* are hyaline, smooth, thin-walled, triradiate with three, 1-3-septate divergent arms each measuring 20-30 x 3 µm.

Specimen examined: Dried submerged leaves, seasonal stream. Molem Wildlife Sanctuary, Goa, India, Herb. No. GUBH-118, Sreekala K. Nair, 18-9-1909.

The genus *Articulospora* Ingold has 4 species so far described (Ingold, 1942; Matsushima, 1975). The present collection does not fall under any of these due to its only 3 radiating arms and therefore recognised as distinct.

10. Bahusutrabeeja angularis Vasant Rao and de Hoog, 1986. Stud. Mwol. 28: 66-67.

(Fig. 9)

Conidial fungus, Hyphomycete. *Colonies* on the substrate superficial, dark brown to blackish, consist of scattered, shiny conidiophores each with a conspicuous colourless, globose slimy conidial mass at its tip. *Mycelium* scanty, immersed, composed of subhyaline to brown, septate, branching hyphae 2-3 µm wide. *Conidiophores* erect, sometimes bent, simple or branched, thick-walled, 150-280 x 5.5-10 µm, usually 5-12 septate, dark brown towards the base and paler towards the apex, terminating in a conidiogenous cell. *Conidiogenous cell* phialidic, slightly narrowed at the neck, with a short collarette above 24-46 µm long, 6-9 µm wide. *Conidia* 1-celled, thin-walled, rounded with 3-4 angles, hyaline with granular cytoplasm, produced singly and successively at the tip of the phialide, 10-14 µm diam, with 3-4 slender, nonseptate, 4-6

μm long appendages arising from the angles.

Specimen examined: Submerged leaves of *Saraca asoca* (Roxb) De Wilde, seasonal stream, Molem Wildlife Sanctuary, Goa, India, Herb. No. GUBH-144. Sreekala K. Nair. 25-12-2000.

The fungus is a terestrial form earlier reported from the forests of Western Ghats in Jog Falls, Karnataka State (Vasant Rao and de Hoog, 1986). Here, it is a first record from aquatic stream environment.

11. *Beltrania rhombica* O. Penzig, 1882. *Nuova G. bot. Ital.* **14**: 72-75. (Fig. 10)

Conidial fungus, Hyphomycete. *Colonies* on MEA effuse, circular, brown, 5 cm diam. in 7 days; reverse of the colony dark brown. *Mycelium* moderate, brown, with thinwalled, septate, smooth, 1.5 µm wide hyphae. *Conidiophores* mononematous, flexuous, brown, septate, smooth, 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, with the free end drawn into a tiny spine like structure, 0-septate, smooth, pale olive brown, with a disticnt hyaline transverse band immediately above the widest part, 22-30 x 8-10 µm; appendages 10-11 µm long, 1.5 µm wide at its base.

Specimen examined: (i) Dried foam slide, seasonal stream, Molem Wildlife Sanctuary, Goa, India, Herb. No. GUBH-150, Sreekala K. Nair, 20-9-2000. (ii) Dried culture mat derived from single conidium, seasonal stream, Molem Wildlife Sanctuary, Goa, India, Herb. No. GUFCC-150-a, Sreekala K. Nair, 20-9-2000. (iii) Dried foam slide.

seasonal stream, Chorlem Ghat, Herb. No. GUBH-151. Sreekala K. Nair, 4-10-2000.

The fungus is a terestrial form commonly found on leaf litter in abundance in the forests of tropical countries. (Ellis, 1971). The fungus has also been reported from foam samples (Ingold, 1975, Subramanian and Bhat, 1981).

12. Beltraniella odinae Subram., 1952. Proc. Indian Acad. Sci. Sect. B. 36: 223-228. (Fig. 36)

Conidial fungus, Hyphomycete. *Colonies* on MEA effuse, olivaceous dark to brown, velvety, 2.8 cm diam. in 10 days; reverse of the colony dark brown. *Mycelium* brown with thin, smooth, septate, branched hyphae 2-3.5 μm wide. *Conidiophores* mononematous, straight, unbranched, smooth, pale brown, arising at the base of the seta, 37-75 x 2.5-4 μm. *Setae* unbranched, erect, thick-walled, dark brown, 85-200 x 3-3.5 μm. *Conidiogenous cells* terminal, discrete, polyblastic, ampulliform, globose to subglobose, denticulate, light brown, 8-10 x 3-4.5 μm. *Conidia* solitary, 0-septate, with a transverse band, just above the centre, obclavate, terbinate, with the base drawn out to a fine point, hyaline, smooth, 20-25 x 5-6.5 μm, attached to the conidiogenous cells by a hyaline, smooth, fusiform, 12.5-14 x 2-3 μm, separating cell.

Specimen examined: (i) Dried submerged roots of *Hopea ponga* (Dennst.) Mabb., perennial stream, Molem Wildlife Sanctuary, Goa, India, Herb. No. GUFCC-123, Sreekala K. Nair, 25-12-2000. (ii) Dried culture mat derived from single spore isolate of the fungus, perennial stream, Molem Wildlife Sanctuary, Goa, India, Herb. No. GUFCC 123-a, Sreekala K. Nair, 25-12-2000.

The fungus is a commonly found terestrial form and reported as a saprophyte on leaves and twigs in tropical forests (Ellis, 1971, 1976). Here, it is a first record of its occurrence as a root endophyte in stream environment.

13. Brachysporiella laxa (Hudson) M. B. Ellis. 1971. Dematiaceous Hyphomycetes.

(Fig. 74)

Conidial fungus, Hyphomycete. *Colonies* effuse, dark blackish brown to black hairy. *Mycelium* immersed. *Conidiophores* solitary or in groups of 2-4, erect, straight, cylindrical to subulate, bearing several branches towards the apex, mid to dark brown, smooth, upto 250 µm long, 6-9 µm thick. *Conidiogenous cells* cylindrical or doliiform, pale to mid brown, 7-13 µm long, 3-5 µm thick. *Conidia* solitary, acrogenous, ellipsoidal to clavate, narrowed to a truncate base, mid to dark brown, end cells often pale, 2-3 septate with dark bands at the septa, 16-30 µm long, 8-12 µm thick in the broadest part.

Specimen examined: On submerged twig of *Ficus benghalensis* Linn., perennial river, Tambdisurla, Goa, India, Herb. No. GUBH-24, Sreekala K. Nair, 8-7-2000.

14. Camposporium pellucidum (Grove) Hughes, 1951. Mycol. Pap. 36: 9. (Fig. 13)

Conidial fungus, Hyphomycete, *Colonies* effuse, grey to brown, attaining a diam. of 2 cm. in 10 days. *Mycelium* partly superficial, composed of smooth, septate, branched, brown hyphae 3-5 µm wide. *Conidiophores* mononematous, straight or flexuous, 7-10-septate, unbranched, smooth and paler towards the apex, up to 150 µm long, 5-8 µm wide. *Conidiogenous cells* integrated, terminal, polyblastic, sympodial, cylindrical or subulate, denticulate, with a denticulate to cylindrical pedicel or separating cell. *Conidia* solitary, dry, appendiculate, 60-140 x 7.5-12 µm, up to 16-septate, cylindrical and round at the apex, with 1-3 filiform, hyaline, septate appendages up to 145 µm long, 2µm thick at the tip.

Specimen examined: (i) Submerged leaves collected from a seasonal stream,

Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUBH-129, Sreekala K.Nair. 19-8-1999. (ii) Dried foam on slide, from a seasonal stream, Molem Wildlife Sanctuary, Goa, India, Herb. No. GUBH-131, Sreekala K.Nair, 20-9-1999. (iii) Dried culture mat derived from a single spore isolate, seasonal stream, Molem Wildlife Sanctuary, Goa, India, Herb. No. GUFCC-131-a, Sreekala K. Nair, 20-9-1999.

The fungus is terestrial form commonly found on twigs in the forests (Ellis, 1971, 1976). It was also recorded earlier from aquatic environment (Ingold, 1975; Subramanian and Bhat, 1981). For Goa, it is a first record.

15. Campylospora chaetocladia Ranzoni, 1953. Farlowia. 4: 373. (Fig. 14)

Conidial fungus, Hyphomycete. *Colonies* on MEA circular, regular, whitish to pale brown. *Mycelium* with hyaline, branched, septate, hyphae 1-2 μm wide. *Conidiophores* simple or rarely branched, hyaline, 20-30 μm long, 1.5-2.5 μm wide, each bearing a single conidium terminally. Conidium develops as an apical swelling of the conidiophore, becomes a spore primordium. *Conidia* with two parts: proximal half triangular, 3-4 septate, 8-12.5 μm high, 10-12 μm wide at the base; distal half allantoid, 3-4-celled, 9-13 μm long, 3.5-5 μm wide; appendages arising from end cells, setae like, 30-40 μm long.

Specimen examined: (i) On submerged leaf litter of *Terminalia paniculata* Roth., Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUBH-119, Sreekala K. Nair, 23-12-1999. (ii) Dried culture mat derived from single spore isolate, Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUFCC-119-a, Sreekala K. Nair. 23-12-1999. (iii) Dried foam slide, perenial stream, Tambdisurla, Goa, India, Herb. No. GUBH-120, Sreekala K. Nair, 8-7-2000.

The genus Campylospora was described by Ranzoni (1953), with chaetocladia as

type species. The fungus was reported from forest streams of Utter Pradesh, Karnataka, Kerala and Andhra Pradesh (Sridhar et al., 1992). This is the first report from Goa State.

16. Campylospora filicladia Nawawi,1974. Trans. Br. mycol. Soc. 63: 603-606. (Fig. 15)

Conidial fungus, Hyphomycete. *Colonies* effuse, white to creamish, circular, margin slightly wavy. *Mycelium* composed of hyaline, branched, septate hyphae 1-3 µm wide. *Conidia* differ from that of *C. chaetocladia* by presence of long, thin, filiform, hyaline appendages arising from apical ends of tetraradiate conidia. These 28-35 µm long appendages extend from the central primordium cell. The conidiophore and conidia are similar to that of *C. chaetocladia*.

Specimen examined: (i) Dried foam slide, seasonal stream, Anmod Ghat, GUBH-114, Sreekala K. Nair, 25-10-1999. (ii) Dried foam slide, seasonal stream, Molem Wildlife Sanctuary, Goa, India, Herb. No. GUBH-121, Sreekala K. Nair, 23-12-1999. (iii) Submerged leaf litter of *Xylia xylocarpa* Taub., Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUBH-122, Sreekala K. Nair, 24-8-2000.

The fungus was earlier reported from different streams of Karnataka, Kerala and Tamilnadu (Sridhar *et al.* 1992). Here, it is reported for the first time from Goa.

17. Canalisporium caribense (Hol.-Jech.and Mercado) Nawawi and Kuthub.1989.

Mycotaxon, 34: 479. (Fig. 76)

Colonies effuse, superficial. Sporododhia on natural substratum punctiform, minute, scattered, granular, black, glistening, up to 135 μm in diam. Conidiophores micronematous, fasciculate, simple or sparsely branched, smooth, hyaline, up to 25 μm long, 2-3.5 μm wide. Conidiogenous cells integrated, terminal, determinate, cylindrical,

often seen attatched to the base of the detached conidia. Conidia 18-25 x 13-15 x 5-8 μ m, acrogenous, solitary, smooth, thick-walled, broadly ellpsoidal to obovoid in surface view, comprising of a single, straight to slightly curved column of vertical septa, towards the peripharal wall of the conidia.

Specimen examined: On the submerged twig of *Careya arborea* Roxb., seasonal stream, Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUBH- 25. Sreekala K Nair. 24-8-2000.

18. Candelabrum brocchiatum Tubaki, 1975. Trans. Mycol. Soc. Japan. 16: 132-140.(Fig. 78)

Colonidial fungus, Hyphomycete. *Mycelium* immersed, formed of branched, septate, hyaline, thin-walled hyphae, 1.5-2.0 μm wide. *Conidiophores* macronematous, mononematous, unbranched, straight, smooth, hyaline, up to 50 μm long, 1.5-2 μm wide. *Conidiogenous cells* terminal, monoblastic determinate, slightly inflated, branches dichotomoustly or rarely trichotomously, each branch then divides in a similar manner in all directions; each short and tubular branch delimited by a septum. By several divisions of branches, final division widens to a spherical cell, which contribute to form conidium. *Conidia* solitary, acrogenous, spherical, hyaline to pale cream-coloured, 80-120 μm in diam. Terminal branch inflated with a short dichotomy, 3-4 μm wide, 4-5 μm long, each of which is ornamented with minute tubercles.

Specimen examined: On submerged twigs, seasonal stream, Bondla Wildlife Sanctuary, Goa, India, Herb. No. GUFCC-102, Sreekala K. Nair, 21-10-2000.

Originally the fungus was collected from Japan on submerged decaying leaves, on rotten wet twigs and submerged wood block (Tubaki, 1975). This is the first record for

India.

19. Centrospora acerina (Hartig) Newhall. An illustrated Guide to Aquatic and Water-Borne Hyphomycetes. C. T. Ingold, 1975. pp. 80-81. (Fig. 17)

Conidial fungus, Hyphomycetes. *Colonies* pale brown, slowing growing, attaining a diam of 1.7 cm in 15 days. Aerial mycelium offwhite, raised at the centre, margin broad, reverse dark brown, composed of septate, hyaline, flexuous, thin walled hyphae up to 1-1.5 µm wide. *Conidiophores* hyaline to very pale brown, simple to sparcely branched, 10-52 x 2-4 µm. *Conidiogenous cells* terminal or lateral, often variously curved, thinwalled, sometimes with convoluted apices. *Conidiogenous cells* blastic. *Conidia* acropleurogenous in group, polyblastic, on inconspicuous denticles, 4.5-10 x 1-1.5 µm, with an elongated tail-like region at the base.

Specimen examined: Dried submerged leaves of *Dillenia indica* Linn., seasonal stream, Bondla Wild Life Sanctuary, Goa, India, Herb. No. GUBH. No. 124, Sreekala K. Nair, 21-10-2000.

This is the first record of the fungus from India.

20. Ceratosporium sp. (Fig. 27)

Conidial fungus, Hyphomycete. *Colonies* effuse, brown to black. *Mycelium* partly superficial, composed of 4-5 µm thick hyphae. Vegetative hyphae branched, septate, pale brown, 2-6 µm wide. *Conidiophores* micronematous, flexuous, irregularly branched with the branches anastomosing, brown, smooth. *Conidiogenous cells* integrated, intercalary, monoblastic, determinate, cylindrical, denticulate; denticles cylindrical to broad conical. *Conidia* solitary, pleurogenous, branched, Y-shaped, mid to dark brown, smooth, with a

swollen, pyriform central cell and 2 divergent, 2-5 septate arms, each measuring 18-30 μ m long, 4-8 μ m at the centre and 2 μ m at the tip. The fungus sporulated well in MEA medium.

Specimen examined: Submerged leaf litter of *Terminalia paniculata* Roth., seasonal stream, Cotigao Wild Life Sanctuary, Goa, India, Herb. No. GUFCC-113. Sreekala. K. Nair. 23-12-1999.

Conidia of *Ceratosporium* sp. were also reported earlier from aquatic foam (Matsushima, 1975). Here it is a first record from India.

21. Chaetendophragmia triseptata Matsushima, 1975. Icones Microfungorum. pp. 25.

(Fig. 19)

Conidial fungus, Hyphomycete. *Colonies* effuse, pale to mid brown. *Mycelium* immersed in the substratum, consisting of branched, septate, hyaline to pale brown hyphae 1-3.5 µm wide. *Conidiophores* solitary, erect, simple, septate, 50-150 µm long, 3-4 µm wide, successively proliferating, moderately brown, with basal darker region and paler tip. *Conidia* solitary, pale brown, triangular, 3-septate, 22-28 x 5.5-7 µm, with a basal truncate region, 2.5-3.5 µm wide, with apical region consisting of 2-5 hyaline appendages 22-32 µm long. The central appendage is erect and others pointing divergently.

Specimen examined: (i) Dried foam slide from the seasonal streams of Molem Wildlife Sanctuary, Goa, India, Herb. No. GUFCC-126. Sreekala K. Nair. 18-9-1999. (ii) Dried submerged leaf litter collected from a perennial streams of Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUBH-127, Sreekala K. Nair, 24-8-2000. (iii) Dried foam slide from the seasonal streams of Bondla Wildlife Sanctuary, Goa, India, Herb. No.

GUBH-128, Sreekala K. Nair, 25-10-2000.

The fungus is commonly seen in the decaying leaves and twigs in the forests.

22. Colletotrichum dematium (Pers. et Fr.) Grove. (Ref. Matsushima, 1975) (Fig. 77)

Colonies effuse, pale brown. Acervuli with setae around its margin. Setae simple, subulate, septate, pale brown, 80-130 μm long, 3.5-5 μm wide. Conidiophores are in dense fascicles, pale brown, branched, ending in phialides. Conidiogenous cells blastic, hyaline, 12-26 x 2.5-4 μm. Conidia falcate, hyaline, 18-24 x 3.5-4.5 μm, pale brown in mass.

Specimen examined: (i) Dried foam slide, seasonal stream, Chorlem Ghat, Goa, India, Herb. No. GUBH-65, Sreekala K. Nair, 2-7-1999. (ii) Dried culture mat, seasonal stream, Chorlem Ghat, Goa, India, Herb. No. GUBH-65-a, Sreekala K. Nair, 2-7-1999.

23. Condylospora spumigena Nawawi, 1976. Trans. Br. mycol. Soc. 66: 363-365.

(Fig. 21)

Conidial fungus, Hyphomycete. *Colonies* on natural substratum whitish, on CMA at 25°C attaining 6 mm diam in 5 weeks, white, smooth, with the middle area turning light brown with age. *Mycelium* mostly superficial, consisting of smooth, branched, septate, hyaline, 1.5-2 μm wide hyphae. *Conidiophores* single, ascending to erect, semi-macronematous, short at first, but becoming longer with progressing conidiogenesis, cylindrical, hyaline, flexuous, up to 52 μm long, with short, cylindrical, truncate, up to 10 denticles 1.5-2 μm long and 0.5 μm wide. *Conidia* cylindrical, recurved in the middle, hyaline, thin-walled, guttulate, with proximal part measuring 45-50 μm long, 1 μm wide at the base, broadening to 2-2.5 μm long.

Specimen examined: (i) Submerged leaf litter of *Saraca asoca* collected from a seasonal stream at Molem Wildlife Sanctuary, Goa, India, Herb. No. GUBH-125, 18-8-1999. (ii) Dried foam slide collected from a seasonal stream of Bondla Wildlife Sanctuary, Goa, India, Herb. No. GUBH-143, Sreekala K. Nair. 21-10-2000.

C. spumigena was earlier reported from streams of Western Ghat regions in Karnataka, Kerala and Tamil nadu (Subram. and Bhat, 1981; Sridhar et al. 1992). Here it is the first record from Goa State.

24. Curvularia lunata (Wakker) Boedijn. (Ref. Ellis, 1971; Matsishima, 1975). (Fig. 75)

Colonies on MEA effuse, brown, grey or black, hairy, irregular, attaining a diam. of 6.2 cm; reverse of the colony brown. *Mycelium* superficial, composed of septate, thinwalled, smooth, hyaline, hyphae 2-3 µm wide. *Conidiophores* mononematous, straight or flexuous, often geniculate, brown, smooth, septate, 3.2 x 60-100 µm. *Conidiogenous cells* polytretic, integrated, terminal, sympodial, intrcalary, swollen at the tip, cicatrized. *Conidia* solitary, simple, often curved, clavate, ellipsoidal, broadly fusiform, pyriform with mostly 3 transverse sepatate, dark brown, 9.4-12.5 x 4.7-6.4 µm.

Specimen examined: (i) On root tissues of *Bambusa arundinaceae* (Retz.) Willd., seasonal stream, Bondla Wildlife Sanctuary, Goa, India, Herb. No. GUBH-30, Sreekala K. Nair, 12-8-2000. (ii) Dried culture mat derived from the culture of the fungus, Bondla Wildlife Sanctuary, Goa, India, Herb. No. GUBH-30-a, Sreekala K. Nair, 12-8-2000.

25. Cylindrocarpon sp.1 (Fig. 79)

Colonies on MEA effuse, white to pale brown, regular, 6.6 cm diam. in 10 days, reverse of the colony brown. *Mycelium* composed of thin-walled, loose, white, smooth, septate, branched, hyaline hyphae, 1.5 µm wide. *Conidiophores* mononematous, straight

to flexuous, branched, hyaline, septate, smooth, $3.5-15.5 \times 1.5 \mu m$. Conidiogenous cells phialidic, integrated, terminal, cylindrical. Conidia simple, rounded to slightly elongated, aseptate, hyaline, smooth, $5-10.5 \times 1.5-2.5 \mu m$.

Specimen examined: (i) Dried foam slide, seasonal stream, Alorna, Goa, India, Herb. No. GUBH-31, Sreekala K. Nair, 20-7-2000. (ii) Dried culture mat, seasonal stream, Alorna, Goa, India, Herb. No. GUBH-31-a, Sreekala K. Nair, 20-7-2000.

26. Cylindrocarpon sp. 2

(Fig. 82)

Colonies on MEA effuse, creamish white in colour, with sparcely superficial mycelium; reverse of the colony offwhite. Mycelium composed of hyaline, septate, branched hyphae, 1-2 μm wide. Conidiophores solitary, erect, simple, erect or flexuous, septate, unbranched, 20-50 μm long and 1-3 μm wide. Conidiogenous cells monophialidic. Conidia cylindrical, smooth, hyaline, elongated, 1-2-septate, 28-38 μm long, 4-6 μm wide.

Specimen examined: (i) On submerged leaf litter of *Dillenia indica* Linn., seasonal stream, Cotigao Wildlife Sancturary, Goa, India, Herb. No. GUBH-32, Sreekala K. Nair, 11-4-1999. (ii) Dried culture mat, seasonal stream, Cotigao Wildlife Sancturary, Goa, India, Herb. No. GUBH-32-a, Sreekala K. Nair, 11-4-1999.

27. Cylindrocarpon sp. 3

(Fig. 83)

Colonies on MEA effuse, white, attaining a diam. of 4.6 cm in 10 days; reverse of the colony white. *Mycelium* immersed, composed of septate, branched, hyaline hyphae, 0.5-2.5 µm wide. *Conidiophores* solitary, erect, simple, branched, 30-50 µm long, 2-3 µm wide. *Conidiophores* phialidic, hyaline, 15-30 µm long, 1-3 µm wide, with 1-2

 μm wide apical region. *Conidia* cylindrical, hyaline, slightly curved, 3-5 septate, 15-30 x 3-4 1-2 μm .

<u>Specimen examined</u>: On submerged leaf litter of *Bambusa arundinaceae* (Retz.) Willd., seasonal stream, Chorlem Ghat, Goa, India, Herb. No. GUBH-33, Sreekala K. Nair, 4-10-2000.

28. Cylindrocladiopsis lagerstraemiae Yen, 1979. Mycotaxon, 8: 233-237. (Fig. 84)

Colonies on MEA effuse, pale brown in colour, attaining a diam. of 3.5 cm in 10 days; reverse of the colony cream. Mycelium sparcely superficial, composed of hyaline, septate, branched, thinwalled hyphae, 2.5-3 μm wide. Conidiophores solitary, erect, hyaline, 18-45 μm long, with the apical region branching and ending into 5-18 conidiogenous cells. Conidiogenous cells cylindrical, hyaline, blastic, 12-20 μm long, 2-4 μm wide. Conidia hyaline, cylindrical, elongate, 3-5 septate and 20-30 x 2-4 μm.

Specimen examined: On submerged leaf litter of *Lagerstroemia lanceolata* Wall. ex Wt. and Arn., seasonal stream, Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUBH-34, Sreekala K. Nair, 23-12-1999.

29. Cylindrocladium cilhounii Peerally var. macroconidialis Crous, Wingfield and Alfenas. 1993. Mycotaxon. 46: 222. (Fig. 81)

Colonies on MEA effuse, reddish brown in colour, irregular, with rhizoidal margin and fast growing attaining a diam. of 3 cm in 7 days, reverse of the colony brown.

Mycelium immersed. Conidiophores arise from vegetative hyphae, thin-walled and hyaline to pale brown, 35-50 µm long, 2-4 µm wide at the base. Conidiogenous cells monophialidic, hyaline with collars, cylindrical, 15-25 µm long, appearing in groups at the

sub-apical region of the conidiophore. *Conidia* large, occasionally pale brown to golden yellow, rod shaped, cylindrical, elongate, 5-8 septate, 20-45 μm long, 2-5 μm wide; stipe sterile, septate, hyaline, 30-40 μm long, terminating in a clavate to spathulate vescicle.

Specimen examined: (i) Dried foam slide, seasonal stream, Bondla Wildlife Sanctuary, Goa, India, Herb. No. GUBH-36, Sreekala K. Nair, 14-2-1999. (ii) As root endophyte of *Bambusa arundinaceae* (Retz.) Willd., Bondla Wildlife Sanctuary, Goa, India, Herb. No. GUBH-37, Sreekala K. Nair, 14-2-1999. (ii) Dried culture mat derived from the culture, Bondla Wildlife Sanctuary, Goa, India, Herb. No. GUFCC-37-a, Sreekala K. Nair, 14-2-1999.

30. Cylindrocladium sp.

(Fig. 80)

Colonies on MEA effuse, creamish to pale brown in colour, moderatily fast growing, attaining a diam. of 3.7 cm in 10 days. *Mycelium* immersed, composed of pale brown, septate, branched hyphae. *Conidiophores* branched, hyaline, penicillate, branching upto six times with a total length of 20-40 μm. *Conidiogenous cells* monophialidic, hyaline, with collars, doliiform to cylindrical, occurring in groups of 4-6 per branch, 10-15 μm long, 2-4 μm wide. *Conidia* hyaline, cylindrical, thin-walled, straight, with 2-5 septa, 8-15 x 1.5-2.5 μm, arranged in cylindrical conidial clusters. Each conidiophore ends in a sterile cylindrical apical region arising from the penicillate cluster of phialides, which are ellipsoidal to clavate towards the apex, up to 45 μm long.

Specimen examined: (i) Dried foam slide, seasonal stream, Cudnem, Goa, India, Herb. No. GUBH-35, Sreekala K. Nair, 4-10-2000. (ii) Dried culture mat, seasonal stream, Cudnem, Goa, India, Herb. No. GUBH-35-a, Sreekala K. Nair, 4-10-2000.

31. Dactylella ellipsospora Grove. Mycologia 29: 492.

(Fig. 22)

Conidial fungus Hyphomycete. Mycelium hyaline, septate and branched. Conidiophores mononematous, simple, erect or flexuous, hyaline, thin-walled, 50-180 μ m long, 1-2 μ m wide. with a single conidium borne at its tip. Conidiogenisis blastic. Conidia hyaline, dome shaped, hyaline, pointed at both ends, 6-15 x 5-8 μ m.

Specimen examined: On submerged leaf of *Hopea ponga* (Dennst.) Mabb, seasonal stream, Molem Wildlife Sanctuary, Goa, India, Herb. No. GUBH-68, Sreekala K. Nair, 25-12-2000.

32. Dactylaria aquatica sp. nov.

(Fig. 24)

Conidial fungus, Hyphomycete. *Colonies* effuse, circular, regular, cream to light grey, attaining a diam. of 4.6 cm in 10 days; reverse of the colony grey. *Mycelium* sparcely superficial, made of thin, hyaline, septate, branched, hyphae 1-2 µm wide. *Conidiophores* arising from vegetative hyphae, solitary, simple, 1-5 septate, rarely branched, pale brown, 15-40 µm long, 3-5 µm wide. *Conidiogenous cells* polyblastic, denticulate with 4-5 denticles per cell; denticles peg like, arranged in a row, resulting in broad hook-like curved conidiogenous cells 3-5 µm wide and 2-4 µm long. *Conidia* solitary, cylindrical, dry, elongate, hyaline, 2-5 septate, 25-40 µm long, 2-3 µm wide at the centre, with truncate base and obtuse apex.

Specimen examined: On submerged live root of *Hopea ponga* (Dennst.) Mabb., isolated as root endophyte, seasonal stream, Molem Wildlife Sanctuary, Goa, India, Herb. No. GUFCC-163, Sreekala K. Nair, 20-9-2000.

The genus Dactylaria Sacc. has accommodated so far 55 described species. None

of these show the recurved nature of conidiogenous apparatus and affinity for an exclusive aquatic habitat as seen in the present species.

33. *Dactylaria* sp. (Fig. 23)

Conidial fungus, Hyphomycete. *Colonies* effuse, regular, cream to pale brown, attaining a diam. of 3.8 cm in 10 days; reverse of the colony brown. *Mycelium* partly superficial, made of thin, hyaline, septate, branched, hyphae 1.5-2.5 μm wide. *Conidiophores* solitary, simple, un-branched, hyaline, 0-septate, 10-15 μm long, 3-10 μm wide. *Conidiogenous cells* polyblastic, denticulate with 4-8 denticles per cell; denticles 2-4 μm wide, 3-8 μm long. *Conidia* solitary, cylindrical, dry, elongate, hyaline, 0-septate, 30-50 μm long, 2-5 μm wide at the centre with a truncate base and obtuse apex.

Specimen examined: On submerged leaf litter of *Dillenia indica* Linn., seasonal stream, Molem Wildlife Sanctuary, Goa, India, Herb. No. GUFCC. No. 190, Sreekala K. Nair, 25-12-2000.

34. *Dendrospora erecta* Ingold, 1944. *Trans. Br. mycol. Soc.* **26**: 104-107. (Fig. 25)

Conidial fungus, Hyphomycete. *Colonies* effuse, white, slow growing in MEA, attaining a diam of 1.7 cm in 12 days; reverse of the colony colourless to white. *Mycelium* immersed, composed of smooth, septate, branched, hyaline hyphae 1-2 µm wide. *Conidiogenous cells* solitary, terminal, integrated, monoblastic, hyaline, 5-15 µm long, 2-4 µm wide. *Conidia* hyaline, solitary, terminal, branched in 3-15 levels, with a straight main axis, 4-12-septate 30-200 µm long and laterals, up to 125 µm long, showing branching at the base.

Specimen examined: (i) Dried foam slide, seasonal stream, Bondla Wildlife

Sanctuary, Goa, India, Herb. No. GUBH-169, Sreekala K. Nair, 21-10-2000. (ii) Dried culture mat derived from single spore isolate of the fungus, Cotigao Wild Life Sanctuary, Goa, India, Herb. No. GUFCC-170, Sreekala K. Nair, 17-12-2000.

So far 11 species were reported in this genus from different parts of the world.

This is the first record from Goa.

35. **Dendrospora** yessemreddia Sreekala et Bhat, 2002. Frontiers in Microbial Biotechnology and Plant Pathology. pp. 295-298. (Fig. 26)

Conidial fungus, Hyphomycete. *Colonies* effuse, white, moderately slow growing, attaining a diam. of 2.3 cm in MEA after 10 days; reverse of the colony colourless to white. *Mycelium* immersed, composed of smooth, septate, branched, colourless hyphae 1-2 µm wide. *Conidiophores* mononematous, simple, erect, flexuous, hyaline, smooth, 0-3-septate, unbranched, 8-40 x 2-3 µm. *Conidiogenous cells* monoblastic, integrated, terminal, truncate at the tip after secession of conidium. *Conidia* solitary, hyaline, smooth, septate, with several (up to 25) lateral branches developing closely from a flexible main axis and resulting into a flat fan-shaped structure, generally seen afloat on surface of water; main axis elongated, flexible, 2-24-septate, 30-280 x 2-4 µm; laterals up to 200 µm long, sometimes branching further into secondary laterals, inserted to the main through a narrow constriction.

The fungus when grown on PDA and in CMA showed a slightly faster growth rate, i.e. 3.7 cm diam in 10 days. In PDA, the colony was flat and did not sporulate whereas the sporulation was moderately good both in MEA and CMA after 10 days of incubation.

Specimen examined: Dried culture mat derived from single spore isolation of the

fungus, Bondla Wild Life Sanctuary, Goa, India, Herb. No. GUFCC-159, Sreekala K. Nair, 15.8.1999.

36. **Dendrosporium lobatum** Plakidas and Edgerton ex Crane, 1972. *Trans. Br. mycol.*Soc. 58: 423-426. (Fig. 30)

Conidial fungus, Hyphomycete. *Colonies* effuse, regular, circular with slightly wavy margin, white to creamy white, center somewhat raised, aerial growth scanty. Older cultures greyish. *Mycelium* septate, hyaline when young, later becoming closely septate with brown swollen cells 10-15 µm wide. Sporulation profuse in young cultures and scanty in old cultures. *Conidiophores* of same thickness and structure as the mycelium, hyaline, developing from mycelial threads, septate, branched, variable in length. *Conidia* hyaline, deeply constricted so that there are usually three lobes on each side, the basal lobes being the largest, flattened to slightly concave or convex, pointed at the apex, pedicillate, uniseptate, the septum just above the basal lobes. The two cells being of unequal size, borne on slender sterigmata, one to several on each conidiophore, 3.5-4.5 x 6.8-10.2 x 10.2-15.3 µm, average about 3.9 x 7.8 x 12.2 µm.

Specimen examined: (i) On submerged live roots of *Hopea ponga* (Dennst.) Mabb. as root endophyte, Molem Wildlife Sanctuary, Goa, India, Herb. No. GUFCC-132, Sreekala K. Nair, 29-12-1999. (ii) Dried foam slide, seasonal stream, Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUFCC-165, Sreekala K. Nair, 24-8-2000.

There is no previous record of this fungus from India.

37. Dichotomophthoropsis aquatica sp. nov. (Fig. 31)

Conidial fungus, Hyphomycete. *Colonies* effuse, white to olivaceous brown in MEA, circular, regular, attaining a diam of 4.5 cm in 10 days; reverse of the colony

brown. *Mycelium* partly immersed, consisting of hyaline to brown, smooth, septate, branched hyphae 2-6 μm thick. *Conidiophores* macronematous, flexuous, septate, branched, dichotomously or trichotomously forked, subhyaline, smooth, up to 150 μm long, 3-7 μm thick. *Conidiogenous cells* polytretic, lobed, terminal and integrated. *Conidia* dry, solitary, borne one each at the tip of each lobe of the conidiogenous cell, simple, helicoid, multiseptate, often constricted at the septa, slightly verucose, pigmented, 150-350 x 10-20 μm.

Specimen examined: On submerged live root of aquatic grass *Glaberus* sp., isolated as root endophyte, Cudnem, Goa, Herb. No. GUFCC-166, Sreekala K. Nair, 4-10-2000.

Besides type species, *Dichotomophthoropsis nympharum* (Rand) M. B. Ellis (Ellis, 1976), the other known species is *D. safeullensis* Sheeba et al. (1991). *D. aquatica* differs from the earlier described species by its larger conidia.

38. *Dictyochaeta assamica* (Agnihotrudu)Hughes et Kendrick, 1968. *N. Z. Jrl. Bot*, 6: 334-335. (Fig. 32)

Colonies effuse, greyish brown to blackish brown, hairy. Setae straight or curved, brown to dark brown, mostly fertile towards the apex with a number of persistent collaretes, smooth, thick-walled, up to 300 μ m long, 4-6 μ m wide just above the swollen base. Conidiophores solitary or in small groups, often associated with setae, closely septate, pale brown, smooth, up to 100 μ m long, 3-5 μ m thick. Conidiogenous cells polyphialidic, collarettes funnel-shaped with thin walls. Conidia fusiform, curved, hyaline, smooth, 10-15 x 2-3 μ m, with a setula 5-10 μ m long at each end.

Specimen examined: (i) Dried foam slide, seasonal stream, Anmod Ghat, Goa, India, Herb. No. GUBH-133, Sreekala K. Nair, 25-10-1999. (ii) On submerged live root of *Bambusa* sp. as an endophyte, Bondla Wildlife Sanctuary, Goa, India, Herb. No. GUFCC-134, Sreekala K. Nair, 12-8-2000. (iii) Dried culture mat of the fungus, Bondla Wildlife Sanctuary, Goa, Herb. No. GUFCC-134-a, Sreekala K. Nair, 12-8-2000.

The fungus is a terestrial form commonly found on leaf litter in the forests of tropical countries (Ellis, 1971; Matsushima, 1975), but often encountered in stream waters.

39. Diplocladiella scalaroides Arnaud ex Ellis, 1976. More Dematiaceous Hyphomycetes. p.229. (Fig. 20)

Conidial fungus, Hyphomycete. *Colonies* effuse, brown, shortly hairy. *Mycelium* mostly immersed in substratum, composed of branched, septate, pale brown, smooth hyphae 2-2.5 μm wide. *Conidiophores* mononematous, simple, geniculate, pale to mid brown, smooth, 25-25 μm long, 3-4 μm thick. *Conidiogenous cells* polyblastic, integrated, sympodial, geniculate, cicatrized. *Conidia* triangular, branched, 2-horned, pale to mid brown, smooth, with horns mostly 2-septate with a paler small terminal cell, 15-30 μm long, basal cell 2-4 x 2-3 μm.

Specimen examined: (i) Dried foam slide, seasonal stream, Chorlem Ghat, Goa, India, Herb. No. GUBH-112, Sreekala K. Nair, 2-7-1999. (ii) Submerged leaf litter of *Terminalia paniculata* Roth., Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUBH-135, Sreekala K. Nair, 19-8-1999.

This is a terrestrial fungus, also encountered in aquatic habitats. Reports of the fungus in foam samples from Western Ghat forest streams are known. (Sridhar and

Kaveriappa, 1989).

40. Diploospora indica Sreekala et Bhat, 2001. Mycotaxon, 80: 101-104. (Fig. 33)

Conidial fungus, Hyphomycete. *Colonies* consisting of concentrically zonate hyphae alternating with conidial mass, effuse, white, with smooth margin, slow growing, reaching 0.5-1.0 cm diam. in 10 days, reverse of the colony colourless to pale brown. *Mycelium* partly superficial, composed of smooth, colourless hyphae 1.5-2 µm wide. *Conidiophores* terminal or lateral, mononematous, erect, flexuous, smooth, moderately thick-walled, 2-7-septate, branched, pale brown, 75-125 x 3-5 µm. *Conidiogenous cells* integrated, polyblastic, 10-22 x 3-4 µm, apically truncate after secession of conidia. *Conidia* in long, acropetal, simple to proximally branched, false chains, fusiform, narrow and truncate at both ends, dorsi-ventrally flat, smooth-walled, aseptate, hyaline to very slightly pigmented, pale brown in mass, 12-33 x 3-4 µm; ramoconidia aseptate, truncate at the base, with 2-5 terminal flat scars.

Specimen examined: On dried culture mat derived from single spore isolate from submerged leaf litter of *Hopea ponga* (Dennst.) Mabb., Cotigao Wildlife Sanctuary, Goa, India, Herb. No. IMI 384550 (1), GUFCC-161, Sreekala K. Nair, 19-7-1999.

41. Endophragmia inaequiseptata Matsushima, 1975. Icones Microfungorum. p.69, Pl.139. (Fig. 73)

Conidial fungus Hyphomycete. *Colonies* effuse, dark brown to black. *Mycelium* immersed. *Conidiophores* mononematous, unbranched, straight or flexuous, brown, smooth, 80-160 µm long, 5-7 µm wide; wall of the lower part of the conidium remain attatched to the apex of the conidiophore forming a cup and the conidiophore then proliferates. *Conidiogenous cells* monoblastic, integrated, terminal and cylindrical.

Conidia cylindrical, with rounded apex and truncate base, 3-septate, dark brown, 18-26 x 5-6.5 µm.

Specimen examined: On the submerged twig of *Dillenia indica* Linn., seasonal stream, Molem Wildlife Sanctuary, Goa, India, Herb. No. GUBH-41, Sreekala K. Nair. 25-12-2000.

42. Flabellospora crassa Alasoadura, 1968. Nova Hedwigia, 15:415-18. (Fig. 34)

Conidial fungus, hyphomycete. *Colonies* white, very pale creamy near the centre, margin diffuse, regular, very slow growing, attaining a diam. of 1.5 cm in 12 days. *Mycelium* immersed, with thin, hyaline, septate, hyphae 1.5-2 μm wide. *Conidiophores* on mycelial hyphae, apical or lateral, prostrate, mononematous, simple, septa indistinct, 20-30 μm x 3-4 μm. *Conidiogenous cells* apical, integrated, monoblastic with naroow detachment scars. *Conidia* acrogenous, solitary, staurosporous with 3-6 radiating arms, 30-75 μm long, arising from a central main axis 5-10 μm long; radiating arms 5-7 μm wide.

Specimen examined: (i) Dried foam slide, seasonal stream, Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUFCC-137, Sreekala K. Nair, 17-12-2000.

This fungus was reported earlier from Uttar Pradesh and Karnataka (Sridhar, et al. 1992). This is the first record from Goa State.

43. Flabellospora multiradiata Nawawi, 1976. Trans. Br. mycol. Soc. 66(3): 543-547.

(Fig. 38)

Conidial fungus, Hyphomycete. *Colonies* on MEA, white to pale cream, attaining a diam. of 3 cm in 15 days; reverse of the colony cream to pale brown. *Mycelium* composed of branched, septate, hyaline, smooth, 1-2 µm wide hyphae. *Conidiophores* arising

laterally from prostrate hyphae, hyaline, smooth, 6-20 μ m long, 3-4.5 μ m wide. *Conidia* holoblastic, developing at apical end of conidiogenous cells, star-like, with main axis 10-13 μ m long, 1.5-2 μ m wide, with 10-30 radiating arms arising simultaneously from the bulbous tip of main axis; each arm 10-15-septate, 55-100 μ m long, 4-5 μ m wide; bulbous tip of main axis 5-6 μ m diam.

Specimen examined: (i) Dried foam slide, seasonal stream, Molem Wildlife Sanctuary, Goa, India, Herb. No. GUBH-136, Sreekala K. Nair, 18-8-1999. (ii) Dried foam slide, seasonal stream, Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUBH-145, Sreekala K. Nair, 24-8-2000. (iii) Dried culture mat derived from single spore isolate of the fungus. Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUFCC-145-a, Sreekala K. Nair, 24-8-2000.

The conidia of *F. multiradiata* are the most attractive of among the aquatic hyphomycetes. One would invariably encounter the conidia of this fungu if samples out foam from a clean running water in the forests of Western Ghats. Although the fungus was previously reported from Kerala, Karnataka and Uttar Pradesh (Sridhar et al. 1992), here it is the first record from Goa State.

44. Flabellospora verticillata Alasoadura, 1968. Nova Hedwigia, 15: 419-21. (Fig. 37)

Conidial fungus, Hyphomycete. *Colonies* white on MEA, attaining a diam. of 3.4 cm in 15 days; reverse of the colony offwhite. *Mycelium* composed of branched, septate, hyaline, smooth, hyphae 1-2 µm wide. *Conidiophores* arising from the vegetative hyphae, inconspicuous, hyaline. *Conidiogenous cells* monoblastic. *Conidia* multiradiate, hyaline, consisting of a main axis and 6-8 radiating arms. Main axis smooth, 2-5 septate, hyaline,

14-30 μ m long, 1.5-2 μ m wide, terminating in an obclavate tip 2-3 μ m diam; each arm 10-15-septate, hyaline, 60-100 μ m long, 4-5 μ m wide.

Specimen examined: (i) Dried foam slide, seasonal stream, Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUBH-138, Sreekala K. Nair, 19-8-1999. (ii) Dried foam slide, perennial stream, Tambdisurla, Goa, India, Herb. No. GUBH-146, Sreekala K. Nair, 8-7-2000. (iii) Dried foam slide, seasonal stream, Mollem Wildlife Sanctuary, Goa, India, Herb. No. GUBH-147, Sreekala K. Nair, 20-9-2000.

Conidia of *F. verticillata* resembles that of *F. multiradiata*, but differs in the number of radiating arms. The fungus has earlier been reported from other parts of Western Ghats (Sridhar et al. 1992), and it is a new record from Goa.

45. Flagellospora curvula Ingold, 1942. Trans. Br. mycol. Soc. 25: 339-417. (Fig. 35)

Conidial fungus, Hyphomycete. *Colonies* effuse, white to cream in MEA, attaining a diam. of 2.3 cm. in 12 days; reverse of the colony offwhite. *Mycelium* composed of hyamine, smooth, thin-walled, branched septate hyphae, 1-2 µm wide. *Conidiophores* mononematous, branched, straight or flexuous, 10-25 x 1-2.6 µm. *Conidiogenous cells* phialidic. *Conidia* hyaline, sigmoid, elongate, aseptate, thin-walled, 15-27 x 1-2.5 µm.

Specimen examined: (i) Dried foam slide, seasonal stream, Chorlem Ghat, Goa, India, Herb. No. GUBH-82, Sreekala K. Nair, 2-7-1999. (ii) Dried culture mat, seasonal stream, Chorlem Ghat, Goa, India, Herb. No. GUBH-82-a, Sreekala K. Nair, 2-7-1999. (iii) Submerged leaf litter of unidentified plant species, Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUBH-83, Sreekala K. Nair, 19-8-1999.

The fungus was reported earlier, from the Western Ghat forest streams (Sridhar et al, 1992).

46. *Gonytrichum macrocladum* (Sacc.) Hughes, 1951. *Trans. Br. mycol. Soc.*, **34**: 565-568 (Fig. 92)

Conidiophores macronematous, mononematous, straight or flexuous, upto 350 μm long, swollen to 5-8 μm at the dark brown base, 4-6 μm thick above, with a paler sterile upper part; collar hyphae arise from the lower half of the connidiophore, stipe, with up to 6 phialides each. Conidiogenous cells monophialidic, 12-21 x 3-4 μm, borne on collar branches, integrated and terminal, determinate, cylindrical with collarrettes. Conidio aggregated in slimy masses, often in columns, acrogenous and semi-endogenous, simple, ellipsoidal or sub-spherical, colourless to pale brown, smooth, 3-4.5 x 2-3 μm.

Specimen examined: (i) On live root tissues, *Bambusa arundinaceae* (Retz.) Willd., Bondla, Goa India, Herb. No. GUBH-46, Sreekala K. Nair, 21-10-2000. (ii) Dried culture mat derived from the pure culture of the fungus, Bondla, Goa India, Herb. No. GUFCC-46-a, Sreekala K. Nair, 21-10-2000.

47. *Hansfordia pulvinata* (Berk. and Curt.) Hughes, 1958, *Can. J. Bot.*, **36**:771. (Fig. 89)

Colonies grey or olivaceous grey. Mycelium partly superficial, composed of smooth, septate, branched and pale brown hyphae, 2-4 μm wide. Conidiophores repent or erect, viable in length, 2-5 μm thick, unbranched, pale to mid brown below; the paler upper part with a number of primary branches, terminating in a conidiogenous cell; primary branches extending into secondary branches. Conidiogenous cells subhyaline, smooth, up to 20 μm long, 1.5-4 μm thick. Conidia spherical or subspherical, colourless to pale brown, verruculose or minutely echinulate, 4-7 μm diam.

Specimen examined: On submerged leaves of *Xylia xylocarpa* Taub., seasonal stream, Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUBH-47, Sreekala K. Nair, 48. *Helicomyces roseus* Link, 1809. *Mag. Ges. Naturf. Freunde, Berlin*, 3:21. (Fig. 39)

Conidial fungus, Hyphomycete. *Colonies* effuse, offwhite to pale brown, superficial on the substratum. *Mycelium* composed of pale brown, septate, thin-walled hyphae 2-4 μm wide. *Conidiophores* mononematous, branched, straight, 5-10 μm long, 3-6 μm wide, arising directly on vegetative hyphae, denticulate, with dark scars on conidial succession. *Conidia* solitary, dry, simple, helicoid, 2-3 times coiled, hyaline, smooth, multiseptate, 240-320 μm long, 4-6 μm wide, 17-20 μm in diam, rounded at the tip and truncate at the base.

Specimen examined: (i) Dried foam slide, seasonal stream, Chorlem Ghat, Goa, India, Herb. No. GUFCC-111, Sreekala K. Nair, 2-7-1999. (ii) Dried foam slide, seasonal stream, Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUBH-153, Sreekala K. Nair. 23-12-1999. (iii) Dried culture mat, seasonal stream, Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUBH-153-a, Sreekala K. Nair. 23-12-1999.

This is a terrestrial form often encountered in foam samples. Agar blocks containing mycelia when submerged in sterile distilled water conidia were readily formed. This is the first report of the fungus from aquatic habitat.

49. *Helicosporium* sp. 1

(Fig. 40)

Colonies effuse, olivaceous brown, delicately hairy. Mycelium partly immersed, partly superficial, composed of branched, smooth-walled, pale brown hyphae 3-5.5 μm wide. Conidiophores arising singly or in groups from superficial hyphae, mononematous, simple or very rarely branched, straight or slightly flexuous, erect, smooth, thick-walled,

2-6-septate, pale yellowish to midbrown, paler towards the apex, 65-160 x 6-8 μm. *Conidiogenous cells* monoblastic, integrated, terminal and intercalary, cylindrical, bearing 1-3 broad lateral cylindrical pegs, 4-9 x 2.5-5 μm. *Conidia* solitary, dry, simple, helicoid, 2-3 times coiled in one plane, hyaline, yellowish in mass, guttulate, indistinctly 6-8-septate, smooth, obtuse at the apex, subtruncate and narrow at the base, 10-18 μm diam, 2-3 μm wide.

Specimen examined: (i) Dried foam slide, seasonal stream, Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUBH-110, Sreekala K. Nair, 17-12-2000.

50. Helicosporium sp. 2

(Fig. 41)

Colonies effuse, light brown. Mycelium partly superficial. Conidiophores mononematous, branched, straight to flexuous, very light brown, septate, smooth, 65-280 x 3-4 μm. Conidiogenous cells monoblastic, discrete, cylindrical, detrminate, denticulate, 1.5-2.5 x 1-1.5 μm. Conidia solitary, dry, simple, hyaline, multiseptae, helicoid, elongate, with each filament 200-500 μm long and 1.5 μm wide.

Specimen examined: (i) Dried foam slide, seasonal stream, Cuncolim, Goa, India, Herb. No. GUBH-109, Sreekala K. Nair, 27-9-1999. (ii) Dried submerged twig, seasonal stream, Bondla Wildlife Sanctuary, Goa, India, Herb. No. GUBH-148, Sreekala K. Nair, 14-2-2000.

51. Helicosporium sp. 3

(Fig. 42)

Colonies effuse, white to pale olive yellow, cottony. *Mycelium* partly immersed, partly superficial, composed of branched, smooth-walled, pale yellowish brown hyphae 2-3 µm wide. *Conidiophores* macronematous, mononematous, simple at first, branched

once or twice distantly at maturity, subulate, branches at right angles to the stipe, constricted towards the point of origin, straight or slightly flexuous, erect, smooth, thick-walled, septate, mid to dark brown in lower part, pale brown to subhyaline at the tip, 370-450 x 4-5 μm, tapering to 2 μm wide. *Conidiogenous cells* polyblastic, discrete, irregularly flask-shaped, thin-walled, hyaline, sympodial, denticulate, denticles short, cylindrical, narrow, produced laterally from the stipe, 5-9 x 3.5-5 μm. *Conidia* solitary, dry, simple, helicoid, 2-3 times coiled in one plane, hyaline, yellowish in mass, guttulate, 6 or more septate, smooth, obtuse at the apex, subtruncate at the base, 15-25 μm in diam, filaments 1.5-2 μm wide.

Specimen examined: On submerged wood, seasonal stream, Molem Wildlife Sanctuary, Goa, India, Herb. No. GUFCC-171, Sreekala K. Nair, 29-12-1999.

52. *Idriella angustispora* Morgan-Jones, 1979. *Mycotaxon*, **8(2)**: 402-410. (Fig. 86) *Colonies* effuse, brown, hairy. *Mycelium* partly superficial, partly immersed in the substratum, composed of branched, septate, hyaline to pale brown, smooth, 1.5-4 μm wide hyphae. *Conidiophores* macronematous, mononematous, crowded, arising from the superficial repent hyphae, main stipe unbranched, cylindrical, erect, straight, pale brown to brown, subhyaline towards the apex, septate, upto 40 μm long and 3-4 μm wide. *Conidiogenous cells* holoblastic, discrete in groups of three of four at the tip of the conidiophore stipe, sympodial or synchronous, cylindrical to ampulliform, upto 15 μm long, 2-3 μm wide. *Conidia* dry, acropleurogenous, falcate, continuous, hyaline, smoth, 1-septate, 29-32 x 1-1.5 μm.

Specimen examined: Submerged leaf litter of Xylia xylocarpa Taub., seasonal

stream, Alorna, Goa, India, Herb. No. GUFCC-49. Sreekala K. Nair, 20-7-2000.

53. *Idriella lunata* Nelson et Wilhelm (Ref: Matsushima, 1975) (Fig. 85)

Colonies in MEA, effuse, grey to blackish brown, attaining a diam. of 4.2 cm in 7 days, reverse pale brown. *Mycelium* partly superficial, with pale brown, septate and branched hyphae. *Conidiophores* macronematous, mononematous, unbranched, straight or flexuous, subulate, usually swollen at the base, geneculate towards the aped, pale brown, smooth, up to 35 μm long and 3-4 μm thick at the base, tapering to 2 μm at the apex. *Conidiogenous cells* polyblastic, integrated, terminal, sympodial, subulate, denticulate, denticles short and conical. *Conidia* solitary, dry, acropleurogenous, simple, falcate, hyaline, smooth, 0-septate, 7-13 x 2-2.5 μm.

Specimen examined: (i) Dried foam slide, seasonal stream, Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUFCC-48, Sreekala K. Nair. 24-8-2000. (ii) Dried culture mat derived from the single spore isolate of the fungus, Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUFCC-48-a, Sreekala K. Nair, 24-8-2000.

54. *Ingoldiella hamata* Shaw, 1972. *Trans. Br. mycol. Soc.* **59**: 255-259. (Fig. 43)

Conidial fungus hyphomycete. *Colonies* effuse, hyaline to white on MEA, circular margin, slow growing, with concentric zones, attaining a diam of 3 cm in 7 days; reverse of the colony colourless to white. *Mycelium* immersed, composed of smooth, septate, branched, colourless, thin-walled hyphae 1-2 µm wide. *Conidiophores* mononematous, simple, erect or flexuous, hyaline, smooth, 0-septate, unbranched, 5-20 µm long and 2-6 µm wide. *Conidiogenous cells* monoblastic, integrated, terminal. *Conidia* very large, hyaline, solitary, smooth, septate, branched, thin-walled, with 1-3 long lateral branches, arising closely from a flexible main axis, constricted at the branching; main axis, 4-12-

septate, constricted at the septa, $50-160~\mu m$ long, $3-10~\mu m$ wide, hooked at the tip; lateral arms 3-10-septate, $40-130~\mu m$ long, $3-10~\mu m$ wide, hooked at their apices. Septa of main axis and laterals possess clamp connections.

Specimen examined: (i) On submerged leaves of *Hopea ponga* (Dennst.) Mabb., Herb. No. GUBH-152, Sreekala K. Nair, 20-9-2000. (ii) Dried culture mat derived from single spore isolate of the fungus, seasonal stream, Molem Wildlife Sanctuary, Goa, India, GUFCC-152-a, Sreekala K. Nair, 20-9-2000.

55. Isthmotricladia britanica Descals, 1982. Trans. Br. mycol. Soc. 78: 416-417.

(Fig. 45)

Colonies pale greyish brown, circular diffuse margin, consisting of sparcely growing, white aerial mycelium. Conidiophores micronematous. Conidiogenous cells lateral, single, discrete, monoblastic, straight, 6-18 x 1-2 μm, on conidial detatchment scars truncate. Conidia solitary, acrogenous, staurosporous, with straight main body, slightly clavate, 3-6 septate, 21-27 x 2.5-3.5 μm; with 3-4 synchronously developing fusiform branches with acute apex, each 35-42 x 2.5-3.5 μm.

Specimen examined: (i) On the submerged leaves, seasonal stream, Bondla Wildlife Sanctuary, Goa, India, Herb. No. GUBH-108, Sreekala K. Nair, 9-8-1999. (ii) Dried foam slide, seasonal stream, Cotigao Wildlife Sanctuary, Goa, India, GUBH-149, Sreekala K. Nair, 17-12-2000.

This aquatic species was initially reported by Descals and Webster (1982) from Great Britain. It differs from *I. laeensis* Matsushima (1971) by the presence of 4 arms. This is the first record from India.

56. Isthmotricladia laeensis Matsushima, 1971. Bull. Nat. Sci. Mus. Tokyo. 14: 478-480. (Fig. 44)

Colonies effuse, white, slow growing, attaining a diam. of 1.2 cm in 10 days; reverse of the colony hyaline to white. Conidiophores arising from the vegetative hyphae, mononematous, micronematous, hyaline. Conidiogenous cells monoblastic, straight, 10-15 μm long, 1-2 μm wide. Conidia solitary, staurosporous, with a straight main axis from which three radiating arms arise. Each arm 5-10 septate, with an acute apex, 25-40 μm x 3-6 μm.

Specimen examined: (i) On submerged leaves of *Careya arborea* Roxb., seasonal stream, Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUBH-107, Sreekala K. Nair, 24-8-2000. (ii) Dried culture mat derived from single spore isolate of the fungus, seasonal stream, Molem Wildlife Sanctuary, Goa, India, GUFCC-171, Sreekala K. Nair, 20-9-2000.

This is the second report of the fungus from Western Ghats, India (Sridhar et al. 1992).

57. Kumbhamaya jalapriya Srikala et Bhat sp. nov. (Fig. 46)

(Etym. Specific epithet in Samskrit, jala (= water) priya (= loving).

Coloniae in cultura MEA planae, obiculatae, ordinatus, cinereus, in colorus olivacio- ad fusco-griseus, 4 cm diam. in 10 dies; marginis ex coloniae serratus, inversum atropunctatus. *Mycelium* perparce superficiale, partim immersum, compositum duo typus hyphae, primus ad secondarius. Ex hyphis primei 3-12 µm lat., crassitunicatus, brunneolus; ex hyphis secondarius 2-4 µm lat, tenuitunicatus, hyalinis, *Conidiophora* enatus hyphis primie, mononematosa, erecta, flexuousa, septata, crassitunicata, paniculata,

pallide brunnea vel brunnea, 6=18 μm longa, 3-5 μm lat. *Cellulae conidiogenae* monophialis, ampullaceus, pallide brunnis, 10-30 μm longa et 3-4 μm lat. ad basim, angustatus longicollus regionis 1=2 μm lat. *Conidiae* solitaria, falcata, vermiformis, cincinnatae, hyalina, aseptata, levia, 15-30 μm longa, 2-3.5 μm lat.

HOLOTYPUS, In culturis derivata radicis submerses, *Hopea ponga* (Denst) Mabb, IMI 386171, Molem Wildlife Sanctuary, Goa, India, Herb. No. GUFCC-172, 20.9.2000, leg. Sreekala. K. Nair.

Colonies on malt extract agar flat, circular, with serrated margin, olivaceous grey to dark green, attaining a diam. of 4 cm in 10 days; reverse of the colony black. *Mycelium* sparsely superficial, composed of two types of hyphae, i.e. primary and secondary; primary hyphae thick-walled, septate, constricted at the septa, medium brown, 3-12 μm wide; secondary hyphae thin-walled, septate, hyaline, 2-4 μm wide, developing from primary hyphae. *Conidiophores* mononematous, erect, flexuous, septate, thick-walled, branched, smooth, pale to medium brown, 6-18 x 3-5 μm, developing from primary hyphae. *Conidiogenous cells* monophialidic, flask-shaped, smooth, with a narrow neck, pale brown, 10-30 μm long, 3-4 μm wide at the base, 1-2 μm wide above, without collarette. *Conidia* solitary, falcate to vermiform, curled, aseptate, smooth, hyaline, up 15-30 μm long, 2-3.5 μm wide.

The genus *Kumbhamaya* Miriam and Bhat (2000) was established to accommodate dematiaceous hyphomycetous fungi with kettle-shaped phialides and phragmosporus conidia. Besides the type species, *K. indica* Miriam and Bhat, *K. goanensis* Maria and Bhat (2001) which differs from the type by its flexible and variedly

shaped phailides and conidia is known. *K. jalapriya* is an endophytic fungus. Besides, it is unique and differs from the former known species by production of aseptate and curved conidia and being aquaitc.

58. Lateriramulosa uniinflata Matsushima, 1975. Icones Microfungorum. pp. 92. Pl. 242. (Fig. 29)

Conidial fungus, hyphomycete. *Colonies* on MEA effuse, offwhite, slow growing attaining a diam of 2.3 cm in 10 days. *Mycelium* consists of sparcely superficial, branched, septate, hyaline, hyphae 1-2 μ m wide. *Conidiogenous cells* polyblastic, minutely denticulate, hyaline, 4-8 μ m long, 2-4 μ m wide. *Conidia* triradiate, with a main axis 6.5-10 x 1.5-2 μ m, with three lateral branches, each 7.5-12.5 x 3.5 x 4 μ m; lower two branches, swollen at the base and with an apical spike-like seta; upper one with a swollen base.

Specimen examined: On submerged leaves of *Careya arborea* Roxb, seasonal stream, Cotigao Wild Life Sanctuary, Goa, India, Herb. No. GUFCC-106, Sreekala K. Nair, 19-8-1999.

The fungus is well known from the forests streams of Western Ghats (Sridhar et al. 1992). This is the first record from Goa.

59. Lemonniera aquatica de Wild., 1894. Ann. Soc. Belge Micr. 18: 143-148. (Fig. 18)

Conidial fungus, Hyphomycete. *Colonies* effuse on MEA, offwhite to light brown, regular with serrate margin, attaining a diam. of 2.8 cm in 15 days. *Mycelium* sparcely immersed composed of thin-walled, hyaline, septate, branched hyphae. *Conidiophors* erect, hyaline, septate, branched, simple, 200-400 µm long, penicillus at the tip. *Conidiogenous cells* phialidic, hyaline, ampulliform to cylindrical, single to penicillus, up

to 3 per branch, 15-30 x 4-7 μ m. Conidial primordia initially spherical, later becoming ovate to tetrahedral with a distal arm developing more rapidly than the laterals; conidial release by cleavage at the basal septum. *Conidia* tetrahedral, with distinct sub-spherical central body 3-5 μ m diam, with arms 34-70 x 4-7 μ m, hyaline, 3-7-septate, with the distal arm slightly longer than the laterals.

Specimen examined: (i) On submerged leaf litter of Lagerstroemia lanceolata Wall. ex Wt. and Arn., seasonal stream, Chorlem Ghat, Goa, India, Herb. No. 105, Sreekala K. Nair, 2-7-1999. (ii) Dried foam slide, seasonal stream, Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUFCC-173, Sreekala K. Nair. 17-12-2000.

The fungus sporulates well after 3-5 days, when a block of agar containing mycelium immersed in sterile distilled water. *L. aquatica* has been reported earlier from U.P., Maharastra and Andhra Pradesh (Sridhar et al., 1992). This is the first record from forests of Western Ghats in Goa State.

60. Lunulospora curvula Ingold, 1942. Trans. Br. mycol. Soc. 25: 404-409. (Fig. 16)

Conidial fungus, Hyphomycete. *Colonies* effuse in MEA, whitish, regular, circular attaining a diam. of 1.7 cm in 10 days. *Mycelium* consists of hyaline, septate, branched hyphae 1-3 μm wide. *Conidiophores* elongated, erect or flexuous, thin-walled, hyaline, 25-35 μm long, 2-4 μm wide. *Conidiogenous cells* monoblastic, hyaline, smooth, erect, up to 10 μm long. *Conidia* cresent-shaped, inflated to 5-7 μm in the middle, tapering to 1.5 μm towards both the ends, with a conspicuous attachment scar just below the inflated region on the convex surface, hyaline, 60-100 μm long.

Specimen examined: (i) On submerged leaf litter of Dillenia indica Linn., seasonal

stream, Molem Wildlife Sanctuary, Goa, India, Herb. No. GUBH-101, Sreekala K. Nair, 18-9-1999. (ii) Dried foam slide, seasonal stream, Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUFCC-104, Sreekala K. Nair, 24-8-2000.

The colony remained sterile on MEA medium. The fungus sporulated well when leaves of *Dillenia indica* collected from the sampling site immersed in sterile water under laboratory conditions. The fungus has been reported from the forests of Western Ghats of Karnataka, Kerala, Tamil Nadu. Besides it has been reported from U.P. and Andhra Pradesh (Sridhar et al. 1992).

61. Mycoleptodiscus indicus (Sahni) Sutton, 1973. Mycologia 59: 970-975. (Fig. 49)

Conidial fungus, Hyphomycete. *Colonies* effuse, dark brown. *Mycelium* superficial, usually sparse, composed of dark brown, septate, brachhed, 3-8 μm wide hyphae. *Sporodochia* sometimes irregular in outline, later becoming confluent, up to 100 μm diam. *Conidiophores* indistinct. *Conidiogenous cells* phialidic, mostly ampulliform, sometimes compressed, 7-17 x 6-16 μm, with apical opening 2.5-5 μm diam. *Conidia* curved, hyaline, often guttulate, smooth, 0-septate, 11-18 x 5-7.5 μm, always with a setula at each end up to 10 μm long.

Specimen examined: (i) Dried foam slide, seasonal stream, Taleigoa, Goa, India, Herb. No. GUFCC-100, Sreekala K. Nair. 2-8-1999. (ii) Submerged leaves of unidentified plants, seasonal stream, Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUBH-174, Sreekala K. Nair, 17-12-2000.

This is the first record of *M. indicus* from aquatic habitat. Otherwise, the fungus has been reported earlier from leaf litter and as an endophyte from the forests of Goa (Miriam, 2000).

62. Nawawia filiformis (Nawawi) Marv. Trans. Br. mycol. Soc. 75 (2): 221-231.

(Fig: 47)

Conidial fungus, Hyphomycete. *Colonies* effuse, light grey. *Mycelium* partly superficial, composed of smooth, pale brown, septate, hyphae 1-2 µm wide. *Conidiophores* mononematous, erect or flexuous, unbranched, moderately dark brown, 20-40 µm long, 2-3 µm wide. *Conidiogenous cells* phialidic. *Conidia* spherical, thinwalled, 6-12 µm in diam., with 3-6, slender, filiform appendages 5-10 µm long, arising from the surface.

Specimen examined: (i) On submerged and decaying leaves, seasonal stream, Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUBH-175, Sreekala K. Nair, 23-12-1999. (ii) Dried foam slide, perennial stream, Tambdisurla, Goa, India, Herb. No. GUBH-176, Sreekala K. Nair, 8-7-2000.

Two additional spore types collected in foam samples of Molem, although were abundant in number, did not sporulate in culture. These spore types were distinct from *N. filiformis*. In Fig. 47-a, spores were with many radiating appendages (6-7) and granular in appearance. Tentatively it is referred as *Nawawia* sp. 1. Another spore type constantly with 3-4 appendages were assigned to *Nawawia* sp.2. (Fig. 47-b)

63. Parathozetella Sreekala et Bhat Gen. nov.

(Fig. 63)

(Etym. Para = similar)

Colonies white to yellowish, circular, regular. Mycelium immersed, composed of thin-walled, hyaline, septate, hyphae. Conidiomata sporodochial, scattered, solitary, discrete, superficial, slightly raised from the substratum, surmounted by greenish mass of slimy conidia, encircled by long, hyaline to light yellowish, thick-walled, septate setae.

Conidiophores compact, hyaline, septate, unbranched, thin-walled. Sterile erect, thin-walled, hyaline, septate, hyphae resembling paraphyses develop between the conidiophores. Conidiogenous cells monophialidic, simple, thin-walled, cylindrical, smooth. Conidia bean-shaped, hyaline, slightly curved, ellipsoid, guttulate, with thin, filiform, appendages developing from the curved edge.

Several genera of sporodochial moniliaceous Hyphomycetes are known (Carmaichael et al., 1980). Of these, *Thozetella* Kuntze, *Tubercularia* Tode ex Persoon and *Volutella* Tode are similar to the fungus described above, in view of its phialidic conidiogenous cells and appeandaged conidia. However, presence of paraphyses-like sterile soft hyphae, stipitate stiff setae and curved apendaged conidia together in the fungus are unique features and warranted establishment of the new taxon *Parathozetella*.

Type species: Parathozetella aquatica Sreekala et Bhat gen et sp. nov.

Paratozetella aquatica gen. et sp. nov.

Colonies white to yellowish, circular, regular, attaining a diam. of 2.7 cm in 7 days; reverse of the colony offwhite. *Mycelium* immersed, composed of thin-walled, hyaline, septate, hyphae 2-4 μm wide. *Conidiomata* sporodochial, scattered, solitary, discrete, superficial, slightly raised from the substratum, surmounted by greenish mass of conidia, encircled by long, hyaline to light yellowish, thick-walled, 320-600 μm long, up to 32-septate setae. *Conidiophores* compact, hyaline, 2-3-septate, unbrachhed, thin-walled 15-30 μm long, 2-3 μm wide. Sterile erect, thin-walled, hyaline, 4-15-septate, up to 120 μm long, 12 μm wide hyphae resembling paraphyses develop between the conidiophores. *Conidiogenous cells* monophialidic, simple, thin-walled, cylindrical, smooth, 8-12 μm

long, 2-3 μ m wide. *Conidia* bean- shaped, hyaline, slightly curved, ellipsoid, guttulate, 4-7 μ m long, 2-4 μ m wide, with 2 thin, filiform, 8-15 μ m long appendages developing from the curved edge.

<u>Holotype</u>: Dried culture mat derived from the single spore isolate of the fungus, perennial river, Siva Temple, Tambisurla, Goa, India, Herb No. GUFCC-167, Sreekala K. Nair, 8-7-2000.

64. *Periconia byssoides* Pers. ex Merat, 1821. (M. B. Ellis, 1971) (Fig. 89)

Colonies effuse, hairy, dark brown to black on the natural substratum. *Mycelium* immersed. *Conidiophores* macronematous, mononematous, 60-200 μm long, 2.5- 3 μm wide. Apical region of the conidiophore is subhyaline, 6-8 μm., smooth, dark brown, septate and thick-walled. *Conidiogenous cells* polyblastic, integrated, terminal and spherical. *Conidia* spherical, dark brown, verrucose, 3-6 μm in diam.

Specimen examined: On submerged twig, seasonal stream, Bondla Wildlife Sanctuary, Goa, India, Herb. No. GUFCC-50, Sreekala. K Nair. 9-8-1999.

65. Phalangispora constricta Nawawi and Webster, 1982. Trans. Br. mycol. Soc. 79: 65-68.(Fig. 48)

Conidial fungus, Hyphomycete. *Colonies* dark greenish-grey, with a paler margin and pale grey aerial mycelium; mycelium composed of hyphae 3-5 µm wide and with frequent anastamoses. *Conidiomata* sporodochial, developing after 4 days at 25°C, scattered or concrescent, appearing at first as whitish pustules of about 0.5mm. diam, funnel-shaped, 300-500 µm high. Each sporodochium consists of parallel strands of cylindrical, hyaline, anastomosing hyphae 3 µm diam, with darker, wider, inflated cells at

the base. *Setae* arising from basal group of darker cells, 4-5, long, dark, septate, subulate, up to 800 μm long, 11-12 μm wide at the base. *Conidiogenous cells* polyblastic, cylindrical, denticulate, 10-12 x 4-5 μm. *Conidia* cylindrical, aseptate, gutulate, 10-12 x 2-2.5 μm. *Conidial chains* branched, sometimes Y-shaped, pale brown, 85-125 μm long, composed of cylindrical, gutulate, aseptate conidia, with branches developing from basal 2-3 cells; basal and end conidial cells tapered.

Specimen examined: (i) On submerged and decaying leaves of *Xylia xylocarpa* Taub, seasonal stream, Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUBH-175, Sreekala K. Nair, 23-12-1999. (ii) Dried foam slide, perennial stream, Tambdisurla, Goa, India, Herb. No. GUBH-176, Sreekala K. Nair, 8-7-2000.

This is a terrestrial fungus reported several times earlier from aquatic system, especially in foam samples (Sridhar et al. 1992).

66. Pithomyces chartarum (Berk. and Curt.) (Ref. M. B. Ellis, 1971) (Fig. 88)

Colonies in MEA, effuse, black, up to 1.3 cm diam. in 10 days. Mycelium sparcely superficial. Conidiophores micronematous, branched and anastomosing, pale olive brown, verruculose, 2-5 μ m thick, denticles 2-10 x 2-3.5 μ m. Conidia broadly ellipsoidal, with 3-4 transverse septa, the middle cells usually divided by longitudinal septa, often constricted at the septa, mid to dark brown when mature, echinulate, verruculose, 18-29 x 10-17 μ m.

Specimen examined: (i) Dried foam slide, Bondla Wildlife Sanctuary, Goa, India, Herb. No. GUFCC-52, Sreekala. K. Nair, 12-8-2000. (ii) Dried culture mat derived from the single spore isolate of the fungus, Bondla Wildlife Sanctuary, Goa, India, Herb. No. GUFCC-52-a, Sreekala. K. Nair, 12-8-2000.

67. Pseudobotrytis terrestris (Timonin) Subram., 1956. (Ref. M. B. Ellis, 1971) (Fig. 90)

Colonies effuse, greyish brown at the centre. Mycelium immersed. Conidiophores micronematous, mononematous, stipe unbranched, straight or flexuous, upto 500 μm long, 4-5 μm thick just above the basal swolling and at the apex, pale to dark brown, smooth. Conidiogenous cells polyblastic, discrete, umbellately arranged over the swollen apex of the stipe, determinate, clavate, denticulate, denticles cylindrical. The umbels are of 6-12 in number, 14-23 μm x 2-3 μm. Conidia are solitary, dry, 7-9 x 3-3.5 μm., formed on denticles which cover the swollen tip of each conidiogenous cell, simple, oblong, rounded at ends or ellipsoidal, pale brown, smooth.

Specimen examined: On submerged twig of *Dellinia indica* Linn., seasonal stream, Bondla Wild Life Sanctuary, Goa, India. Herb. No. GUFCC-53. Sreekala. K. Nair. 21-10-2000.

68. Scolecobasidium sp.

(Fig. 93)

Colonies effuse, usually slow growing, slightly raised at the center, grey to brown, reverse brown. *Mycelium* partly superficial, composed of smooth hyphae which are branched and septate. *Conidiophores* macronematous, mononematous, often short, unbranched, sraight of flexuous, smooth, pale olivaceous, 4-25 x 1.5-3 μm. *Conidiogenous cells* polyblastic, integrated, terminal, sympodial, cylindrical and denticulate. *Conidia* solitary, dry, acropleurogenous, simple, ellipsoidal, oblong with rounded ends, 1-3 septate, sometimes constricted at the septa, pale olivaceous, verruculose or finely echinulate, 7-8 x 2.5-4.5 μm.

<u>Specimen examined</u>: On submerged twig of *Bambusa arundinaceae*, seasonal stream, Bondla Wild Life Sanctuary, Goa, India. Herb. No. GUFCC-55, Sreekala. K.

Nair, 21-10-2000

69. Scutisporus brunneus Ando and Tubaki, 1985. Trans. mycol. Soc. Japan. 26: 151-160. (Fig.11)

Conidial fungus, Hyphomycete. *Colonies* on MEA efffuse, thin, colourless to pale grey. *Mycelium* composed of septate, branched, smooth, hyphae 1-2 μm wide and colourless at first but darkening to pale brown with age. *Conidiophores* arising laterally, hyaline, 25-70 μm long, 1-2 μm wide. *Conidia* aleurio-type, sometimes sympodial, consisting of a short basal cell and 4-celled main body bearing a slender, appendage from each cell; basal cell cuniform, hyaline or pale brown, 2-3 μm long, 2-3 μm wide on the sides, 0.5-1.5 μm wide at the base. The four celled main body butterfly-shaped, pale to dark brown, cross-septate, 9-15 x 7.5-11 μm; vertical septum 7-9 μm long and horizontal septum 5.5-7.5 μm long. Appendages ciliate, hyaline, non-septate, 7-25 μm long, those from upper cells of same length and longer than those of lower cells.

Specimen examined: (i) Dried foam slide, seasonal stream, Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUBH-179, Sreekala K. Nair, 24-8-2000. (ii) Dried foam slide, seasonal stream, Molem Wildlife Sanctuary, Goa, India, Herb. No. GUBH-180, Sreekala K. Nair, 20-9-2000.

This interesting fungus was known earlier only from foam samples (Subram and Bhat, 1981).

70. **Sopagraha sibika** Subram. and Sudha 1978. Can. J. Bot. **57**: 34-39. (Fig. 51)

Conidial fungus, Hyphomycete. *Colonies* effuse, superficial. *Mycelium* superficial, composed of loose network of hyphae which are olivaceous brown to dark brown,

smooth, thick-alled, distantly septate, branched, 3.3-4.2 μm wide. *Conidiophores* erect, flexuous, simple or branched, smooth brown when young becoming darker at maturity, thick-walled, 2-9 septate, arising terminally and laterally from repent hyphae, 26-105 μm long, 3.5-6 μm wide. *Conidia* developing singly and terminally on the conidiophore or on its branches, gangliar, brown to dark brown at maturity, appearing as shiny black bodies on the leaf, variable in size, shape, and configuration, subglobose, oval to ellipsoidal, deeply constricted at the septa, with variable number of cells. Mature conidium typically consists of a basal cell, 26-42 μm x 19-32 μm, with two very darkly pigmented central cells, 4-12 primary cells arising from the central cell and 1-4 secondary cells arising from the primary cell.

<u>Specimen examined</u>: On the submerged leaves of *Hopea ponga* (Dennst.) Mabb., seasonal stream, Molem Wildlife Sanctuary, Goa, India, Herb. No. GUFCC-54, Sreekala. K. Nair, 25-12-2000.

This terrestrial litter fungus is reported here for the first time from an aqautic habitat.

71. *Spegazinia sundara* Subram., 1956, *J. Indian bot. Soc.* **35**: 76-78. (Fig. 67)

Conidia dark brown in colour, flower like in appearance, consisting of 4 petal like region, each measuring 7-10 x 5-8 μ m. These are heart-shaped and are arranged on a samll stalk which are slightly curved. Stalks are pale brown in colour, 4-8 μ m long and 3-5 μ m wide at the region where it is attached to the conidia.

Specimen examined: Rarely observed in the foam sample, seasonal streams, Molem Wild Life Sanctuary, Goa, India. Herb. No. GUBH-56, Sreekala K. Nair, 20-9-2000.

72. Speiropsis hyalospora Subram. and Lodha, 1964. Can. J. Bot. 42: 1057-1063.

(Fig. 52)

Conidial fungus, Hyphomycete. Colonies on MEA effuse, dark brown, attaining a diam. of 3.5 cm in 10 days. Mycelium scanty, composed of hyaline to subhyaline, septate, branched hyphae 2.5-3.5 µm wide. Conidiophores mononematous, erect, septate, slightly thick-walled, 80-120 x 4.5-5.0 µm, producing, by branching once or twice, an apical cluster of sporogenous cells. Conidiogenous cells vary in shape, wider towards the tip, 9.6-11.2 µm long, 4-4.8 µm wide below, 4.8-5.6 µm wide above, polyblastic, denticulate at the tip. The lower cells are darker than the distal ones. The spores are borne on the upper surface of the sporogenous cells, frequently several on a single sporogenous cell. Conidia in branched chains, usually with three arms, hyaline. Each conidial chain consists of a main axis with 6-8 cells, each separated by a narrow isthmus and usually two lateral arms similarly composed of several cells, but fewer in number, and seperated by isthmi. One of the arms arises laterally from near the distal end of the basal cell of the spore, the second arm similary from the second cell from the base, the first arm being longer than the second. Each arm is separated from the cell from which it arises by an isthmus. The first arm 5-6-celled, $40-60 \times 4.8-5.2 \mu m$, the second arm 3-4 celled, $27-38 \times 4-4.8 \mu m$.

Specimen examined: (i) Dried foam slide, seasonal stream, Bondla Wildlife Sanctuary, Goa, India, Herb. No. GUFCC-99, Sreekala. K. Nair, 25-10-1999. (ii) Dried culture mat, single spore isolation, seasonal, stream, Bondla Wildlife Sanctuary, Goa, India, Herb. No. GUFCC-99-a, Sreekala. K. Nair, 25-10-1999.

The fungus was originally described from terrestrial litter and is now recorded for the first time from aquatic habitat.

73. *Speiropsis pedatospora* Tubaki 1958. J. *Hat. Bot. Lab.* **20**: 142-244. (Fig. 53)

Conidial fungus, Hyphomycete. *Colonies* effuse, dark brown, 3.2 cm diam. in 7 days. *Mycelium* partly superficial, composed of septate, branched, dark brown hyphae 2.5-3.5 µm wide. *Conidiophores* dark brown at the base, light brown towards the apex, 60-130 µm long, 3.5-5 µm wide, 3-8-septate, arising laterally from vegetative hyphae, erect, slightly thick walled, by branching once or twice at the apex forming an apical cluster of polyblastic conidiogenous cells. *Conidia* in branched chains, light brown; each arm 45-85 µm long, with 2-8 conidia in each chain, 4.5-6 µm wide. Lateral bhanches arise from main axis, with 2-5 conidia.

Specimen examined: (i) On submerged leaf litter of *Dellenia indica* Linn., seasonal stream, Bondla Wildlife Sanctuary, Goa, India, Herb. No. GUBH-95, Sreekala K. Nair, 9-8-1999. (ii) Dried foam slide, seasonal stream, Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUFCC-96, Sreekala K. Nair, 19-8-1999.

74. Sporoshisma uniseptata Bhat and Kendrick, 1993. Mycotaxon 59: 71-73

(Fig. 91)

Colonies effuse, black. Mycelium immersed. Conidiophores micronematous, macronematous, scattered, unbranched, straight or flexuous, dark brown, smooth, each composed of a swollen base, a cylindrical stipe, and a large, lageniform spore sac with long cylindrical neck. Conidiogenous cells monophialidic, integrated, terminal, determinate. Conidia catenate, formed endogenously in a few transverse septa, pale to dark brown, 1-septate, smooth, 15-20 x 5-8 μm.

Specimen examined: On submerged twig, seasonal stream, Cotigao Wild Life Sanctuary, Goa, India, Herb. No. GUFCC-58, 24-8-2000.

Conidial fungus Hyphomycete. *Colonies* effuse, regular, circular, greyish, attaining a diam. of 4.3 cm in 7 days; reverse of the colony dark grey to black. *Mycelium* consists of superficial, cottony, septate, branched hyphae 2-4 µm wide. *Conidiophores* erect, solitary, simple or branched, septate, brown throughout except the fertile region, straight or slightly curved, length increasing by sympodial growth, 120-320 µm long, 4-5 µm wide at the base, 3-4 µm at the apex; upper fertile region geniculate, up to 80 µm long. *Conidiogenous cells* polyblastic, sympodial, with truncate tip on secession of conidia. *Conidia* borne apically, subuliform, hyaline, with truncate base, with acute and slightly curved tip, 2-5 septate, 30-45 µm long, 2-2.5 wide.

Specimen examined: (i) On submerged leaf litter of *Bambusa aundinaceae* (Retz.) Willd, seasonal stream, Bondla Wildlife Sancturary, Goa, India, Herb. No. GUBH-182, Sreekla K. Nair, 12-8-2000. (ii) Dried culture mat derived from the single spore isolate of the fungus, Bondla Wildlife Sancturary, Goa, India, Herb. No. GUBH-182-a, Sreekala K. Nair, 12-8-2000.

The fungus is well known from both terrestrial and aquatic habitats (Matsushima, 1971, 1975).

76. Subulispora sp.2

(Fig. 55)

Colonies effuse, grey to pale brown, with sparcely superficial mycelium emerging out from the substratum. *Mycelium* consists of septate, branched hyphae 2-4 µm wide. *Conidiophores* erect, flexuous, solitary, simple, septate, pale brown, with a hyaline tip, 50-180 µm long, 3.5-4.5 µm wide at the base, 2-4 µm at the tip. *Conidia* hyaline, elongated,

3-8 septate, Y-shaped, 20-35 μm long, 2-2.5 wide, with a truncate base and tapering apical region, with appendage measuring 12-20 μm long; rarely conidia produce two appendages.

Specimen examined: On submerged leaf litter of *Dellenia indica* Linn., seasonal stream, Molem Wildlife Sancturary, Goa, India, Herb. No. GUBH-83, Sreekala. K. Nair, 20-9-2000.

Conidia of this fungus, variable from previous speicies, were gathered once from stream habitat

77. Tetrachaetum elegans Ingold, 1942. Trans. Br. mycol. Soc. 25: 339-417. (Fig. 50)

Conidial fungus, Hyphomycetes. *Colonies* effuse, with sparce mycelium. *Mycelium* slow growing, composed of septate, branched hyphae 1.5 µm wide. *Conidiophores* very long, thin-walled, hyaline, septate, 20-50 µm long. *Conidogenous cells* thallic. *Conidia* get liberated from conidiophores under submerged conidion from nartural substratum, terminal, tetraradiate, consists of a main axis bent in the middle, with two lateral branches in the ragion of the bent making up the other two arms, with all arms of similar thickness, 100-150 µm long, 2-4 µm wide.

Specimen examined: On submerged leaf litter of *Grewia hirsuta* Vahl., seasonal stream, Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUBH-86, Sreekala K. Nair, 23-12-1999.

78. *Tetraploa aristata* Berk. and Br., 1850. *Ann. Mag. nat. Hist.*, **5**: 459. (Fig. 56)

Conidial fungus, Hyphomycete. *Colonies* effuse, brown to dark greyish brown. *Mycelium* superficial. *Conidiophores* micronematous, branched, anastomosing to form a

network, flexuous, hyaline to pale yellowish brown, often verruculose, 1.5-3 μ m wide. *Conidiogenous cells* monoblastic, occassionally polyblastic, integrated, intercalary, determinate, cylindrical. *Conidia* solitary, dry, pleurogenous, appendaged, brown, verruculose, muriform; mature conidia mostly with 4 cells in each column, 25-40 x 14-30 μ m, with septate appendages 12-80 x 3.5-8 μ m.

Specimen examined: Dried foam slide, seaosnal stream, Cotigao Wildlife Sanctuary, Goa, India. Herb. No. GUFCC-98, Sreekala K. Nair, 19-8-1999.

T. aristata is a common terestrial fungus often encountered in foam. It has been reported from almost all countries (Ingold, 1975; Dix and Webster, 1995). Earlier reports make several references of this fungus in foam from Indian subcontinent (Sridhar et al. 1992).

79. Tricladium angulatum Ingold, 1942. Trans. Br. mycol. Soc. 25: 339-417. (Fig. 58)

Conidial fungus, Hyphomycetes. *Colonies* on MEA white with concentric rings of denser mycelium, glabrous, with diffuse margin, attaining a diam. of 1.8 cm in 15 days. *Conidiophores* arising from vegetative hyphae, mononematous, simple. *Conidiogenous cells* monoblastic. *Conidia* hyaline, 55-75 μ m in span, consisting of 3-4-septate main axis and 2-3, 0-1-septate laterals. Main axis 45-60 μ m long, 3.5-5 μ m at the widest, 1.5-2 μ m at the tip; laterals 20-50 μ m long, 2.5-4 μ m wide at the base and tapering to 1.5 μ m at the apex.

Specimen examined: Dried foam slide, seasonal stream, Cotigao Wildlife Sanctuary, Goa, India. Herb. No. GUFCC-97, Sreekala K. Nair, 19-8-1999.

The fungus sporulates sparcely on agar medium but produces abundant conidia when placed in aerated water.

80. Tricladium splendens Ingold, 1942, Trans. Br. mycol. Soc. 25: 339-417. (Fig. 57)

Conidial fungus, Hyphomycetes. *Colonies* slow growing, smooth, whitish, with sparce areal mycelium. *Conidiophores* simple, up to 55 μ m, 3 μ m wide. *Conidiogenous cells* terminal, integrated, swollen near the apex after release of the conidium. *Conidia* apical, solitary, axis 50-80 μ m, broadest in the middle, bent or sigmoid, with sharp apex and truncate base, with 2 arms, constricted bilaterally at the point of insertion, acicular at the distal end, almost of the same length and width, 30-40 μ m x 2-4 μ m.

Specimen examined: (i) Dried foam slide, seasonal stream, Molem Wildlife Sanctuary, Goa, India, Herb. No. GUBH-91, Sreekala K. Nair, 29-12-1999. (ii) Dried foam slide, seasonal stream, Bondla Wildlife Sanctuary, Goa, India, Herb. No. GUBH-93, Sreekala K. Nair, 9-8-1999.

The fungus sporulates well under water.

81. Trinacrium indica Soosamma et al. 2001. Mycologia 93: 1200-1202. (Fig. 59)

Colonies on malt extract agar slow growing, with circular margin, white to grey, flat, 1.2 - 1.5 cm diam in 10- -12 d at 25 C. *Mycelium* mostly immersed, composed of branched, septate, hyaline hyphae 1-3 μm wide. *Conidiophores* micronematous, mononematous, formed laterally on vegetative hyphae, simple, hyaline, smooth, 1.5-2.5 x 1.5-2.5 μm. *Conidiogenous cells* monoblastic, simple, determinate to sympodial, hyaline, truncate at the apex after conidial secession. *Conidia* solitary, triradiate, asymetrically Y-shaped, septate, hyaline, smooth, thin-walled, slightly constricted and septate at bifurcation point, pale brown in mass, developing in groups of 1-10 from one loci, with main axis cylindrical, erect or flexuous, 1-3-septate, 28-56 x 1.2-2.5 μm, with two

cylindrical, synchronously developing and divergent arms at the apex of the main axis; short arm straight or slightly flexuous, rounded at the tip, 1-2-septate, 23-35 x 1.2-2.5 μ m; long-arm 1-3-septate, distinctly recurved at midway between, 46-65 x 1.2-2.5 μ m.

Specimen examined: On decaying and submerged leaves of *Coffea arabica*, 15 Oct. 1998, Seasonal stream, Ponda, Goa, Herb. No. GUBH-169, Sreekala K. Nair, 18-11-1998.

82. Trinacrium subtile Ando, 1992. Trans. Mycol. Soc. Japan 33: 223-229. (Fig. 96)

Conidial fungus Hyphomycete. *Colonies* effuse, pale brown to mid brown. *Mycelium* composed of smooth, septate, branched, hyaline hyphae, 1-2 μ m wide. *Conidiophores* mononematous, formed on vegetative hyphae, simple, hyaline, smooth, 2-4 x 1.5-2.5 μ m. *Conidiogenous cells* monoblastic, simple, terminal, integrated, hyaline, truncate at the apex after conidial secession. *Conidia* solitary, triradiate, septate, hyaline, smooth, thin-walled, Y-shaped, slightly constricted at the point of attachment, with a cylindrical, erect or flexuous, main axis, 1-5-septate, 50-60 x 1-2.5 μ m, with two cylindrical, divergent arms arising from the main axis; both arms septate, hyaline, cylindrical, 25-40 x 1-2.5 μ m.

Speciemen examined: On the submerged leaf litter of *Dalbergia lanceolaria* Linn., seasonal stream, Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUBH-91, Sreekala. K. Nair, 11-4-1999.

The fungus was originally described by Ando (1992) from rain drops draining from leaves of *Quercus acutissima* Carr., in Japan. This is the first record of this fungus from India.

83. *Tripospermum myrti* (Lind) Hughes, 1951. *Mycol. Pap.*, **46**: 17-18. (Fig. 28)

Colonies effuse, brown, blackish brown or black. Mycelium superficial, composed of much branched, anastomosing hyphae 4-8 μm wide. Conidiophores semi-macronematous, mononematous, erect, unbranched, occasionally branched, pale to mid brown, smooth, up to 90 μm long, 4-8 μm thick. Conidiogenous cells monoblastic or rarely polyblastic, integrated, terminal, determinate, cylindrical or dolliform. Conidia solitary, dry, branched, usually made of a pyriform and ellipsoidal stalk cell and 4 divergent, subulate mutiseptate arms, pale to midbrown and smooth. The stalk cell of the conidia measures 6-10 x 4-7 μm, arms upto 30 μm long, 4-8 μm at the base, tapering to 1-2 μm, 1-4 septate, often constricted at the septa. One of the arms in this species usually lies parallel to the stalk cell.

Specimen examined: Submerged leaf litter of *Lagerstroemia lanceolata* Wall. Ex Wt. and Arn., seasonal stream, Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUFCC-92, Sreekala K. Nair, 23-12-1999.

Known generally from the terrestrial habitat, the fungus has earlier been reported from the aquatic habitat in the Western Ghats (Subram. and Bhat, 1981). This is a new record from Goa.

84. Triscelophorus acuminatus Nawawi, 1975. Trans. Br. Mycol. Soc. 64: 345-348. (Fig. 60)

Conidial fungus, Hyphomycete. *Colonies* on MEA, moderately growing, light grey, attaining a diam. of 2.3 cm in 10 days; reverse of the colony grey to brown. *Mycelium* sparcely superficial, composed of smooth, septate, branched, colourless, thinwalled hyphae 1-3 µm wide. *Conidiophores* mononematous, simple, erect, flexuous,

hyaline, smooth, up to 40 µm long. *Conidiogenous cells* monoblastic, integrated, terminal. *Conidia* resembles to that of *T. monosporus* in size, shape and measurement; difference between the two is the presence of septa on the main axis and lateral branches; arms strongly taper distally.

Specimen examined: (i) Dried foam slide, seasonal stream, Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUBH-94, Sreekala K. Nair; 19-8-1999. (ii) Dried culture mat, seasonal stream, Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUFCC-94-a, Sreekala K. Nair; 19-8-1999.

85. Triscelophorus konajensis, Sridhar and Kaveriappa, 1987. Indian Phytopathol. 40: 102-105. (Fig. 61)

Conidial fungus, Hyphomycete. *Colonies* on MEA, slow growing, sparcely superficial, regular, creamish, attaining a diam. of 3 cm in 7 days. *Mycelium* composed of smooth, septate, branched, colourless, thin-walled hyphae 1-4 µm wide. *Conidiophores* mononematous, simple, erect or flexuous, hyaline, smooth, unbranched, 0-3-septate, 3-25 µm long, 1-4 µm wide. *Conidiogenous* cells monoblastic, integrated, terminal. *Conidia* solitary, hyaline, smooth, tetraradiate; main axis measuring 10-20 µm long, 3-5 µm wide at the base, tapering at the tip; lateral arms arise from the broad basal region of the main axis, 2-5 µm wide at the base with 1-3 µm wide apical region.

Specimen examined: (i) Dried foam slide, seasonal stream, Bondla Wildlife Sanctuary, Goa, India, Herb. No. GUBH-93, Sreekala K. Nair, 12-8-2000. (ii) Dried culture mat, seasonal stream, Bondla Wildlife Sanctuary, Goa, India, Herb. No. GUBH-93-a, Sreekala K. Nair; 12-8-2000.

86. Triscelophorus monosporus Ingold, 1943. Trans. Br. mycol. Soc. 26: 148-52.

(Fig. 62)

(Fig. 95)

Conidial fungus, Hyphomycete. *Colonies* on MEA effuse, cream to light grey, attaining a diam. of 4.5 cm in 7 days; reverse of the colony light grey. *Mycelium* sparcely superficial, composed of smooth, septate, branched colourless, thin-walled hyphae 1-1.5 µm wide. *Conidiophores* mononematous, unbranched, simple, erect or flexuous, hyaline, smooth, 0-5-septate, 15-45 µm long, 2-4 µm wide. *Conidiogenous cells* monoblastic, integrated, terminal. *Conidia* solitary, hyaline, smooth, tetraradiate; main axis of the conidia consisting of a triangular base and a tapering tip, separated by a narrow septa at its broader region, 18-45 µm long, 3-6 µm broad at the base and 1-4 µm at the tip; lateral branches 3, arising from below septum, 10-30 µm long, with tapering ends, 2-4 µm broad at the region of attatchment to the main axis.

Specimen examined: (i) Dried culture mat derived from single spore isolate of fungus, seasonal stream, Chorlem Ghat, Herb. No. GUFCC-90, Sreekala K. Nair, 2-7-1999. (ii) On submerged leaf litter of *Careya arborea* Roxb., seasonal stream, Molem Wildlife Sanctuary, Goa, India, Herb. No. GUBH-87, Sreekala K. Nair, 18-9-1999. (iii) Dried foam slide, seasonal stream, Bondla Wildlife Sanctuary, Goa, India, Herb. No. GUBH-88, Sreekala K. Nair, 25-10-1999.

87. Vermiculariopsiella sp.

Colonies effuse, usually slow growing, slightly raised at the center, grey to black, attaining a diam. of 4.8 cm in 10 days, reverse of the colony black. *Mycelium* partly superficial, composed of smooth, septate, brached, pale brown hyphae, 2-3 µm wide. *Sprodochia* seen scattered in the colony, which are 0.3 mm in diam. Setae, simple, long,

acutely pointed at the apex, septate, brown, smooth, upto 350 μm *long. Conidiophores* macronematous, mononematous, often short, unbranched, sraight or flexuous, smooth, hylaine, 4-25 x 1.5-3 μm. *Conidiogenous cells* polyblastic, integrated, terminal, and cylindrical. *Conidia* solitary, dry, acropleurogenous, simple, ellipsoidal, oblong with rounded ends, aseptate, hyaline to very slightly pigmented, smoothwalled, 7-12 μm long and 2.5-4.5 μm wide.

Speciemen examined: (i) Dried foam slide, seasonal stream, Molem Wild Life Sanctuary, Goa, India. Herb. No. GUBH-59, Srekala. K. Nair, 20-9-2000. (ii) On live root as root endophyte of *Hopea ponga* (Dennst.) Mabb, seasonal stream, Molem Wild Life Sanctuary, Goa, India, Herb. No. GUFCC-60, Sreekala. K. Nair, 20-9-2000. (iii) Dried culture mat, seasonal stream, Molem Wild Life Sanctuary, Goa, India. Herb. No. GUBH-60-a, Sreekala. K. Nair, 20-9-2000.

88. Verticillium sp. (Fig. 97)

Colonies effuse, white. Mycelium hyaline, septate and branched. Conidiophores hyaline, branched, 10-18 x 1.5-2.5 μ m. Conidia hyaline, circular, smooth-walled, with a diam. of 3-5 μ m.

Specimen examined: (i) Dried foam slide, seasonal stream, Alorna, Goa, India. Herb. No. GUBH-65, Sreekala. K. Nair, 20-7-2000. (ii) Dried culture mat, seasonal stream, Alorna, Goa, India. Herb. No. GUFCC-65-a, Sreekala. K. Nair, 20-7-2000.

89. *Virgaria* sp. (Fig. 94)

Colonies white to pale brown. Mycelium pale brown, composed of branched, septate, 1-2.5 µm wide hyphae. Conidiophores mononematous, simple, branched, 15-25 x

2-4 μ m. Conidiogenous cells deticulate, with several, small peg like denticles at the tip of the conidiogenous cells. Conidia ovoid to elliptical, with a prominant scar at the place of detachment, 5-10 x 3-6 μ m.

Specimen examined: (i) Dried foam slide, seasonal stream, Cotigao Wild Life Sanctuary, Goa, India. Herb. No. GUBH-66, Sreekala. K. Nair, 11-4-1999. (ii) Dried culture mat, seasonal stream, Cotigao Wild Life Sanctuary, Goa, India. Herb. No. GUFCC-66-a, Sreekala. K. Nair, 11-4-1999.

90. Weisneriomyces javanicus Koorders, 1907. Bot. Untersuch. 246-247. (Fig. 98)

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 dark green. *Mycelium* white to 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; mature sporodochia 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 borne, 8-12 x 3-4 μm. *Conidia* slimy, in chains, individually colourless, in mass pale brown, smooth, 0-septate, 10-12 x 3-4.5 μm; end cells in the chain tapered, intermediate cylindrical.

Specimen examined: On submerged leaves of *Xylia xylocarpa* Taub., seasonal stream, Cotigao Wild Life Sanctuary, Goa, India, Herb. No. GUBH-89, Sreekala. K. Nair, 24-8-2000.

This is a terrestrial leaf litter fungus often recorded from aquatic foam (Ingold, 1975).

2. UNDETERMINED TAXA

A number of aquatic spora with unique and unusual shapes were recognised from foam gathered from various streams or in culture derived from single spores. These taxa were not assignable to any known species. Therefore they are described here tentatively as 'Undetermined' species 1-10.

91. Undetermined sp.1

(Fig. 64)

Colonies effuse, hyaline to white, attaining a diam. of 3.4 cm in 12 days; reverse of the colony white. *Mycelium* immersed, composed of hyaline, thin-walled, septate, smooth, branched hyphae 1.5-2.5 μm wide. *Sporodochia* scattered, with compactly arranged hyaline conidiophores and conidiogenous cells. *Conidiophores* hyaline, branched, septate, 25-15 μm long, 1-2.5 μm wide. *Conidiogenous cells* monoblastic, compactly arranged in the sporodochia, hyaline, 5-10 μm long, 1-2 μm wide. *Conidia* helicoid, hyaline, solitary, terminal, long-fusoid, 150-290 x 6.5-9 μm, 0-septate, with truncate base, obtuse at the apex.

Specimen examined: (i) Submerged root of *Bambusa arundinaceae* (Retz.) Willd., seasonal stream, Bondla Wildlife Sanctuary, Goa, India, Herb. No. GUBH-184, Sreekala,

K. Nair; 9-8-1999. (ii) Dried culture mat, root endophyte, seasonal stream, Bondla Wildlife Sanctuary, Goa, India, Herb. No. GUBH-184-a, Sreekala, K. Nair, 9-8-1999.

92. Undetermined sp. 2

(Fig. 65)

Conidia tri-radiate, with a triangular central body 3-5 μm in diam. Each protuberence, of same size and shape, 4-8 μm long, 3-5 μm wide in the middle region.

Specimen examined: Dried foam slide, seasonal stream, Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUFCC-185, Sreekala, K. Nair, 24-8-2000.

93. Undetermined sp.3

(Fig. 66)

Conidia resembling to *Tricladium splendens*, in its shape, but differs in their size and aseptate, flexible, thin lateral branches. *Conidia* are hyaline, 10-20 μ m long main axis, consisting of 3-4 septate two, 0-septate laterals. Main axis is 1.5-2.5 μ m wide at the broader region, 0.5-1 μ m at the tip, 2 laterals, 4-15 μ m long, 0.5-1.5 μ m wide at the base and tapering to 0.3 μ m at the apex.

Specimen examined: Rarely observed in the foam sample collected from the seasonal streams of Mollem. Herb. No. GUFCC-186. Molem Wild Life Sanctuary, Goa, India, Sreekala, K. Nair; 20-9-2000.

94. Undetermined sp. 4

(Fig. 68)

Conidia hyaline, aseptate, with a main axis, 8-16 μ m long, 2-3 μ m wide at the basal truncate ragion. Tip of the main axis tapers towards apical region, and consisting of two hyaline aseptate lateral branches, 8-12 μ m long and 1-2.5 μ m wide at the region of attachment at the main axis.

Specimen examined: Dried foam slide, seasonal stream, Cotigao Wild Life

Sanctuary, Goa, India, Herb. No. GUBH-188, Sreekala, K. Nair, 24-8-2000.

95. Undetermined sp. 5

(Fig. 69)

Conidia 'Y' shaped, resembling *Trinacrium gracile* in shape, differing by its smaller size, hyaline, measuring 18-23 μ m in its span, 1-4 μ m wide at the center of the spore, with two branches, each measuring 8-12 μ m long from the centre, 1-septate, with rounded ends.

Specimen examined: On submerged leaf litter of unidentified plant, seasonal stream, Bondla Wild Life Sanctuary, Goa, India, Herb. No. GUFCC-189, Sreekala, K. Nair, 25-10-1999.

96. Undetermined sp. 6

(Fig. 70)

Conidia hyaline, solitary, terminal, branched in 2-4 levels, with a flexible, 6-12-septate, main axis, 30-80 μ m long, bends at the point of attachement of the lateral branches. The lateral branches 3-4 in number, 3-5 septate, up to 60 μ m long, 1-2 μ m wide, showing basal branching.

Specimen examined: Dried foam slide, seasonal stream, Cotigao Wild Life Sanctuary, Goa, India, Herb. No. GUFCC-200, Sreekala K. Nair; 17-12-2000.

97. Undetermined sp. 7

(Plate. 10. 1, 2)

Colonies black, hairy on the natural substratum. Mycelium partly immersed. Conidiophores macronematous, synnematous, up to 0.5 cm. high, with 20-30 µm diam. at the base, 12-45 µm at the apex, where the synnema ends like a bottle brush, with spores at their apical region, pale to dark brown, smooth, straight or flexuous, septate, 2-3 µm thick. Conidiogenous cells monophialidic, 12-20 µm long, integrated and terminal,

cylindrical, with a narrow neck region, 1.5-3 μ m wide. *Conidia* solitary, dry, acropleurogenous, simple, fusiform, spherical, hyaline to very pale brown, smooth, aseptate, 3-8 x 1.5-2.5 μ m.

Speciemen examined: On submerged twig, seasonal stream, Bondla Wild Life Sanctuary, Goa, India, Herb. No. GUBH-61, Sreekala K. Nair, 21-10-2000.

98. Undetermined sp. 8

(Plate. 12.2)

Mycelium sparcely superficial, composed of pale brown, septate, branched hyphae, 1-2 μm wide. Sporodochia is encircled with mid to dark brown setae, 100-120 μm long. Conidiophores compactly arranged, hyaline, branched, septate, 10-20 μm long. Conidiogenous cells phialidic. Conidia slightly curved, hyaline, 8-10 x 2-3 μm with small setulae at either side of the conidia.

Specimen examined: On submerged wood, seasonal stream, Anmod, Goa, India, Herb. No. GUBH-62, Sreekala K. Nair, 25-10-1999.

99. Undetermined sp. 9

(Plate. 11.5)

Mycelium hyaline, immersed in natural substratum. Conidiophores sporodochial in appearance, unbranched, occationally branched. Conidia holoblastic, integrated, solitary, clavate, rough walled, hyaline, aseptate, $15-24 \times 5-10 \mu m$.

Specimen examined: On submerged twig of *Terminalia tomentosa* Wt. and Arn., seasonal stream, Cotigao Wildlife Sanctuary, Goa, India, Herb. No. GUBH-51, Sreekala K. Nair, 24-8-2000.

100. Undetermined sp. 10

(Plate. 12.4)

Conidiophores synnematous in appearance. Conidia resembles the above species,

but differs by slightly longer cells.

Specimen examined: On submerged twig of *Xylia xylocarpa*, seasonal stream, Cotigao Wild Life Sanctuary, Goa, India. Herb. No. GUBH-57, Sreekala K. Nair, 24-8-2000.

3. COELOMYCETES

Six species of Coelomycetes were recovered in this study. These included Chaetomella sp., Robillarda phragmitis, Seimatosporium sp, Phoma sp., Pestalotiopsis sp.1 and Pestalotiopsis sp.2. Two of these, Robillarda phragmitis and Seimatosporium sp, are described and illustrated below.

101. Robillarda phragmitis Cunnel 1958. Trans. Br. mycol. Soc. 41(4): 405-412.

(Fig. 72)

Colonies effuse in MEA, dark grey, fast growing, attaining a diam.of 3.7 cm in 7 days; reverse of the colony black. Mycelium sparcely superficial, consisting of dark green, thick-walled, septate, branched hyphae 3-5 µm wide. Pycnidia superficial on agar yellow-brown, medium, 150-320 diam. with wall mostly elliptical, μm pseudoparenchymatous, 8-15 µm thick, composed of angular, isodiametric cells 4-8 µm wide; ostiole dark around the pore, 15-20 µm wide. Conidia closely packed and embedded in the mucilage within the pycnidial cavity and easily separated when mounted in water, fusiform, straight or slightly curved, smooth, colourless, constricted at the median septum, with one end rounded and other end obtuse, consisting of 2-4 (3), divergent, tapering, flexible, appendages 15-23 µm long.

Specimen examined: (i) As root endophyte on live roots of *Hopea ponga* (Dennst.) Mabb., seasonal stream, Molem Wildlife Sanctuary, Goa, India. Herb. No. GUBH-177, Sreekala K. Nair, 11-5-2000. (ii) Dried culture mat, seasonal stream, Molem Wildlife Sanctuary, Goa, India, Herb. No. GUFCC-177-a, Sreekala K. Nair, 11-5-2000. (iii) Dried foam slide, seasonal stream, Alorna, Goa, India, Herb. No. GUBH-178, Sreekala K. Nair, 20-7-2000.

This is one of the most common pycnidial fungi encountered in foam samples. It has been earlier recorded from the forests of Western Ghats (Subram. and Bhat, 1981). However, here it is a first record as root endophyte.

102. Seimatosporium sp.

(Fig. 71)

Colonies effuse, white to cream, attaining a diam. of 3.7 cm in 12 days; reverse of the colony off white. Mycelium immersed, composed of smooth, septate, branched, hyaline hyphae 1.5 µm wide. Pycnidia found scattered in the colony, with mass of conidia present in a slimy mucilage, at the osteole. Conidiophores mononematous, hyaline, arranged compact within the pycnidia, short, brnahced, 6-12 x 2-3 µm. Conidiogenous cells monoblastic. Conidia oval, hyaline, smooth-walled, 8-15 x 5-7 µm, with two long narrow appendages arising from either side of the conidia.

Specimen examined: (i) Dried foam slide, seasonal stream, Cuncolim, Goa, India, Herb. No. GUBH-63, Sreekala K. Nair, 4-10-2000. (ii) On submerged live root as endophyte, seasonal stream, Cotigao Wild Life Sanctuary, Goa, India, Herb. No. GUBH-64, Sreekala K. Nair, 11-4-1999. (iii) Dried culture mat, seasonal stream, Cotigao Wild Life Sanctuary, Goa, India, Herb. No. GUBH-64-a, Sreekala K. Nair, 11-4-1999.

4. ASCOMYCETES

Of the seven species of Ascomycetes recovered during the study, 3 were treated as undetermined morphotypes since their identity was not known. The recorded species are Chaetomium nigricolor, Emericella sp., Nectria haematococca, Xylaria sp. and Undetermined sp.1-3.

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Fig. 2. Alatospora acuminata	-conidia
Fig. 3. Anguillospora crassa	-conidiogenous cells and conidia
Fig. 4. Anguillospora furtiva	-conidia
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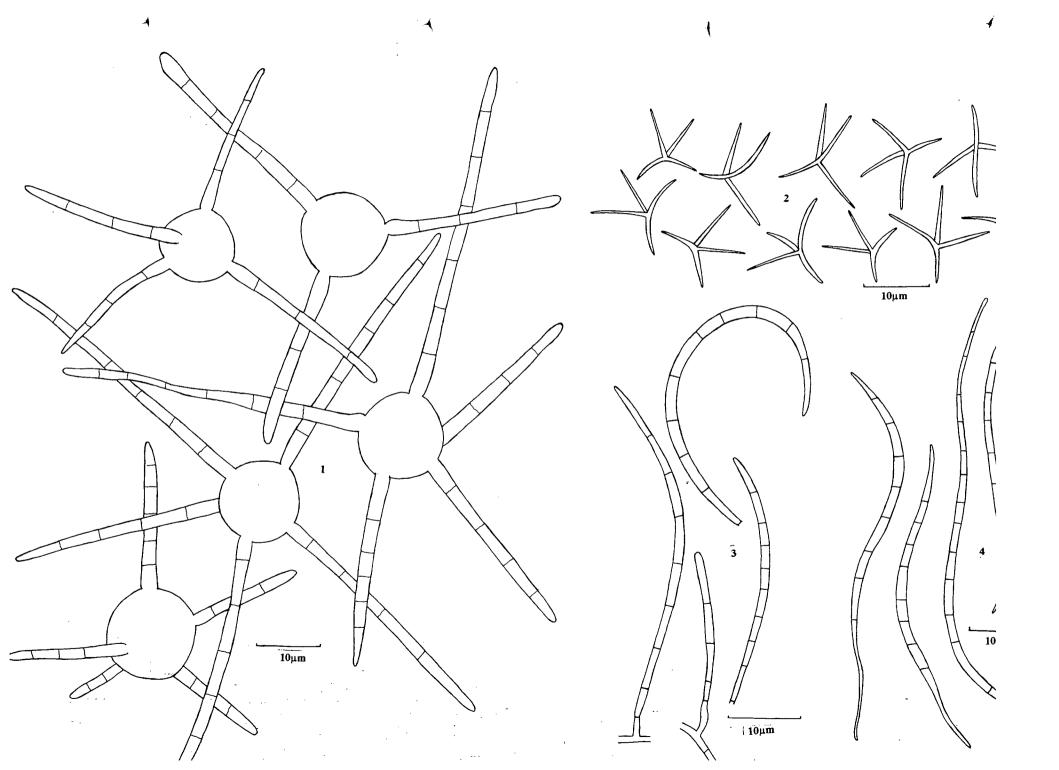
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Fig. 30. Dendrosporium lobatum	-conidiophore, conidiogenous cells and conidia	
Fig. 31. Dichotomophthoropsis aquatica -conidiophore, conidiogenous cells and conidia		
Fig. 32. Dictyochaeta assamica	-conidiophore, conidiogenous cells and conidia	
Fig. 33. Diploospora indica	-conidiophore, conidiogenous cells and conidia	
Fig. 34. Flabellospra crassa	-conidiophore, conidiogenous cells and conidia	
Fig. 35. Flagellospora curvula	-conidiophore, conidiogenous cells and conidia	
Fig. 36. Beltraniella odinae	-conidiophore, conidiogenous cells and conidia	
Fig. 37. Flabellospora verticillata	-conidia	
Fig. 38. Flabellospora multiradiata	-conidia	
Fig. 39. Helicomyces roseus	-conidiophore, conidiogenous cells and conidia	
Fig. 40. Helicosporium sp.1	-conidiophore, conidiogenous cells and conidia	
Fig. 41. Helicosporium sp.2	-conidiophore, conidiogenous cells and conidia	
Fig. 42. Helicosporium sp.3	-conidiophore, conidiogenous cells and conidia	
Fig. 43. Ingoldiella hamata	-conidiophore, conidiogenous cells and conidia	
Fig. 44. Isthmotricladia laeensis	-conidia	
Fig. 45. Isthmotricladia britanica	-conidiophore, conidiogenous cells and conidia	
Fig. 46. Kumbhamaya jalapriya	-conidiophore, conidiogenous cells and conidia	
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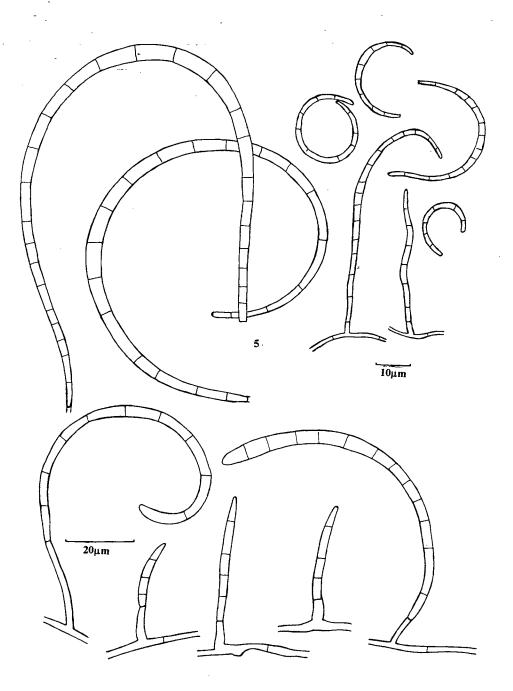
Fig. 47. Nawawia filiformis	-conidiophore, conidiogenous cells and conidia
Fig. 47-a. Nawawia sp.1	-conidia
Fig. 47-b. Nawawia sp.2	-conidia
Fig. 48. Phalangispora constricta	-conidia
Fig. 49. Mycoleptodiscus indicus	-conidia
Fig. 50. Tetrachaetum elegans	-conidiogenous cells and conidia
Fig. 51. Sopagraha sibika	-conidiophore, conidiogenous cells and conidia
Fig. 52. Speiropsis hyalospora	-conidiophore, conidiogenous cells and conidia
Fig. 53. Speiropsis pedatospora	-conidiophore, conidiogenous cells and conidia
Fig. 54. Subulispora sp. 1	-conidiophore, conidiogenous cells and conidia
Fig. 55. Subulispora sp.2	-conidiophore, conidiogenous cells and conidia
Fig. 56. Tetraploa aristata	-conidia
Fig. 57. Tricladium splendens	-conidiogenous cells and conidia
Fig. 58. Tricladium angulatum	-conidiophore, conidiogenous cells and conidia
Fig. 59. Trinacrium indica	-conidiophore, conidiogenous cells and conidia
Fig. 60. Triscelophorus acuminatus	-conidiophore, conidiogenous cells and conidia
Fig. 61. Triscelophorus konajensis	-conidiophore, conidiogenous cells and conidia
Fig. 62. Triscelophorus monosporus	-conidiophore, conidiogenous cells and conidia
Fig. 63. Parathozetella aquatica	-sporodochia, conidiophores and conidia
Fig. 64. Undetermined sp.1	-sporodochia, conidiophores and conidia
Fig. 65. Undetermined sp.2	-conidia
Fig. 66. Undetermined sp.3	-conidia

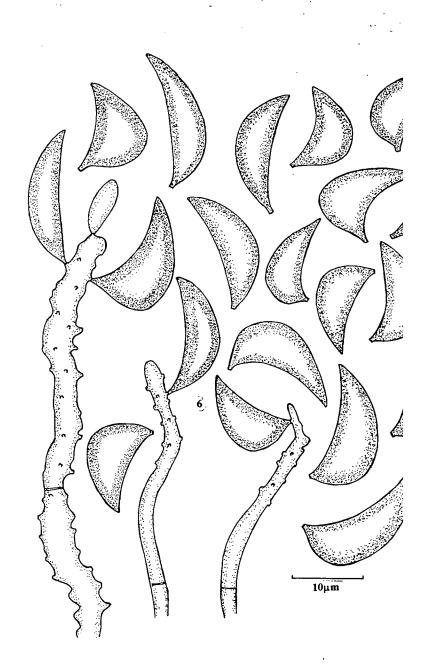
Fig. 67. Spegazinia sundara	-conidia	
Fig. 68. Undetermined sp.4	-conidia	
Fig. 69. Undetermined sp.5	-conidia	
Fig. 70. Undetermined sp.6	-conidia	
Fig. 71. Seimatosporium sp.	-pycnidia, conidiophores and conidia	
Fig. 72. Robillarda phragmitis	-pycnidia, conidiophores and conidia	
Fig. 73. Endophragmia inaquiseptata -conidiophores and conidia		
Fig. 74. Brachysporiella laxa	-conidiophores and conidia	
Fig. 75. Curvularia lunata	-conidiophores and conidia	
Fig. 76. Canalisporium caribense	-conidiophores and conidia	
Fig. 77. Colletotrichum dematiatum	-conidiophores and conidia	
Fig. 78. Candelabrum brochiatum	-conidia	
Fig. 79. Cylindrocarpon sp. 1	-conidiophores and conidia	
Fig. 80. Cylindrocladium sp. 1	-conidiophores and conidia	
Fig. 81. Cylindrocladium sp. 2	-conidiophores and conidia	
Fig. 82. Cylindrocarpon sp. 2	-conidiophores and conidia	
Fig. 83. Cylindrocarpon sp. 3	-conidiophores and conidia	
Fig. 84. Cylindrocladiopsis lagerstraemiae -conidiophores and conidia		
Fig. 85. Idriella lunata	-conidiophores and conidia	
Fig. 86. Hansfordia pulvinata	-conidiophores and conidia	
Fig. 87. Idriella angustispora	-conidiophores and conidia	
Fig. 88. Pithomyces chartarum	-conidiophores and conidia	
Fig. 89. Periconida byssoides	-conidiophores and conidia	

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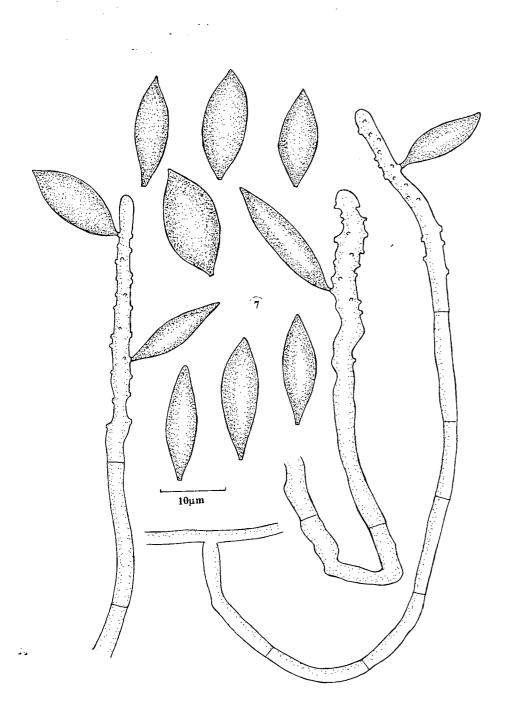
Fig. 90. Pseudobotritis terrestris -conidiophores and conidia Fig. 91. Sporoshisma uniseptata -conidiophores and conidia Fig. 92. Gonitrichum macrocladum -conidiophores and conidia Fig. 93. Scolecobasidium sp. -conidiophores and conidia Fig. 94. Virgaria sp. -conidiophores and conidia Fig. 95. Vermiculariopsiella sp. -conidiophores and conidia Fig. 96. Trinarium subtile -conidiophores and conidia Fig. 97. Verticillium sp. -conidiophores and conidia Fig. 98. Weisneriomyces javanicus -conidia

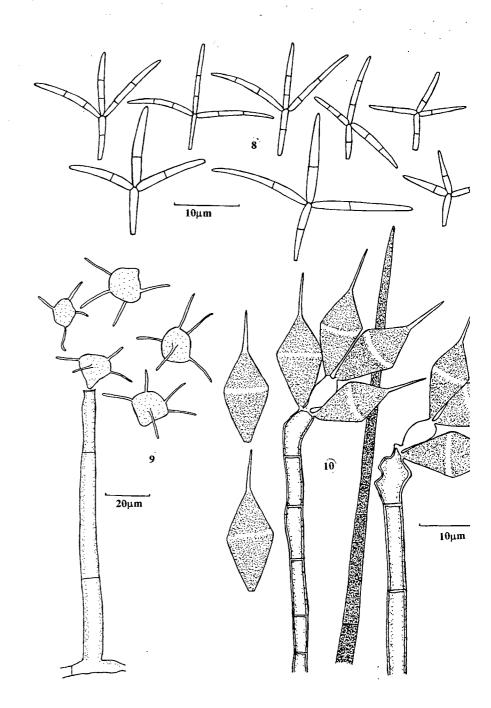


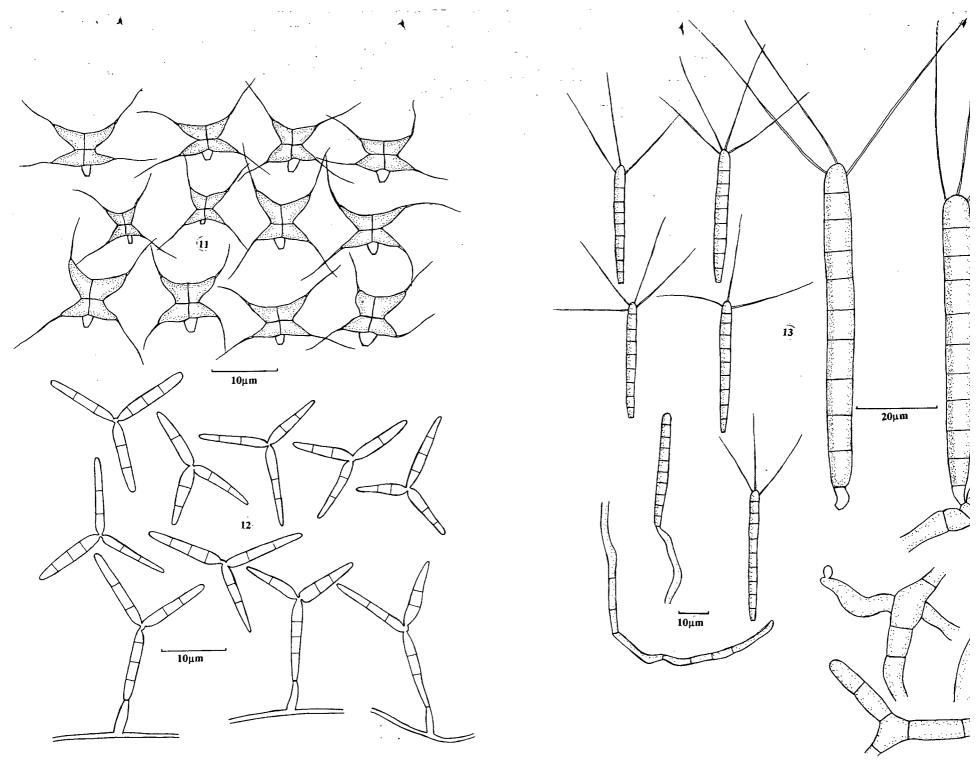




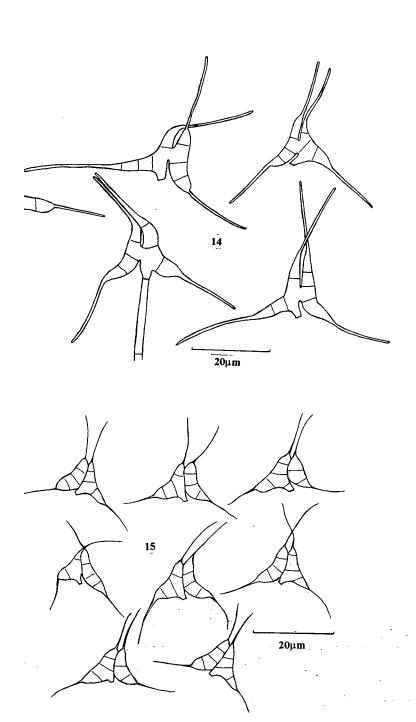
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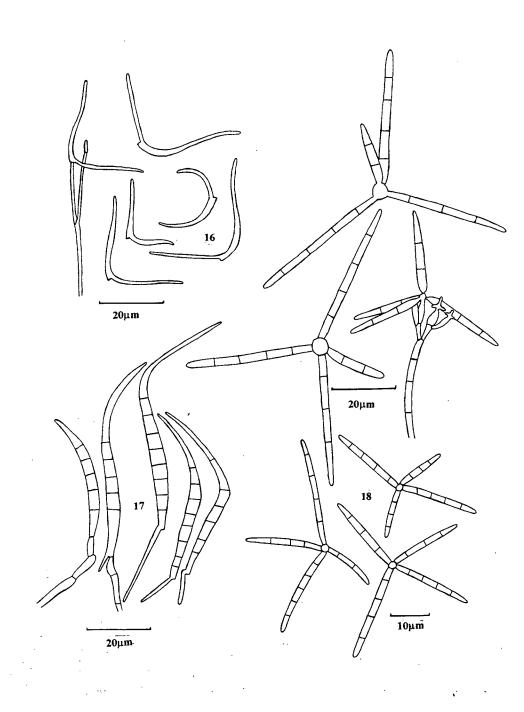


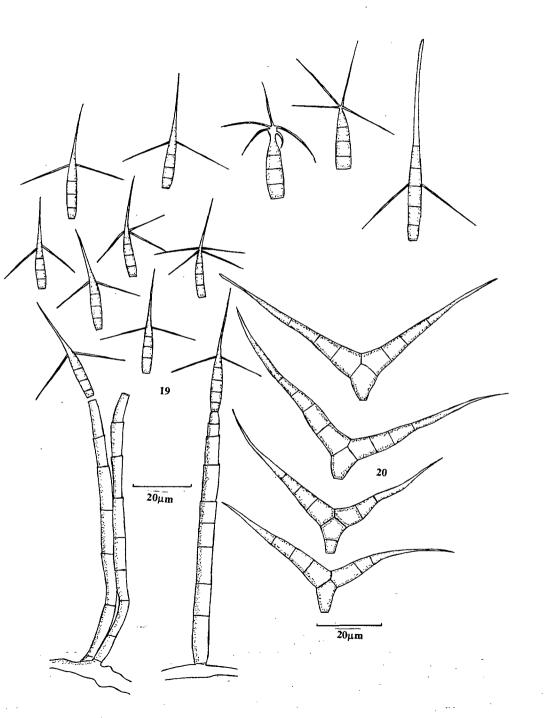


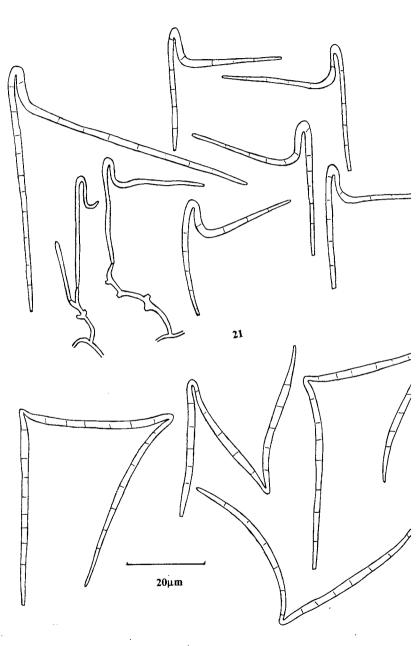


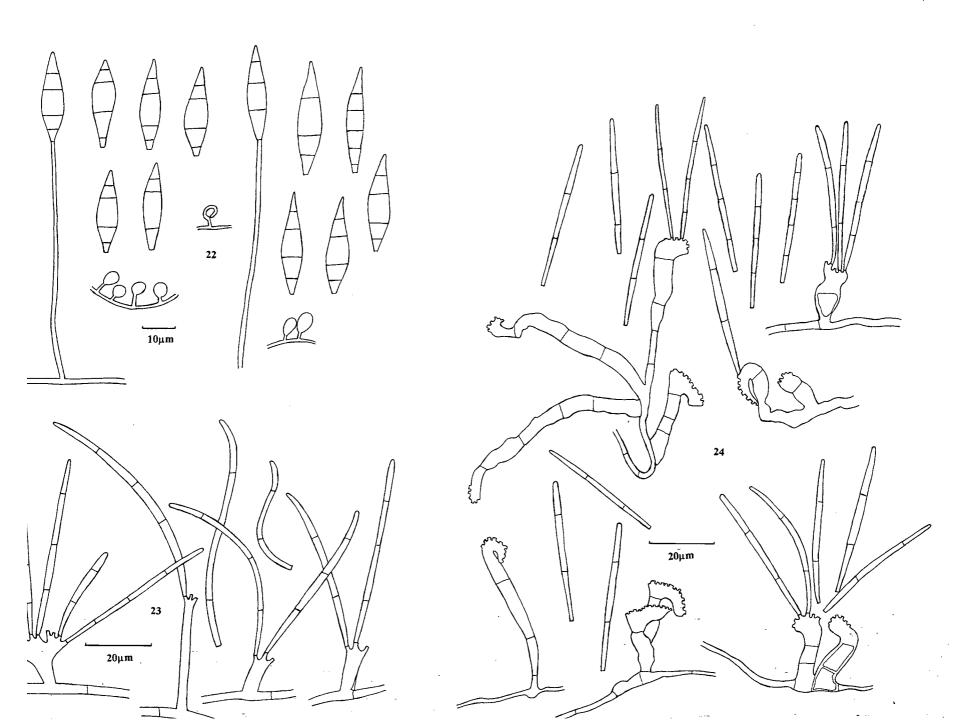
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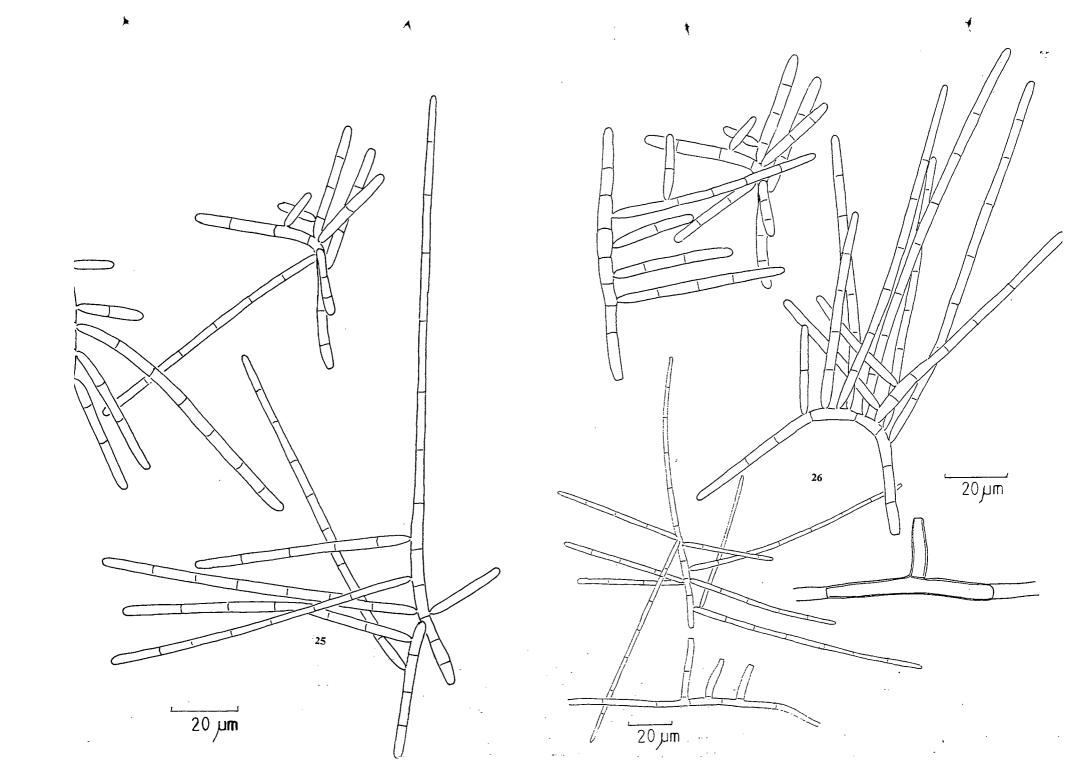


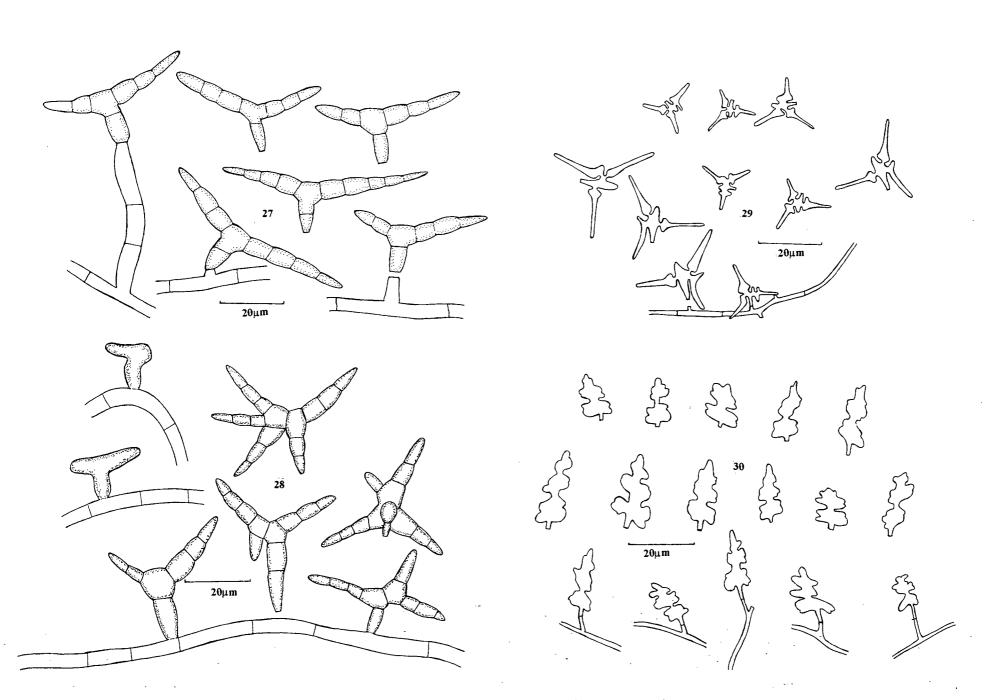




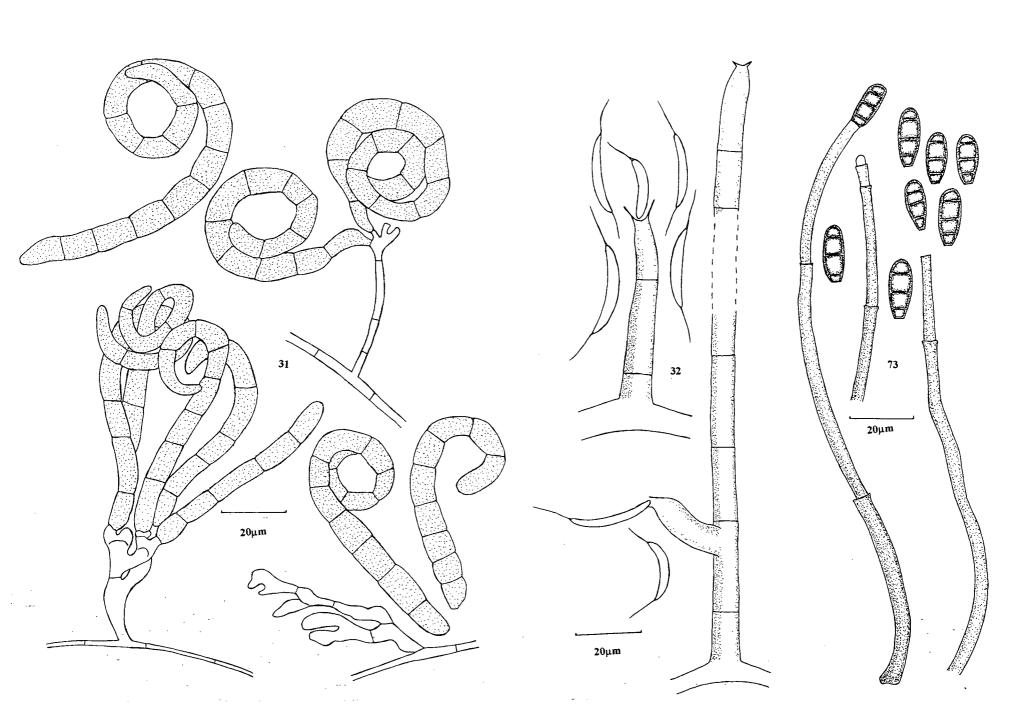


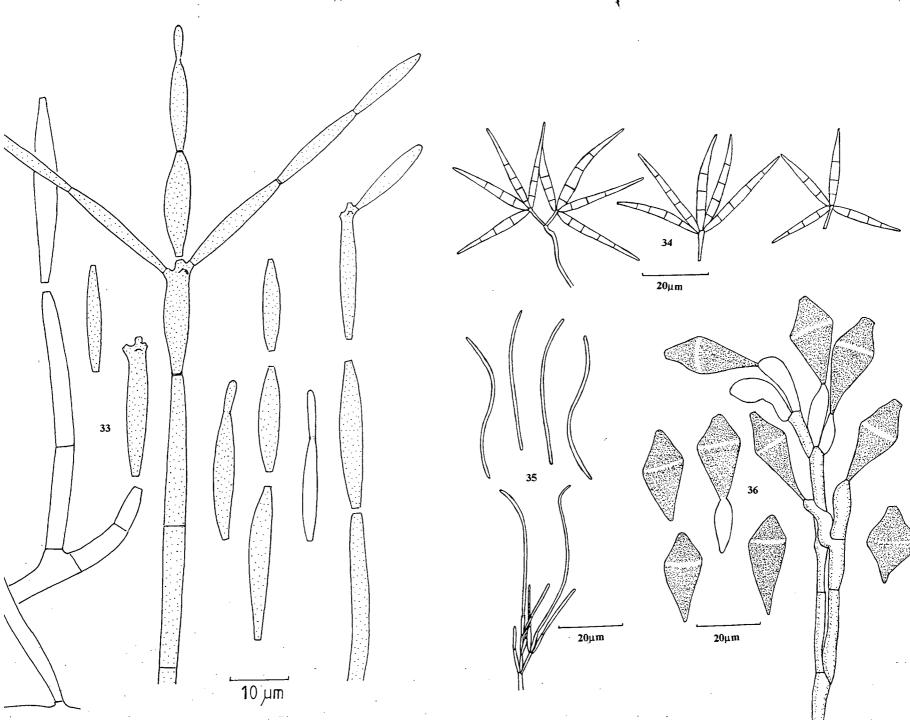




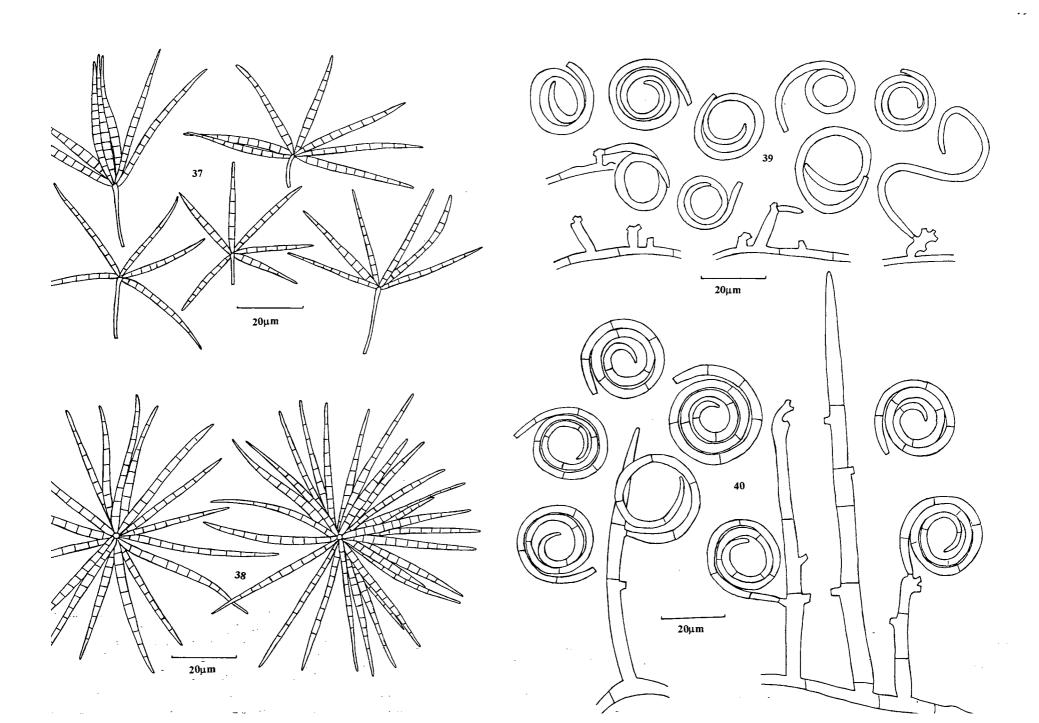


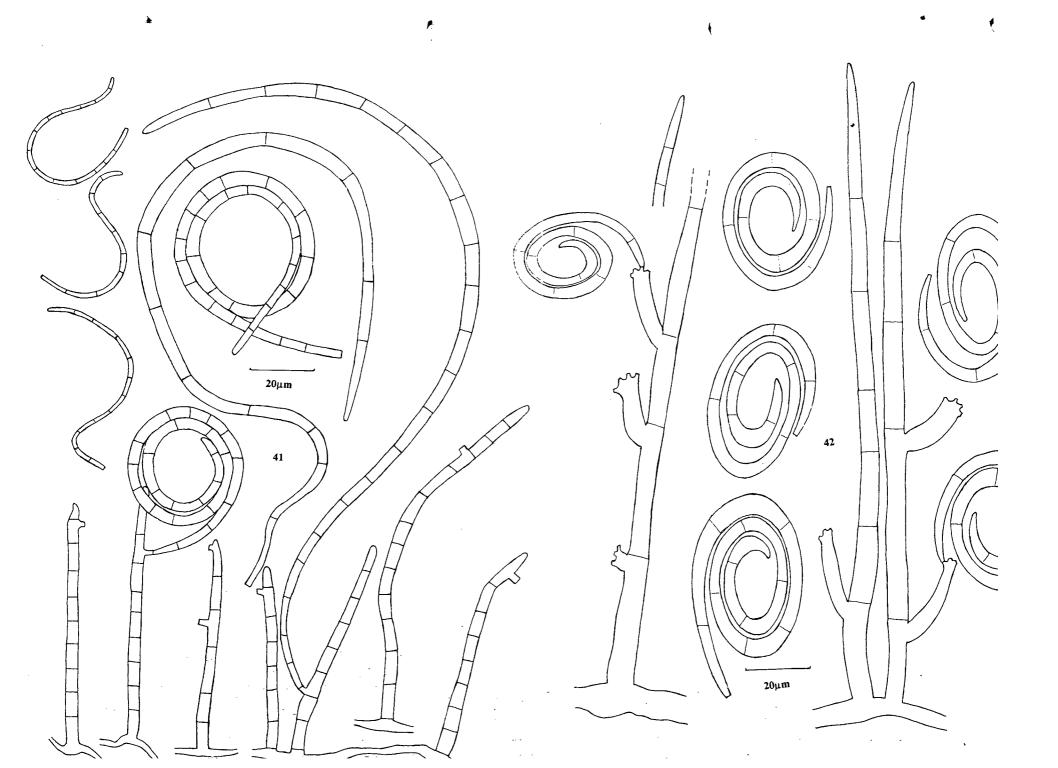
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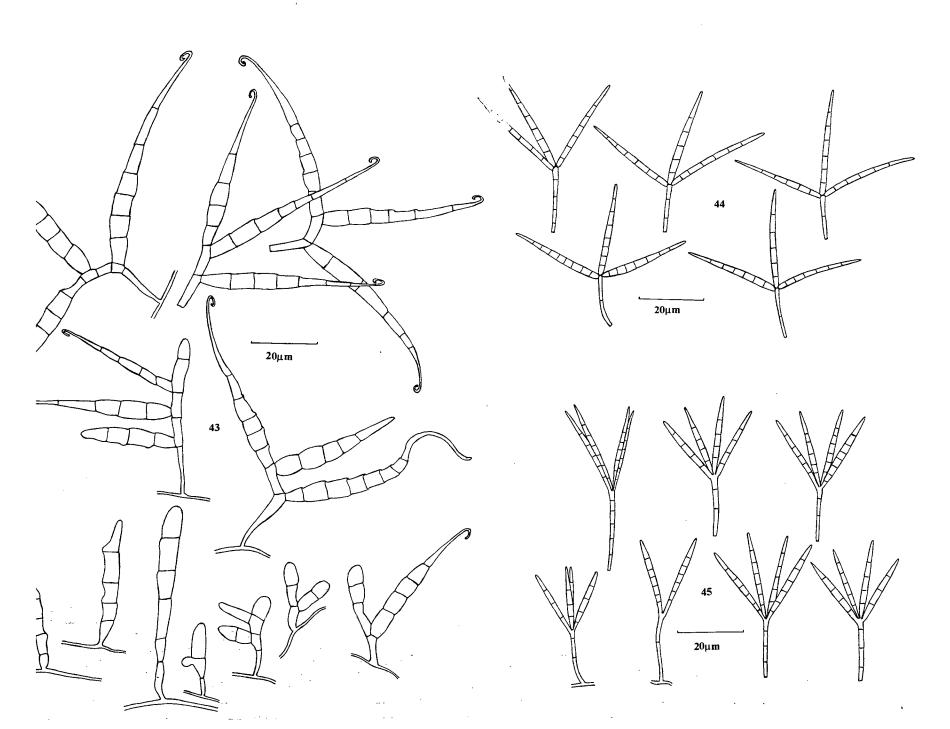




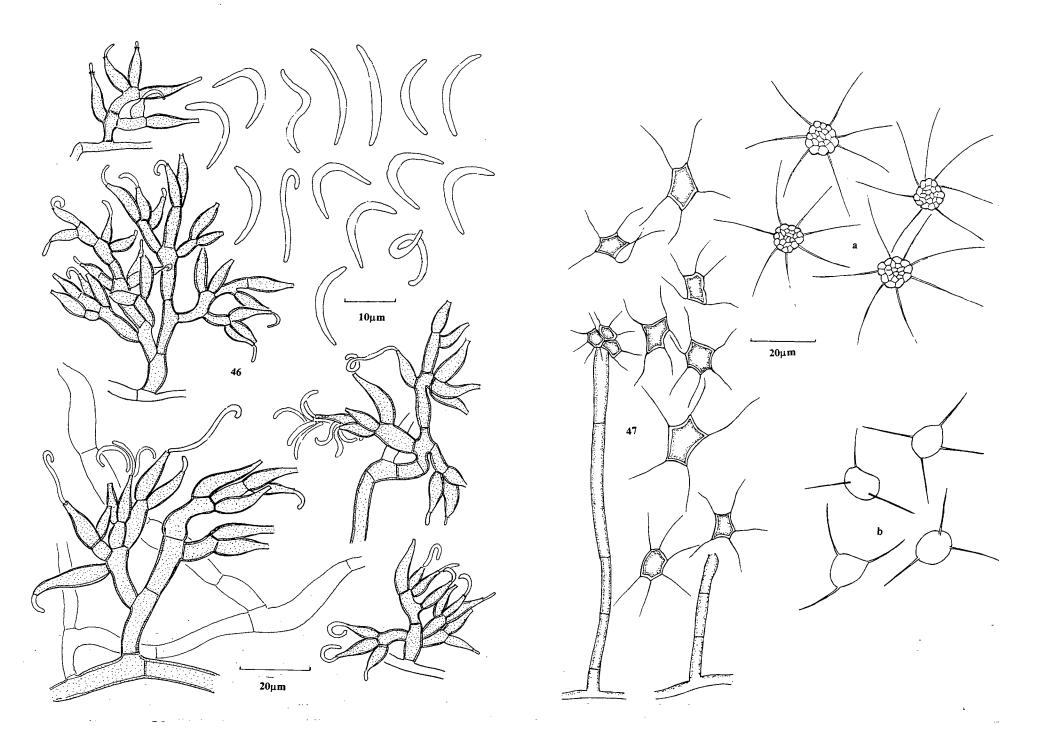
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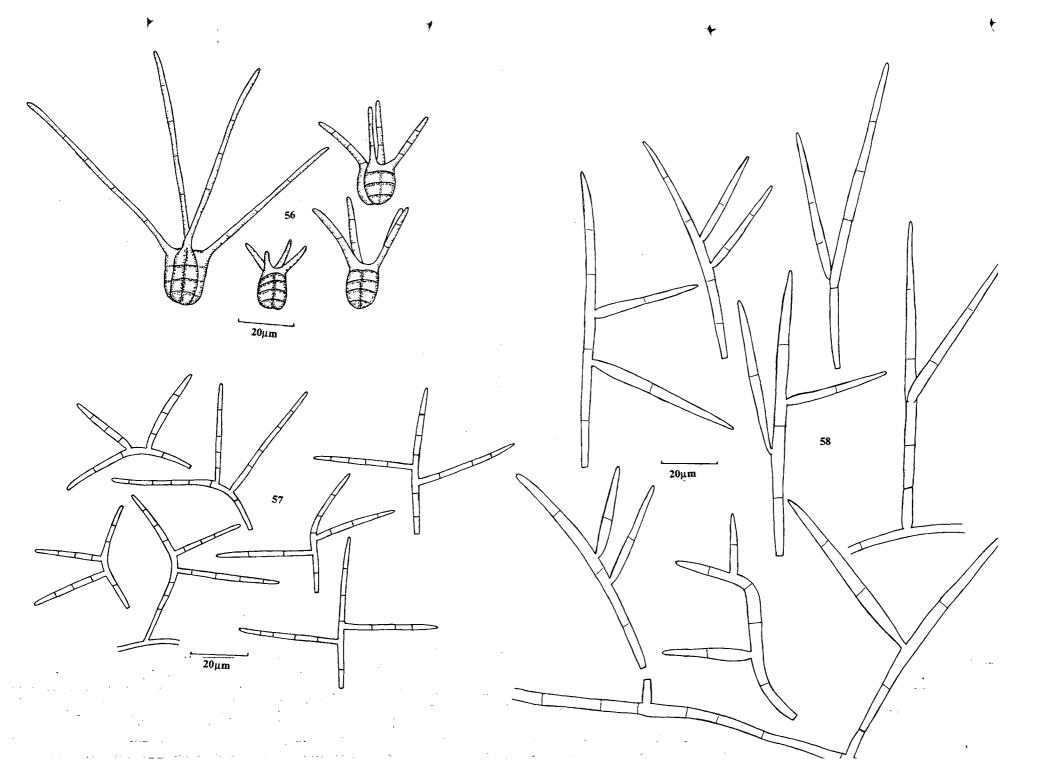


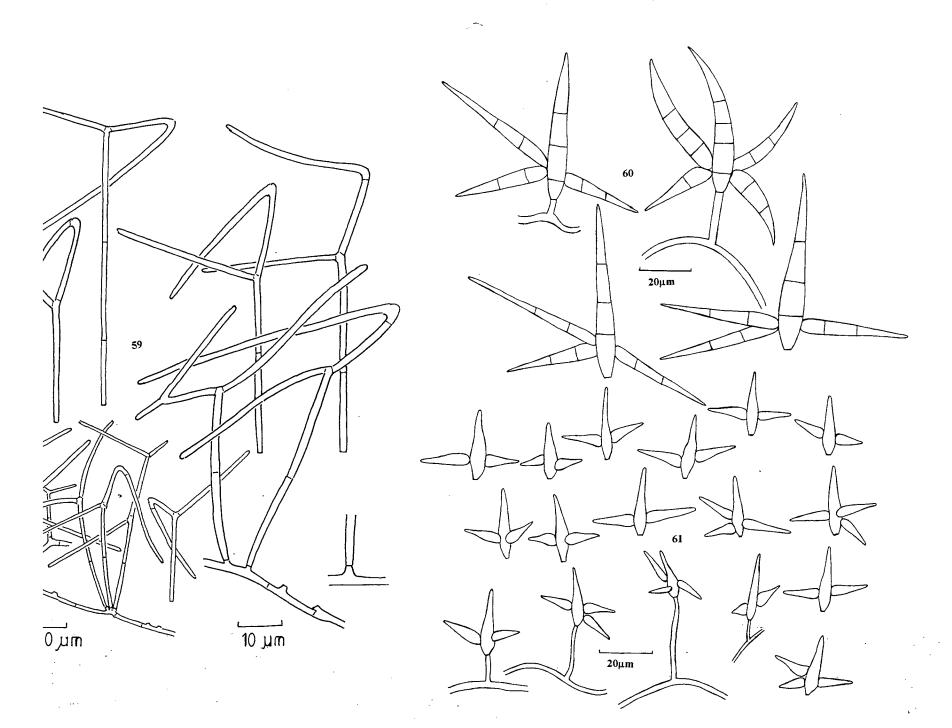


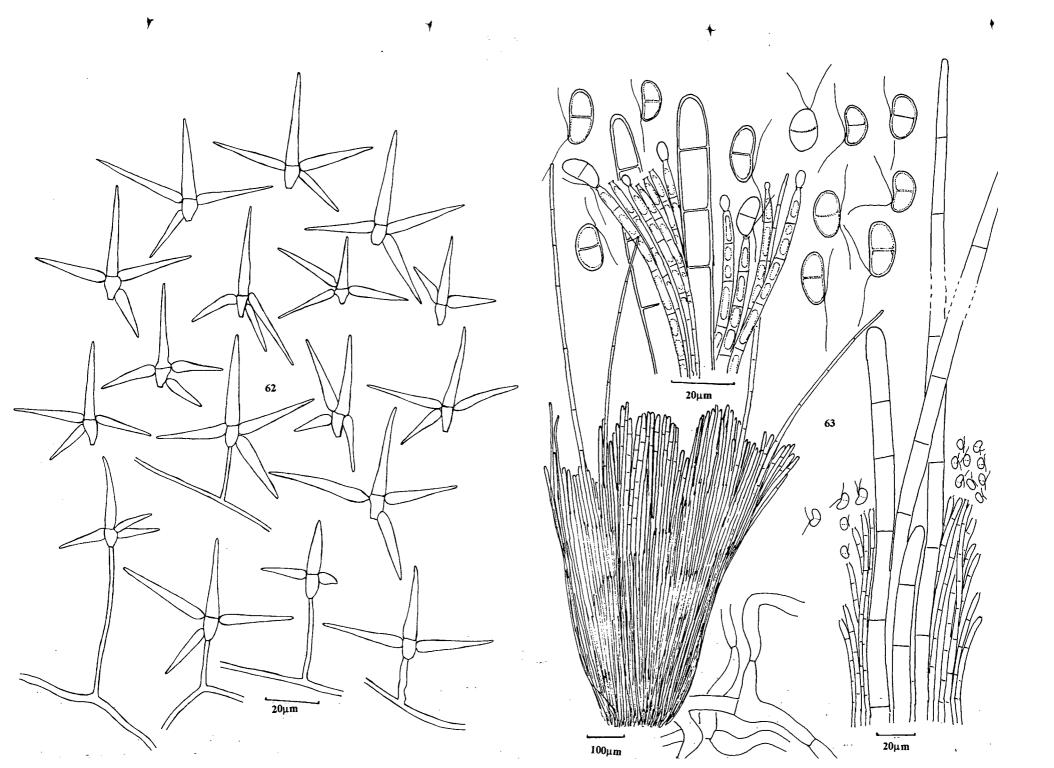


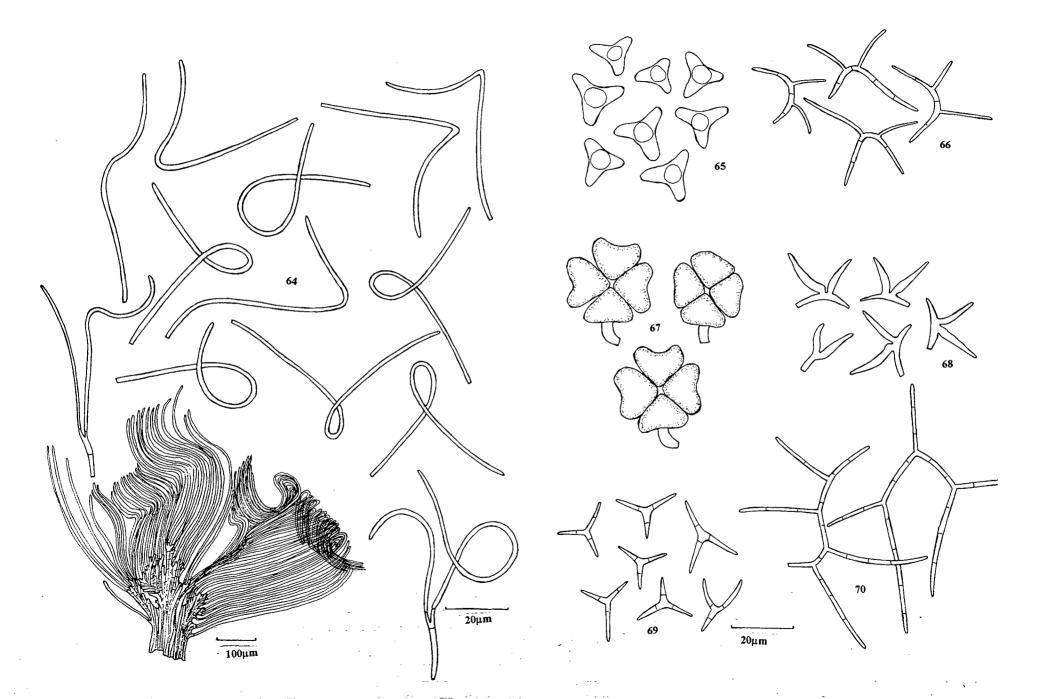
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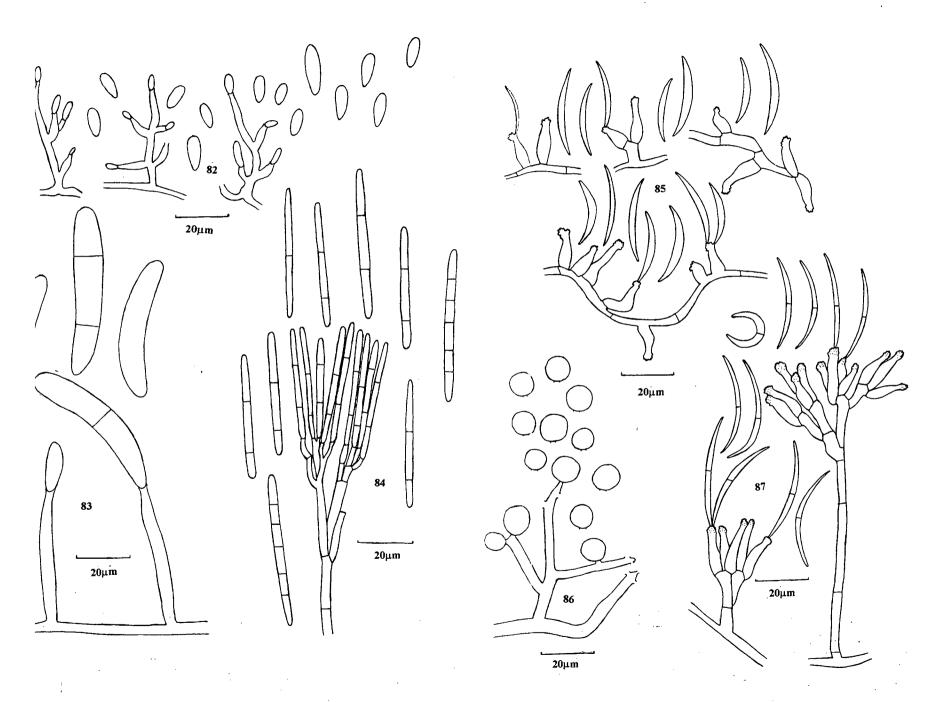








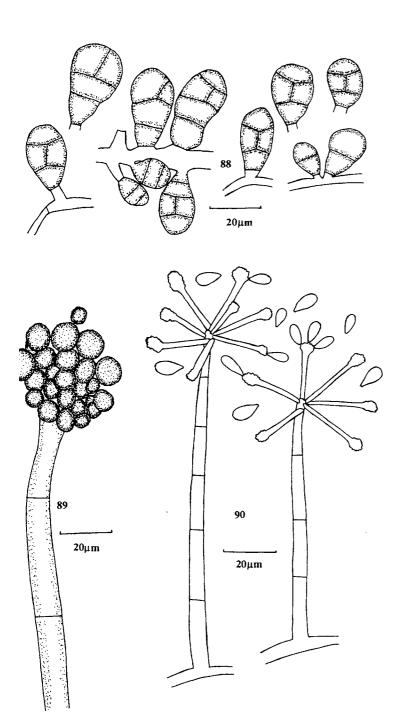
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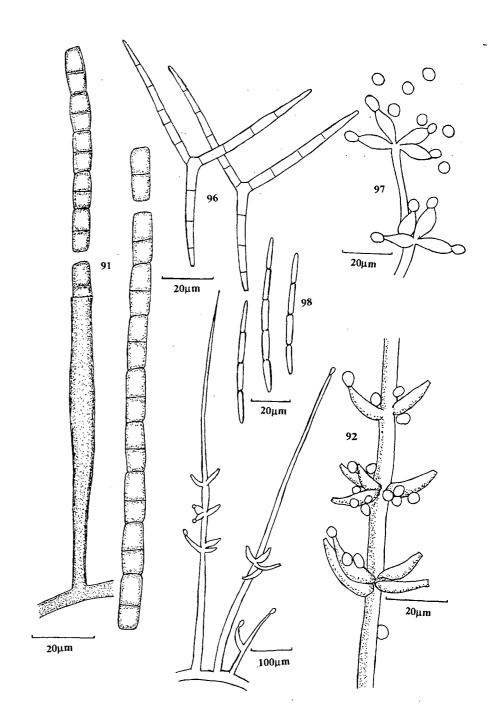


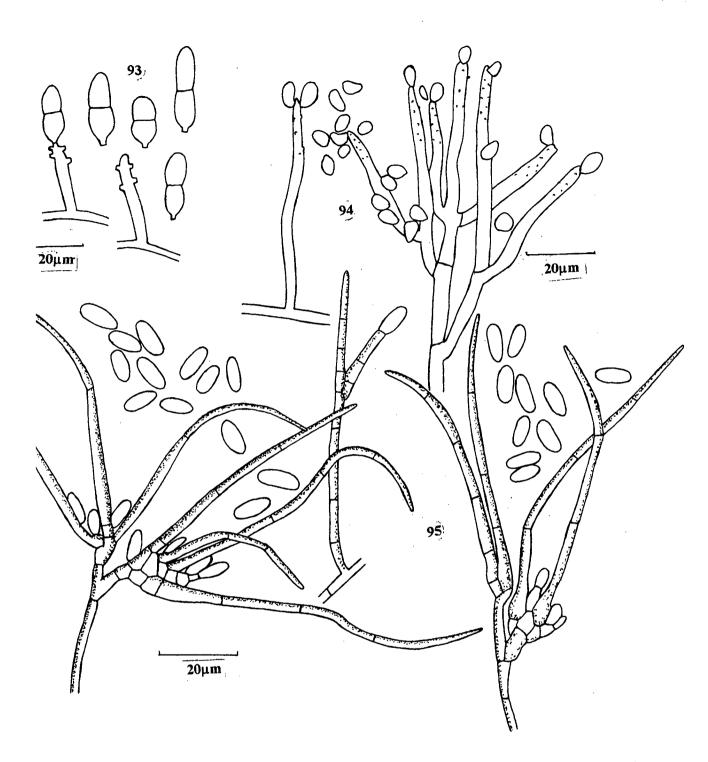
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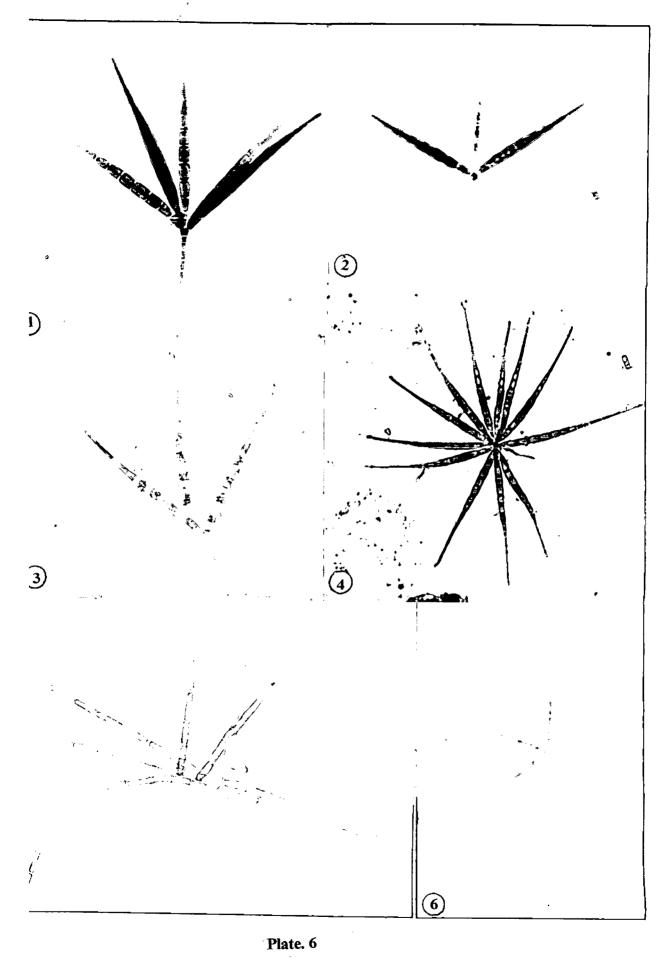






Single conidium of

- 1. Flabellospora verticillata
- 2. Isthmotricladia laeensis
- 3. Ingoldiella hamata
- 4. Flabellospra multiradiata
- 5. Dendrospora erecta
- 6. Alatospora acuminata



- 1. Actinospora megalospora single conidium
- 2. Ceratosporium sp. Conidiophores and conidia.
- 3. Triscelophorus monosporus-conidiophores and conidia.
- 4. Helicopsorium sp. -conidiophores and conidia.
- 5. Helicomyces roseus conidiophores and conidia.

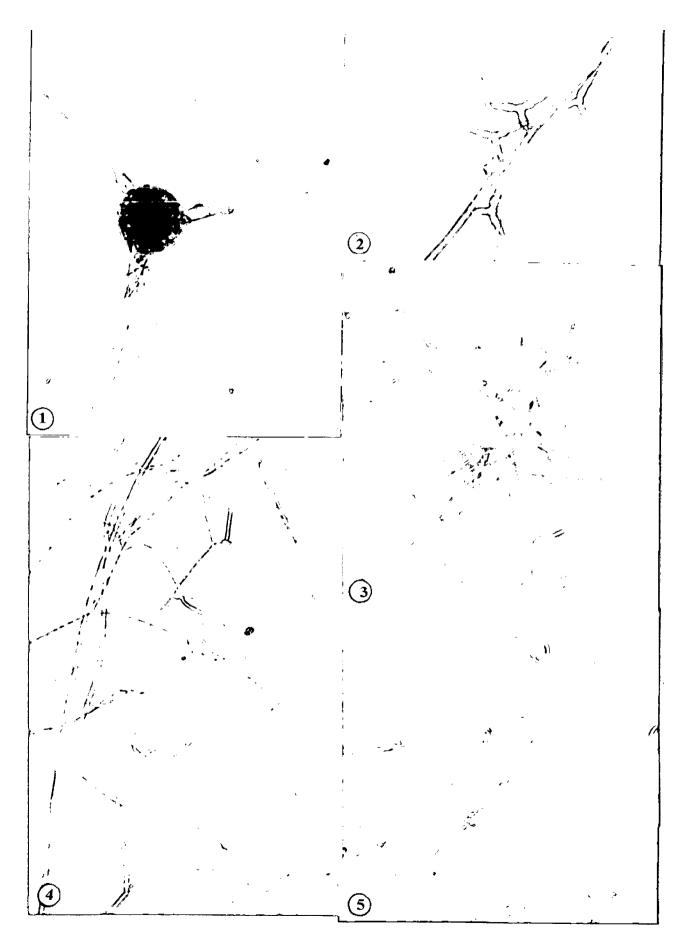


Plate. 7

- 1. Tricladium splendens single conidium
- 2. Anguillospora crassa single conidium
- 3. Diploospora indica conidiophore and conidia.
- 4. Anguillospora longissima single conidium
- 5. Undetermined ascomycete. sp.2

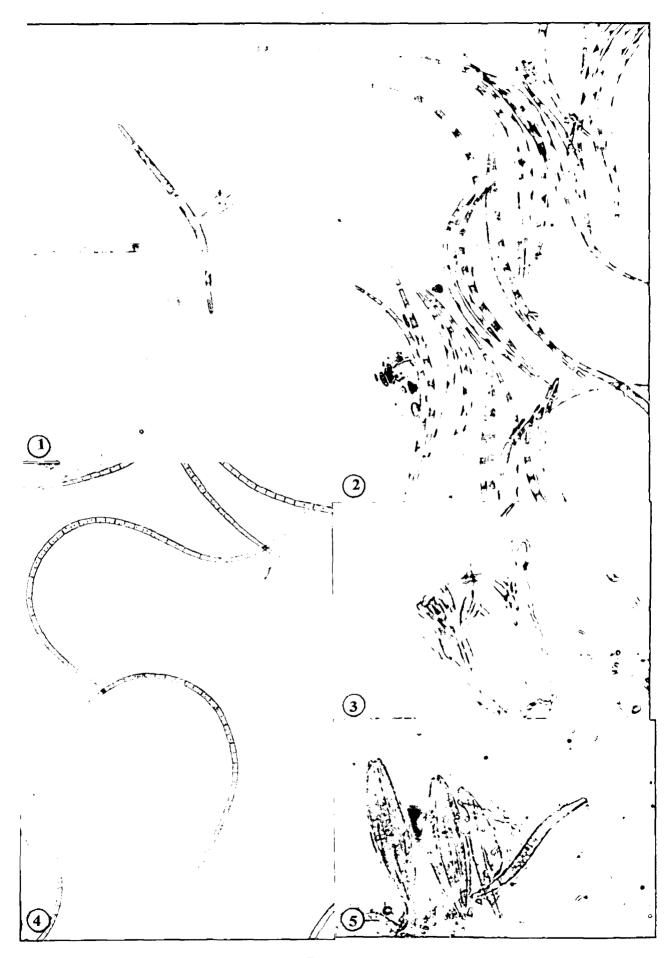


Plate. 8

- 1. Dichotomophthoropsis aquatica conidiophore and conidia.
- 2. Dichotomophthoropsis aquatica conidia.
- 3. Parathozetella aquatica sporodochium
- 4. Dictyochaeta assamica conidiophore and conidia.

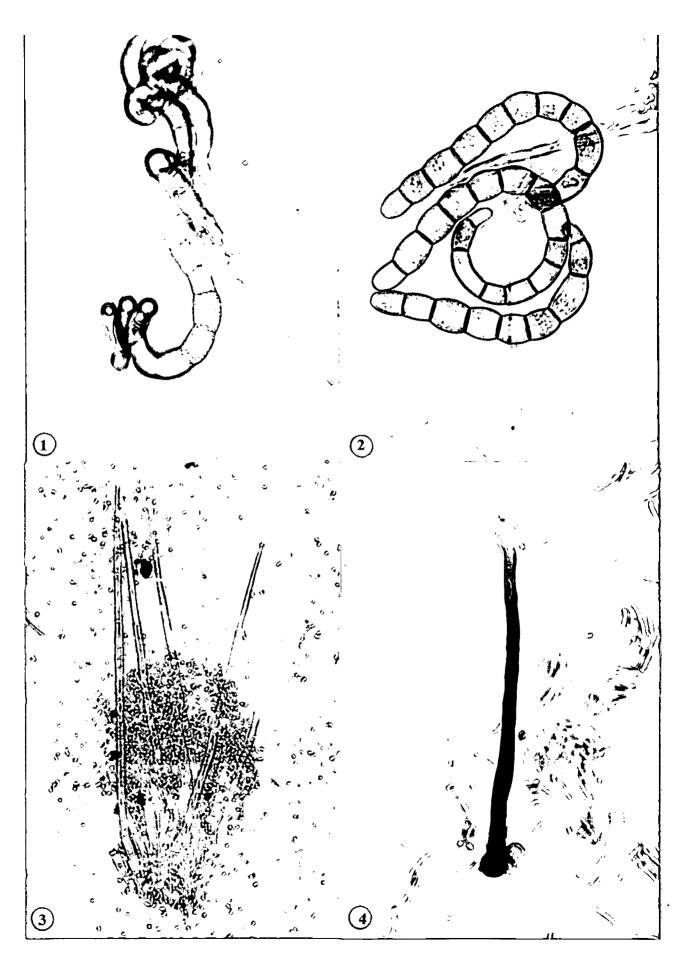


Plate. 9

- 1. Undetermined sp. 7 synnemata
- 2. Undetermined sp. synnema in close-up view.
- 3. Speiropsis hyalospora- conidiophores and conidia.
- 4. Ardhachandra sp. conidiophores and conidia.



Plate. 10

- 1. Anguillospora longissima conidiophores and conidia.
- 2. Beltrania rhombica conidiophores and conidia.
- 3. Canalisporium caribense sporodochium
- 4. Ingoldiella hamata developing conidia.
- 5. Undetermined sp. 9
- 6. Undetermined sp.1

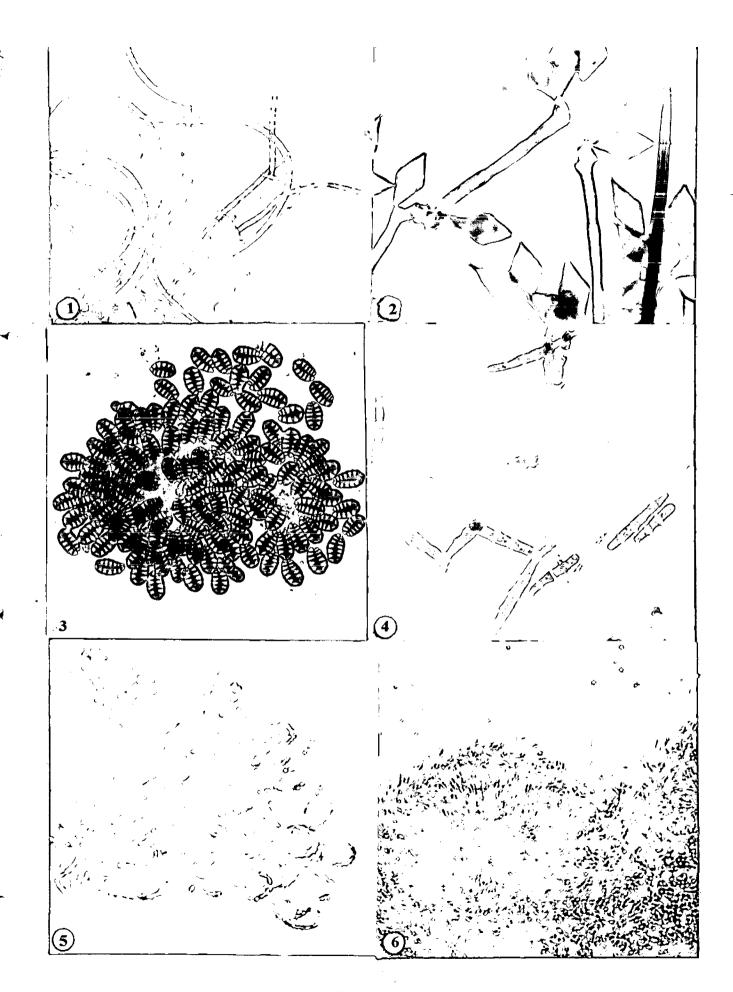


Plate. 11

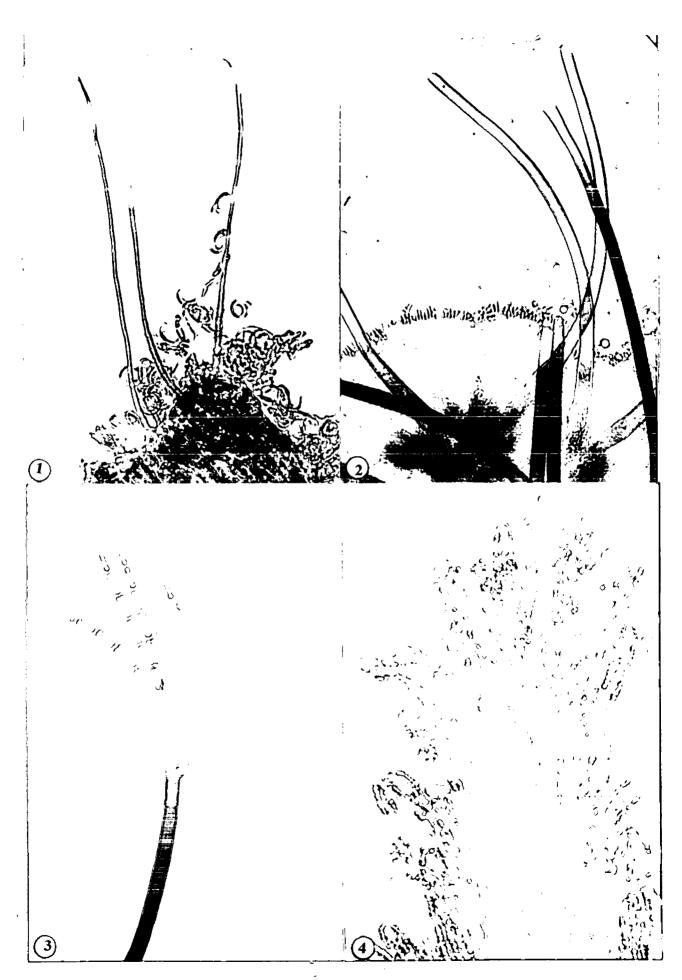


Plate. 12

<u>PART-II</u>: SEASONAL APPEARANCE AND ABUNDANCE OF AQUATIC FUNGI

The presence and density of aquatic fungi in a stream ecosystem in different seasons of the year are topics of considerable significance from ecological point of view. The occurrence of aquatic fungi on a substrate at a given time indicates that the fungi grow and colonize and are able to degrade them. This is true with all kinds of plant litter, be it terrestrial or aquatic (Dix and Webster, 1995).

2.1. SEASONAL APPEARANCE OF AQUATIC FUNGI IN 3 STREAMS OF GOA:

To study the abundance and periodicity of occurrence of aquatic fungi on submerged leaf litter in different seasons, one stream each in three wildlife sanctuaries of Goa - Bondla, Cotigao and Molem were chosen. The sites are indicated in Figure 3.1 and described in detail in the materials and methods. All the three sites had dense riparian canopy. Fallen and submerged leaves were collected at each site, brought to the laboratory in sterile polythene bags in an ice pack container and subjected for sporulation.

Aquatic fungi sporulate under water, when conditions are favourable. Of the decayed and well-skeletonised leaf litter brought to the laboratory, 2 to 3 leaves were washed thoroughly in deionized tap water and placed in large specimen jars containing 1 L sterile distilled water. The jars were aerated continuously for 3-7 days using a fish-tank aerator. The aerated water was filtered through a millipore filter (8 µm pore size) and the aquatic spora collected on the filter were counted (Iqbal and Webster, 1973).

The density of fungi on leaf litter, as expressed in water by aquatic spora, during the three different seasons, monsoon (M), post-monsoon (PM) and summer (S) is given below in Table 4.2.1. and their significance of occurrence, statistically analysed using 'Analysis of Varience' (ANOVA) test, is described.

In all, conidia of 65 species of aquatic fungi were recorded in varying concentration in the aerated water. Fungal taxa seen as conidia in water and their average relative abundance (%) are given in the Table: 4.2.1 and Table: 4.2.4.

The relative abundance of species of fungi in 3 different seasons at three sites, namely Cotigao, Bondla and Molem indicates (Table 4.2.1.) indicates that during monsoon, highest number of species were at Bondla (55), followed by Cotigao (54) and Molem (48). During post-monsoon the species richness remained in the same order of abundance in three sites, i.e., Bondla (41), Cotigao (37) and Molem (31). However, the abundance was in different order in summer; highest number of species being at Cotigao (19), followed by Bondla (15) and Molem (15).

Analysis of Variance Test (ANOVA) on seasonal sampling carried out during the year 1999-2000 showed a highly significant variation in the species richness in three different seasons at three sites and the order of significance was as follows: Cotigao (4.47), Bondla (3.13) and Molem (2.45). Less significant variation was observed between the sites during the same seasons: Monsoon (1.69), Post-monsoon (1.18) and Summer (0.92).

The relative abundance of species of fungi in 3 different seasons at three sites, namely Cotigao, Bondla and Molem (Table 4.2.3.) indicated that during monsoon, the highest number of species were at Cotigao (55), followed by Bondla (50) and Molem (48).

During post-monsoon, the species richness remained in the same order of abundance at three sites, i.e., Cotigao (45), Bondla (38) and Molem (33). The order of abundance remained the same also in summer; i.e., highest at Cotigao (23), followed by Bondla (19) and Molem (18).

ANOVA Test showed that the concentration of conidia in water was significantly affected by 3 different seasons when compared to that of the 3 sampling sites. In the significant level tested between the three seasons, in three different sites, the F ratio obtained for Bondla, Cotigao and Molem was 7.9, 4.85 and 8.46 respectively. In all the three sites, the density and species abundance in three different seasons varied significantly.

The significant level tested between the places during same seasons did not show much variation. The F ratio obtained in monsoon, post-monsoon and summer from the three sampling sites were 1.84, 1.37 and 0.44 respectively. This similarity in significance may be attributed to the similar type of vegetation composition seen at three sites.

A total of 65 species of freshwater fungi were encountered on randomly sampled leaves, during the three different seasons. In both years (July 1999 - May 2000 and July 2000 - May 2001), highest number of species observed were during monsoon (from June to September) and the lowest during summer season (from February to May).

The density and average number of species recorded showed a negative correlation with the temperature. As the ambient temperature increased, the number and kinds of fungi in stream ecosystem decreased (Table 4.2.5). Nevertheless, *Anguillospora crassa*, *Campylospora chaetocladia*, *C. filicladia*, *Lunulospora curvula* and *Triscelophorus monosporus* were found to be the major colonizers of leaf material round the year.

Besides, the following species which occurred throughout the year, namely Chaetendophragmia triseptata, Dictyochaeta asamica, Helicomyces roseus, Pestalotiopsis sp. and Speiropsis pedatospora can be treated as predominant species in the regional aquatic system. It may also be said that presence of aquatic hyphomycetes is primarily dependent on the availability of stream water and suitable submerged substrates such as leaf litter in the stream.

The results also indicated that the density and number of species recorded did not show a significant correlation with the pH (Table 4.2.6). The species density varied irrespective of the near uniform pH of stream water.

As an overall analysis, it may be said that occurrence and species density of fungi of a given stream ecosystem are largely dependent on factors such as (i) rainfall and substrate availability or leaf deposition. The water temperature and pH had very little to impact on fungal presence. Similar results were obtained by Sridhar and Kaveriappa (1984, 1989) who did studies in a freshwater stream in Konaje, Mangalore.

2.2. ABUNDANCE OF AQUATIC FUNGI IN DIFFERENT STREAMS OF GOA:

The distribution and abundance of aquatic fungi as represented by conidia in water were significantly correlated with the sampling sites. The mean number of conidia was higher at the sampling sites of Bondla, Cotigao and Molem where the streams were covered by dense riparian canopy round the year. Less number of conidia and species of fungi were observed at the sites of Taleigao and Harvalem wherein very few riparian

plants were observed overhanging the streams. This clearly showed that distribution of fungi is directly correlated with the vegetation around.

No clearcut distribution pattern for the most abundant species was recognised in relation to stream water chemistry (pH and /or nutrients). The pattern of species richness seemed to be independent of the water chemistry. The distribution of fungi was more consistent in the presence of dense riparian canopy. Species richness was higher at all the three sites which are endowed with rich riparian canopy.

The present results suggest that abundance and diversity of conidia of aquatic fungi are primarily correlated with substrate availability rather than water chemistry (pH, alkalinity or inorganic nutrient concentration). Similar findings from temperate streams have been reported by other authors (Iqbal and Webster, 1977; Shearer and Webster, 1985; Barlocher, 1987; Chauvet, 1991; Mothe-Jean-Louis, 1997). Despite factors such as rainfall (Willoughby and Archer, 1973; Mothe-Jean-Louis, 1997), the abundance of aquatic fungi in many streams of deciduous forests seems to be well-correlated with the prevailing litter fall pattern.

A comparable phenomenon occurs in two sites lacking riparian canopy. The species density was less in Taleigao and Harvalem where the streams had sparce tree lining. The species richness was maximum at all the three sites with rich riparian canopy. It may be noted that in these three sites, not only the streams had good substrate availability but also were supported by dense terrestrial tree coverage. This can be seen by the presence of a substantially large number of 'casual aquatics' in these sites. The spora were washed into the stream mostly by flash floods.

From the results, it can be inferred that dissolved nutrients as well as pH or

alkalinity, seems to be of minor importance determining the abundance of aquatic fungi. The site with higher Sodium (Na) and Potassium (K) status, Cuncolim, showed a relatively less fungal species diversity (Fig.4.3).

The results obtained from rare fraction studies (Fig. 4.3) point out that rainfall and temperature did not influence much the aquatic species diversity. It is clear that species richness largely regulated by the prevailing litter fall. The richness indices R1 and R2 showed decrease in the number of species between Cotigao (35) and Taleigao (10). The expected number of species calculated for each sampling site [E(S_n)], with regard to the data obtained, showed a drastic decrease relating to the canopy cover in the surroundings of the site. Higher number and density of species were seen in all the three wildlife sanctuaries when compared to other collecting sites. Although higher concentration of individuals was found in Molem, as shown in the rarefaction curve, when the sample size was 150, the expected number of species for Cotigao sample is 32 and that of Molem 22. The diversity index slided from Cotigao to Molem. This may be attributed to the continuous (100%) tree canopy found along the streams of Cotigao.

Table 4.2.1: Relative abundance (%) of fungi at sampling sites during July1999-May 2000.

		Bondla	l.	(Cotigao			Molem	
Name of fungus	M	PM	S	M	PM	S	M	PM	S
						-			
Actinospora megaolospora	0.46	0.09	-	0.19	0.05	-	0.17	-	-
Alatospora acuminata	0.93	0.49	-	0.47	0.39	0.05	1.15	0.39	_
Anguillospora crassa	3.17	1.22	0.59	3.12	0.74	0.29	1.82	1.24	-
Anguillospora furtiva	1.05	0.46	_	1.38	0.92	0.42	-	-	-
Anguillospora longissima	1.75	0.95	0.16	2.30	2.20	0.49	1.02	0.53	0.22
Ardhachandra selenoides	3.89	2.24	1.88	3.41	1.36	1.19	11.23	7.73	3.46
Ardhachandra sp.	0.39	-	-	1.09	0.57	-	-	-	-
Articulospora tetracladia	0.19	0.03	-	0.29	0.12	-	0.39	0.18	-
Bahusutrabeeja dwaya	0.82	0.63	-	0.17	-	-	-	-	-
Beltrania rhombica	0.19	0.19	-	2.13	1.04	0.59	0.79	0.75	0.35
Camposporium pellucidum	1.45	0.59	0.19	1.83	0.92	0.62	1.24	0.04	-
Campylospora chaetocladia	0.75	0.33	-	3.54	1.86	-	3.24	0.62	-
Campylospora filicladia	0.93	0.36		3.19	1.31	0.42	3.46	1.11	0.44
Centrospora acerina	0.53	0.06	-	-	-	-	-		-
Ceratosporium sp.	0.63	0.29	_	-	-	-	-	_	-
Chaetendophragmia triseptata	1.92	0.85	0.29	1.61	1.04	0.29	3.06	0.84	0.13
Condylosporda spumigena	1.78	0.59	-	3.04	0.64		1.28	1.06	_
Dactylaria sp.	0.56	0.06	-	-	-	-	-	-	_
Dactylella ellipsospora	-	-	-	0.47	0.29	0.19	0.53	0.08	_
Dendrospora erecta	0.53	0.16	_	0.42	0.17	-	0.44	0.13	-
Dendrosporium lobatum	-		-	-	-	-	1.99	0.79	
Dictyochaeta assamica	4.36	_		5.25	1.78	1.38	8.21	1.55	0.75
Diplocladiella scalaroides	2.47	0.76	0.29	4.16	2.03	0.82	2.39	0.88	
Flabellospora crassa	0.49	-	-	0.59	•		0.53		-
Flabellospora multiradiata	1.25	0.69	-	1.53	0.67	•	0.84	0.88	
Flabellospora verticillata	0.92	-	-	2.15	1.06	0.37	0.53	0.35	0.26
Flagellospora curvula	0.46	0.56	-	0.69	-	0.39	0.75	0.35	0.22
Helicomyces roseus	0.86	0.69	-	0.29	0.19	0.12	0.48	-	-
Helicosporium sp.1	0.39	0.06	0.09	0.05	0.19	0.09	0.62	0.22	-
Helicosporium sp.2	0.16	0.06	-	0.07	-	-	0.39	-	-
Helicosporium sp.3	0.49	-	_	0.12	0.27	-			
Ingoldiella hamata	-	-	_		-	-	1.19	0.57	0.09
Isthmotricladia britanica	-	-	-	0.12	0.09	-	-	_ -	-
Isthmotricladia laeensis	2.84	1.88	_	1.46	0.29	-	1.24	0.66	0.08
Lateriramulosa uniinflata	0.96	0.33	_	-		-		-	
Lemonniera aquatica	0.86	0.36	0.16	0.32	0.05	-	0.35	0.13	
Lunulospora curvula	0.43	0.29	0.19	0.22	-		0.66	0.35	0.08
Nawawia filiformis	0.49	0.39	0.09	0.72	0.12	_	0.35	-	-
Nawawia sp.	0.56	0.26	-	0.05	-	-	-		-

Pestalotiopsis sp.	8.45	6.97	0.92	6.23	2.87	_	2.57	-	-
Phalangispora constricta	1.75	0.36	0.26	0.77	0.29		0.84	0.53	0.18
Robillarda phragmitis	3.23	1.92	-	2.00	-	-	0.22	1	
Scutisporus brunneus	2.08	1.16	-	1.68	1.26	-	0.93		-
Seimatosporium sp.	0.56	0.26	-	0.05	0.12	-	1.99	1.15	-
Speiropsis hyalospora	-	-	-	1.46	-	0.05	-		-
Speiropsis pedatospora	0.53	0.49	0.36	0.69	0.29	0.19	0.84	0.44	0.13
Subulispora sp.1	0.66	0.36	-	0.29	0.19			-	-
Tetrachaetum elegans	0.19	0.19	•	0.22	0.05	-	0.13	0.04	
Tetraploa aristata	0.49	-	•	0.54	0.35	-	0.26		-
Tricladium gracile	0.69	0.39	0.26	0.72	0.37	-	0.44	0.04	-
Tricladium splendens	0.46	0.16	-	0.07		-	0.35	0.31	0.08
Trinacrium indica	0.65		_	-	-	-		-	
Trincarium sp.	0.39	•	-	0.42	-	-	0.84		-
Tripospermum myrti	0.23	-	-	0.05	_		0.79		-
Triscelophorus acuminatus	0.63	-	-	0.39	-				-
Triscelophorus konajensis	0.23	-	-	•	-	-	0.53	-	_
Triscelophorus monosporus	3.17	0.63	0.36	1.28	0.64	0.17	3.86	0.66	0.26
Weisneriomyces javanicus	-	-	-	0.29		-	0.62	-	-
Undetermined sp.1	-	-	-	0.39	-	-	-	-	-
Undetermined sp.2	0.39	-	-	-	-	-	-	_	-
Undetermined sp.3	-	-	-		-	-	0.22		-
Undetermined sp.4	0.26	-	-	-	-	-	-	-	-
Undetermined sp.5	-		-	-		<u>-</u>	0.39		_
Undetermined sp.6	-	-		0.29	-	-	0.35	-	-
Total No. of Species	55	41	15	54	37	19	48	31	15

Table: 4.2.2. Analysis of Variance (ANOVA) for 1999-2000:

Places and	Seasons		Experimental Method									
		Sum of squares	Degree of freedom	Mean Square	F ratio	Signifi- cance						
Bondla	Between seasons	8604.65	2	4302.33	3.13	Yes						
	Residual	148591.24	108	1375.84								
Cotigao	Between seasons	16697.25	2	8348.63	4.47	Yes						
	Residual	199862.1	107	1867.87								
Molem	Between seasons	6875.37	2	3437.69	2.45	Yes						
	Residual	127646.46	91	1402.71								
Monsoons	Betweeen places	7979.44	2	3989.72	1.69	No						
	Residual	361992.46	154	2350.60								
Post-Monsoons	Between places	2281.51	2	1140.79	1.18	No						
	Residual	102324.58	106	965.33								
Summer	Between places	469.65	2	234.83	0.92	No						
	Residual	11782.76	46	256.15								

Table 4.2.3: Relative abundance (%) of fungi at sampling sites during June 2000-May 2001.

	T	Bondla	ì		Cotigao		Molem		
Name of fungus	M	PM	S	M	PM	S	M	PM	S
	1			<u> </u>					
Actinospora megaolospora	0.17	_	-	0.40	0.16	_	0.91	-	-
Alatospora acuminata	0.63	0.27	-	0.77	0.50	0.23	0.61	0.56	-
Anguillospora crassa	0.51	0.46	0.23	0.84	0.43	0.06	0.86	-	0.10
Anguillospora furtiva	0.68	-	-	1.57	0.60	-	-	-	
Anguillospora longissima	4.73	1.77	0.46	1.94	1.20	0.20	1.47	0.10	0.10
Ardhachandra selenoides	1.48	1.14	1.03	6.61	5.14	2.61	2.64	-	0.30
Ardhachandra sp.	-	-	-	2.07	0.04	0.10	-	-	-
Articulospora tetracladia	0.28	0.11	-	0.87	0.30	_	0.81	0.20	-
Bahusutrabeeja dwaya	0.97	-	0.34	0.40	0.27	-	-	-	-
Beltrania rhombica	3.08	0.57	0.51	1.10	0.37	0.63	1.32	-	0.46
Camposporium pellucidum	1.08	0.28	<u> </u>	0.97	0.50	0.27	2.33	0.61	<u> </u>
Campylospora chaetocladia	2.79	1.48	0.57	0.87	0.67	0.17	3.45	1.83	0.35
Campylospora filicladia	3.31	2.22	0.34	3.14	1.44	0.40	1.72	0.91	0.76
Centrospora acerina	0.68	0.51		0.73	0.43	-	-	-	_
Ceratosporium sp.		-	<u> </u>	0.50	0.07		-	-	-
Chaetendophragmia triseptata	3.31	2.17	0.63	2.44	1.64	0.40	6.38	3.25	0.61
Condylosporda spumigena	1.48	0.51	-	1.78	0.40	0.07	1.77	0.30	
Dactylaria sp.	1.19	1.03	0.51	-	_		2.78	0.86	-
Dactylella ellipsospora	0.46	-	-	-	_	-	1.32	0.40	0.10
Dendrospora erecta	0.74	0.29	-	0.50	0.30	-	0.66	0.10	-
Dendrospora yessemreddia	0.63	-	-	-	_	-	-	-	-
Dendrosporium lobatum	-	-	-	-	_	-	2.99	1.22	0.61
Dictyochaeta assamica	3.31	1.65	0.34	8.55	1.80	0.37	1.93	2.28	0.81
Diplocladiella scalaroides	2.17	0.91	0.28	1.40	0.60	-	3.24	0.51	-
Flabellospora crassa	-	0.85	-	0.70	0.17	-	0.91	-	-
Flabellospora multiradiata	2.39	0.86	-	1.07	0.20	_	2.74	1.01	-
Flabellospora verticillata	0.51	0.17	-	0.40	0.20	-	0.25	0.10	-
Flagellospora curvula	3.07	0.91	0.39	0.87	0.43	_	0.61	0.71	0.30
Helicomyces roseus	0.68	0.11	-	0.53	0.27	0.17	0.71	1.06	0.25
Helicosporium sp.1	0.34	-	-	0.53	-	-	0.35	0.15	-
Helicosporium sp.2	0.51	0.23	-	-	0.17	0.07	_	-	-
Helicosporium sp.3	-	-	-	0.20	0.07	-	0.41	0.05	-
Ingoldiella hamata	-	-	-	_	-	-	1.62	1.47	0.30
Isthmotricladia britanica	-	-	-	0.07	-	-	-	-	-
Isthmotricladia laeensis	0.68	0.23		0.77	0.43	-	1.27	0.96	-
Lateriramulosa uniinflata	0.46	0.11	-	0.40	0.16	_	-	-	-

Lemonniera aquatica	0.97	0.86	-	0.86	0.67	<u> </u>	0.61	0.41	-
Lunulospora curvula	1.48	0.63	0.51	1.00	0.43	<u> </u>	1.57	0.81	0.61
Mycoleptodiscus indicus	-	-	-	0.33	-				<u> </u>
Nawawia filiformis	0.68	0.46	0.23	0.60		<u> </u>	0.81	-	-
Nawawia sp.	0.17	-	-	0.17	0.13	0.06	0.46	-	-
Pestalotiopsis sp.	3.82	2.57	1.08	3.10	1.80	0.93	4.26	0.61	-
Phalangispora constricta	0.91	0.39	0.11	0.93	0.50	<u> </u>	1.62	0.66	0.46
Robillarda phragmitis	1.08	-	-	1.64	0.70	0.57	1.32	0.91	0.81
Scutisporus brunneus	0.46	0.11	-	0.77	0.57	<u> </u>	0.81	0.20	-
Seimatosporium sp.	0.34	0.51	-	0.17	0.07	<u> </u>	0.56	-	<u> </u>
Speiropsis hyalospora	T-	-	-	0.94		<u> </u>	-	<u> </u>	ļ -
Speiropsis pedatospora	1.65	0.34	0.17	1.77	1.37	0.07	1.01	0.35	<u> </u>
Subulispora sp.1	0.28	-	-	-		<u>l </u>			-
Tetrachaetum elegans	0.91	0.11	-	0.40	0.27	<u> </u>	0.46	0.05	-
Tetraploa aristata	0.28	0.11	-	0.57	_	0.10	0.30	-	
Tricladium gracile	1.19	0.63	0.23	0.94	0.33	-	0.96	0.30	
Tricladium splendens	0.91	0.28	_	0.73	0.47	0.07	0.46		-
Trincarium sp.	0.74	-	-	0.50	0.07	-	-	-	
Tripospermum myrti	-	-	_	0.77	0.50	0.17	<u> </u>	<u> </u>	-
Triscelophorus acuminatus	1.03	0.11	-	0.13	0.07		0.66	-	<u> </u>
Triscelophorus konajensis	0.34	-	-	-	-	-	0.35	-	
Triscelophorus monosporus	2.74	1.19	0.85	1.87	1.43	0.43	4.72	1.22	0.30
Weisneriomyces javanicus	0.51	-	-	0.60	-	0.13	0.61	0.15	0.10
Undetermined sp.1	T-	-	_	0.37] -	-	-		-
Undetermined sp.2	0.46	Ţ -	_	-	-	-	<u> </u>	<u> </u>	<u> </u>
Undetermined sp.3	1-	-	-	_	-	-	0.61		-
Undetermined sp.4	-	T -	-	0.57	T -	_	-		-
Undetermined sp.5	-	-	-	0.10	-	-	0.25	-	-
Undetermined sp.6	-	-	-	-	-	-	0.30	-	-
Total no. of species	50	38	19	55	45	23	48	33	18

. ,

Table: 4.2.4. Analysis of variance (ANOVA) for 2000-2001:

Places/			Exper	imental metho	<u>od</u>	
<u>Seasons</u>				- _Y		
		Sum of	df	Mean	F	Sig.
:		squares		Square		
Bondla	Between	3618.36	2	1809.18	7.9	Yes
	seasons					
	Residual	23815.15	104	228.99		
Cotigao	Between	10903.96	2	5451.98	4.85	Yes
	seasons					
	Residual	134853.7	120	1123.78		
Molem	Between	6669.42	2	3334.71	8.46	Yes
	seasons					
	Residual	37843.93	96	394.21		
Monsoons	Betweeen	3725.84	2	1862.92	1.84	No
	places					
	Residual	151845.94	150	1012.31		
Post-Monsoons	Between	925.37	2	462.69	1.37	No
	places					
	Residual	38149.46	113	337.61		
Summer	Between	101.55	2	50.775	0.44	No
	places					
	Residual	6517.38	57	114.34		

Table 4.2.5: Temperature in different seasons at three sites during 1999-2000:

	-	1999		<u>2000</u>					
	M	PM	S	M	PM	S			
Bondla	23	25.4	32	23.5	24.5	29.5			
Cotigao	22	26	28	20	25	27.8			
Mollem	24	25.8	29	24	26	30			

Table 4.2.6: pH in different seasons at three sites during 1999-2000:

		1999		<u>2000</u>					
	M	PM	S	M	PM	S			
Bondla	5.7	6	6	5.5	5.8	5.7			
Cotigao	6.3	6.5	6.2	6.0	6.5	6.3			
Mollem	5.8	5	5.5	4.5	6	5.5			

Table 4.2.7: Aquatic fungi in different streams of Goa:

Name of fungus	<u>S1</u>	S2	S3	<u>S4</u>	<u>S5</u>	<u>S6</u>	S7	S8	<u>S9</u>	S10	S11	S12
Ivainc of fungus)	<u>=</u>			-					_		
Activopanous messalassass		+	+					-		+	_	
Actinospora megaolospora	-		+	+	+		+		+	+		<u> </u>
Alatospora acuminata	+	-		 					+			- +
Anguillospora crassa		+	+	-	+	+	-	- -	Т	+		
Anguillospora longissima	+	-		+	+	-	-			+	+	-
Ardhachandra selenoides		-	+	+	-	+		+	-			
Articulospora tetracladia	-	+	-	-	+		-	-	-	+	-	
Beltrania rhombica	+		+	-	+	+	+		-	-		-
Camposporium pellucidum	+	+	+	+	+		+	-	+	+	+	+
Campylospora chaetocladia		+	-	-	-	+	-	+	-	+	-	+
Campylospora filicladia	+	+	+	+	+	-	+		+	+	· +	
Centrospora acerina			-	+	+		-	-	-	<u>-</u>	-	-
Ceratosporium sp.			-	<u> </u>	+	-		-	-	+	-	_
Chaetendophragmia triseptata	+	+	+	-	+	+		+	+	-	-	+
Condylosporda spumigena	<u> </u>	+	+		+	-	+	-	<u> </u>	+		+
Dactylaria sp.1	-	<u> </u>		-	-		-	-		+ .	<u> </u>	-
Dactylella ellipsospora		+	<u> </u>			-		-	+	+		+
Dendrospora erecta			+		+		<u> - </u>		-	+		+
Dendrospora yessemreddia			+		-		-			-	-	-
Dendrosporium lobatum			-	-	-		-	-		+		-
Dictyochaeta assamica	+	-		+	-		+	+	+		+	-
Diplocladiella scalaroides	+	-	+	+	+	+		<u> </u>		+	-	+
Flabellospora crassa	T +	_		+	-				-	-		
Flabellospora multiradiata	-	+	+	-	+	-	+		+	+	-	+
Flabellospora verticillata	+	T -	+	+	+	-	-	+	-	+	+	
Flagellospora curvula	+	+	+	-	+	_	-			+	-	+
Helicomyces roseus	+	+	-	+	_	-	+	-	+	_	-	_
Helicosporium sp.1	-	+	+	-	-	-	-	-	-	+	+	-
Helicosporium sp.2	+	-	-	+	_	-	-	-	-	-	-	+
Helicosporium sp.3	-	+	+	-	-	-	+	T -		-	-	_
Ingoldiella hamata	-	-	-	-	-	-		-	_	+	-	-
Isthmotricladia britanica	† -	-	-	-	+	-	T -	-	<u> </u>	-	-	_
Isthmotricladia laeensis	+	+	+	+	+	-	+	+	+	+	+	-
Lateriramulosa uniinflata	+	-	-	-	+	-	-	 -	-	-	-	-
Lemonniera aquatica	1 -	+	-	-	+	+	T -	 -	T -	+	-	-
Lunulospora curvula	+	 -	+	+	+	+	-	+	-	+	_	+
Mycoleptodiscus indicus		+	† -	-	 	-	 	-	-	-	+	-
Nawawia filiformis	† -	-	-	 -	+	-	-	-	+	+	-	-
Pestalotiopsis sp.	 	+	<u> </u>	+	 _	+	-	-	+	+	-	<u> </u>
Phalangispora constricta	+-	+ +	+	† <u>-</u>	+	† -	 -	-	_	+	-	-
Robillarda phragmitis	+	+	 	+-	<u> </u>	+ +	+	+	+	 -	 -	_
Scutisporus brunneus	'-	 	+	+-	+	<u> </u>	 	+-	† <u>-</u>	+	1 -	-
Seimatosporium sp.	+	+	+ -	+	 	+	+-	+	+-	 	+	 _
	+-	-	+	+ -	 -	 	+-	 	+-	-	† <u>-</u>	-
Speiropsis hyalospora	+	+	+	+	+	+	+-	 -	+-	+	 -	 -
Speiropsis pedatospora	+		† 	+	+ -	+-	+	+-	 - -	+	+ -	 -
Subulispora sp.1		1-	1	<u> </u>				1 -			<u> </u>	т

Subulispora sp.2	-	-	-	-	+	•	-		-	-		-
Tetrachaetum elegans	-	+	-	+	+	-	•	+	-	+	-	+
Tetraploa aristata	+	+	-	1	+	+	ı	+	-	+	+	-
Tricladium gracile	T -	_	+	-		+	1	•	+	+	-	-
Tricladium splendens	T	+	-	+	+	•	-	+		+	-	<u> </u>
Trinacrium indica	-	-	-	-	+	-				-	<u> </u>	
Trincarium sp.	-	-	-	-	•	-	+	-	-	+	-	
Tripospermum myrti	T -	+_		_	+	-	-	+	-		<u> </u>	+
Triscelophorus acuminatus	-	-	-	-	+	-	-	_	+	+		<u> </u>
Triscelophorus konajensis	+	-	+		+	+	+	-	-	-	+	+
Triscelophorus monosporus	+	+	+	+	+	+	-	+	+	+	+	
Weisneriomyces javanicus	+			-	-		+	-	-	-	-	-
Undetermined sp.1			-	-	-	-	-	-	+		-	+
Undetermined sp.2	-	-	+	-	-	-	-	-	_	-	-	· -
Undetermined sp.3		-	-	+				-		+	-	-
Undetermined sp.4	-	-	+	-	-	-	-	-	-	-	-	+
Undetermined sp.5			-		+.				-	+	-	-
Undetermined sp.6	-	-	-	-	-	-	-	-	-	-	-	-
Total no. of species	23	27	27	20	35	15	18	14	16	29	10	26

(+: presence; -: absence)

S1: Alorna S2: Anmod S4: Chorlem

S7: Cuncolim

S10: Molem S11: Taleigao

S3: Bondla

S5: Cotigao S6: Cudnem S8: Harvalem S9: Kesarval

S12: Tambdisurla

Table. 4.2.7.1: Richness indices:

									·			
NO	23	27	27	20	35	12	18	16	14	29	10	-26
R1	4.27	5.01	4.72	4.11	6.11	2.52	3.51	3.07	2.89	4.53	2.91	4.73
R2	1.98	2.01	1.72	1.98	2.17	1.36	1.59	1.39	1.47	1.32	2.13	2.52

Table.4.2.7.2: <u>Diversity indices</u>:

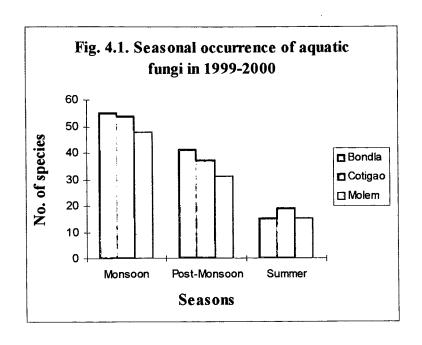
λ	0.13	0.06	0.12	1.10	0.07	0.08	0.63	0.07	0.15	0.29	0.08	0.11
H'	2.44	2.95	2.53	2.55	2.99	2.44	2.75	2.63	2.23	2.06	2.19	2.73
N1	11.4	19.1	12.6	12.9	19.9	11.5	15.7	13.7	9.29	7.87	9.01	15.4
N2	8.0	17.5	8.08	9,93	13.5	13.3	15.8	13.4	6.79	3.46	12.2	9.01

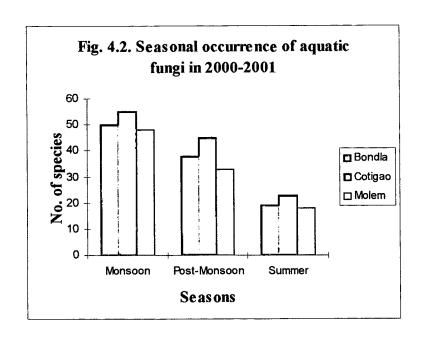
Table.4.2.7.3: Evenness:

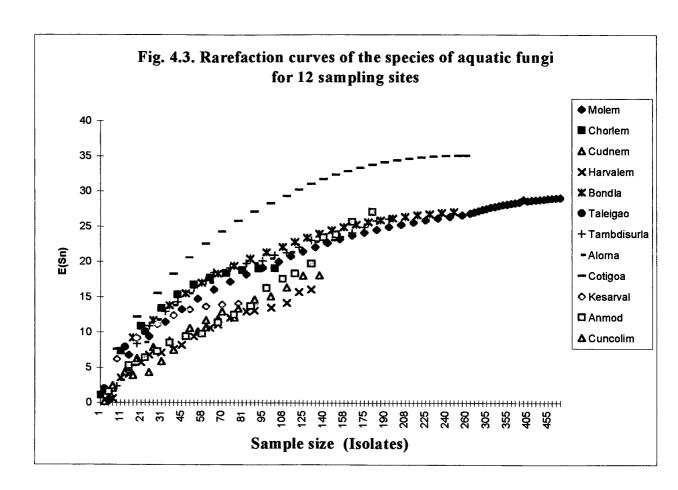
E1	0.78	0.89	0.77	0.85	0.84	0.98	0.95	0.95	0.85	0.61	0.95	0.84
E2	0.49	0.71	0.47	0.64	0.57	0.96	0.87	0.87	0.64	0.27	0.90	0.59
E3	0.48	0.69	0.45	0.62	0.56	0.96	0.86	0.86	0.64	0.25	0.89	0.58
E4	0.69	0.92	0.64	0.77	0.68	1.57	1.01	0.97	0.73	0.44	1.35	0.59
E5	0.66	0.91	0.12	0.72	0.66	1.17	1.01	0.97	0.69	0.36	1.39	0.56

Table.4.2.7.4: Physical and chemical characters of 12 streams:

	Al.	An.	В	Ch.	Cot.	Cud.	Cun.	Har.	Kes.	Mol	Tal	Ta
												m
pН	5.7	5.3	6.3	6.5	6	5.8	6	5	5	6.2	4.5	7
Alkalinity (mg/L)	132.5	60	70	85.6	120	106	105	87	40	83.2	37.5	98
CO ₂ conc. (mg/L)	12.3	10.9	9.2	11.6	7.2	8.2	14.4	16	16.4	8.6	15.4	12.6
Dissolved O ₂ (mg/L)	14.9	14.8	10.5	9.2	15.6	11.3	8.7	8.2	6	12.8	8.47	9.6
Tempera- ture (°C)	25.5	23	24	26	23.2	24	24.4	26	27	23.5	29	26
Na. cont. (mg/L)	23.2	18.9	14.8	12.3	20.3	18.6	26.7	8.4	4.5	15.5	3.2	21
K. cont. (mg/L)	17.4	12.6	13.4	9.8	19.6	20.4	28	12.6	5.2	16	2.9	15.2
Canopy cover (%)	75	55	85	60	100	50	30	10	40	80	30	50







PART III: SURVIVAL MECHANISM ADAPTED BY AQUATIC FUNGI FOR DRY SEASONS

During monsoon, the freshwater fungi grow well and sporulate in abundance on submerged organic matter in streams and rivers. This can be seen by sourcing foam and leaf litter for fungi from running waters. As the water dries up in post-monsoon and summer months how the fungi survive and manage until next rain, was one of the objectives of this investigation.

The ability of and mechanism adapted by aquatic fungi to tide over the dry season every year was studied by setting up a simple *in situ* experiment labelled 'litter bag experiment'. Leaf litter of two riparian plant species, *Hopea ponga* and *Dellinia indica*, collected from adjoining terrestrial habitat, were placed in separate A4 size nylon mesh bags (mesh size # 3 mm) and immersed in a stream at Mollem wildlife sanctuary in Goa in August 2000 and maintained for the subsequent 8 months. One bag each containing submerged leaf litter was lifted up at monthly intervals and analysed in the laboratory for presence of aquatic fungi by placing the litter in glass jars containing distilled water and aerating through a fish-aerator. Foam generated in the jar was analysed for fungi.

The fungi recorded on incubated leaf litter of both plants, at each monthly sampling, are given in Table, 4.3.1 and 4.3.2. In all, 24 species of aquaitc fungi belonging to 21 genera were recovered from the leaf litter of *Dillenia indica* during the 8 month litter-bag study period. (Table, 4.3.1). Of these, *Anguillospora crassa*, *Angulospora aquatica* and *Triscelophorus monosporus* appeared in 6, 5 and 6 times of the monthly isolations respectively.

From the leaf litter of Hopea ponga, 28 species belonging to 25 genera of aquatic

fungi were recovered (Table, 4.3.2). Amongst these, Ardhachandra selenoides, Flagellospora curvula, Lunulospora curvula and Triscelophorus konajensis appeared 7, 6, 5 and 6 times of the monthly sampling respectively. The appearance of remaining species ranged from once to 4 times in both plant litter.

The results indicated that, in terms of species density, the recovery of aquatic fungi was maximum in the post-monsoon months, especially during September, October and November. Generally, the flow of water in the streams of Western Ghat forests recedes in November and in certain parts only patches of water remain. This was reflected in the occurrence of aquatic fungi in litter. In both types of plant litter, maximum number of species was obtained in September and October (Figs. 4.4 and 4.5). In *Dillenia indica* the number of species recorded in September and October were 15 and 16 respectively (Table: 4.3.1) whereas in *Hopea ponga* the fungi documented were 18, 16 and 16 during September, October and November (Table: 4.3.2). The species density ranged from 9 to 6 in the subsequent months (November to April) in *Dillinia indica* and from 13 to 5 in December to April in *Hopea ponga*. The study shows that the species density in *Dellinia indica* is reduced sharply in summer months whereas in *Hopea ponga* it was grdual (Figs. 4.4 and 4.5)

The results further indicated that sizable number of the aquatic fungi were common to both plant litter. Fourteen out of 28 species were common to both plants. Whereas, the following species occurred only on *Dellinia indica*: Cylindrocladium sp.2, *Helicomyces sp.*, *Thozetella sp. Speiropsis pedatospora*, *Trinacrium gracile* and *vermispora sp.* and *Camposporium pellucidum*, *Lunulospora curvula*, *Ingoldiella hamata*, *Triscilosporus konajensis*, *Tripospermum myrti*, *Phalangospira constricta* only on *Hopea ponga*.

The study also pointed out that in the summer months, from February to April, while the litter bags were exposed to dryness due to drying of stream bed, on incubation several of the otherwise terrestrial forms were recovered from leaf litter. In the litter of Dellinia indica, species such as Ardhachandra selenoides, Cylindrocladium sp.1, Dactylella ellipsospora, Pestalotiopsis sp., Sporoschisma uniseptata and Weiseriomyces javanicus were found in the months of January to April (Table: 4.3.1). In Hopea ponga these included Ardhachandra selenoides, Cylindrocladium sp.1, Corynespora sp., Beltrania rhombica, Dactylella ellipsospora, Dictyochaeta assamica and Fusarium sp (Table: 4.3.2).

3.1. AQUATIC FUNGI AS ROOT ENDOPHYTES:

Riparian trees extend their roots into water and some of the aquatic fungi are believed to be taking sanctuary as root endophytes, when the water level in streams goes down or completly dries up. This was studied by examining submerged roots of 7 plant species, namely Alstonia scolaris, Calamus thwaitesii, Dellinia indica, Glaberus sp., Glochidion hohenackeri, Hopea ponga and Saraca asoca in which Dellinia indica and Hopea ponga were the same subjected to 'litter bag experiment'.

The study provided an opportunity to compare and analyse the fate of aquatic fungi during summer months. The fungi such as Cylindrocladium sp.2, Dictyochaeta assamica, Fusarium sp.1, Pestalotiopsis sp, Robillarda sp., Seimatosporium sp., Triscelophorus monosporus recovered from litter bags of Dellinia indica and Anguillospora crassa, Campylospora filicladia, Corynespora sp., Dendrosporium

lobatum, Dictyochaeta assamica, Flagellospora curvula, Robillarda phragmitis, Seimatosporium sp., Triscelophorus monosporus recovered from Hopea ponga were also found as root endophytes of same plants (Table: 4.3.3)

The root endophytes were isolated from primary, secondary and tertiary roots of all the 7 plants species. In the order of abundance, the highest percent colonisation was observed in primary roots (80%), followed by secondary (50%) and tertiary root tissues (20%). The results (Table: 4.3.4) clearly indicate that primary roots are the major hosts or sanctuary for aquatic endophytes.

Together, 96 nonsporulating morphotypes were isolated as root endophytes from the 7 plants. These are recorded in the last row of the Table 4.3.3. Amongst these, maximum numbers were recovered from *Saraca asoca* (24), followed by *Dellinia indica* (23), *Hopea ponga* (18) and *Calamus thwaitesii* (16).

3.2. EFFECT OF MINING REJECTS ON GROWTH AND

SPORULATION OF AQUATIC FUNGI

Excavation and export of Iron and manganese ore are the two major industries in the State of Goa. Mining activities are mostly confined to hinterlands and ghats. Significant amounts of mining reject were washed into the stream and river during rain. The effect of mining on the growth and sporulation rate of aquatic fungi has been studied in the laboratory condition.

Foam and submerged leaf samples were collected from two perennial streams of Cudnem Iron Ore Mines, Goa. Single spore isolation was carried out to recover maximum

number of fungi from foam and incubated leaf litter.

Different quantities mining rejects (5g, 10g and 15 g) were added to glass jars containing leaf litter in sterile distilled water. A continuous jet of air was introduced from a fish aerator. Control was run using natural soil gathered from away from mining site. The floristic analysis for aquatic fungi appeared in each jar was conducted by observing the spora under an inverted microscope.

The results obtained are given in Table 4.3.5 (for mining rejects) and Table 4.3.6. (control). As can be seen, in the control, when the same leaves were subjected for aeration with different concentration of natural soil, the number of fungi recovered were more. In all, 14 species of fungi belonging to 12 genera were observed from the aerated leaflitter in control conditions. When the same leaf litter was aerated with mining rejects, 12 species of fungi were reported from 12 different genera. It was also observed that there was a gradual decrease in the number (from 9 to 5) of species as the concentrations of mining rejects were increased

Table: 4.3.1 Litter bag experiment: Occurrence of fungi on Dillenia indica

Name of fungus	Sept.	Oct.	Nov.	Dec.	Jan.	Feb.	Mar.	Apr.
Anguillospora crassa	+		+	+	+	+		
A. longissima	+	+				+		
Angulospora aquatica	+	+	+	+	+		+	
Ardhachandra selenoides	+	+	+	+		Ī		+
Condylospora spumigina	+				+			I
Cylindrocladium sp.1	+	+	+				+	+
Cylindrocladium sp.2		+			+			+
Dactylella ellipsospora		+			+			
Dictyochaeta assamica	+	+					+	
Flabellospora multiradiata	+	+						
Flagellospora curvula	+	+	+	+	+			
Fusarium sp.		+			+	+		+
Helicomyces roseus	+	+		+	+		+	
Nawawia sp.			+	+	+		+	
Pestalotiopsis sp.1	+				+	+	+	
Robillarda phragmitis	+	+						+
Seimatosporium sp.	+	+						
Speiropsis pedatospora		+	+	+		+		
Sporoschisma uniseptata		+						
Thozetella sp.		+					+	
Trinacrium gracile	+					+		
Triscelophorus monosporus	+	+	+	+		+		+
Vermispora sp.			+			+		
Weiseriomyces javanicus						+	+	
Total	15	16	9	8	9	9	8	6

Table 4.3.2. Litter bag experiment: Occurrence of fungi on *Hopea ponga*:

Name of fungus	Sept.	Oct.	Nov.	Dec.	Jan.	Feb.	Mar.	Apr.
						<u> </u>		
Anguillospora crassa	+	+	<u> </u>			<u> </u>		ļ
Anguillospora longissima	+	+					+	
Angulospora aquatica	+	+	+			+		
Ardhachandra selenoides	+	+	+	+	+	+		+
Beltrania rhombica			+	+		+	<u> </u>	
Bahusutrabeeja dwaya				+				
Campylospora filicladia	+	+	+					
Camposporium pellucidum	+	+	+	+	<u> </u>	1	+	
Chaetendophragmia triseptata				+	+	+		
Cylindrocladium sp.1	+	+	+	+	+			+
Corynespora sp.		+	+					
Dendrosporium lobatum	+		+				+	
Dactylella ellipsospora			+	+				+
Dictyochaeta assamica	+				+		+	
Flabellospora verticillata	+	+						
Flagellospora curvula	+	+	+	+	+	+		
Fusarium sp.					+	+	T	+
Ingoldiella hamata	+	+	+					
Lunulospora curvula	+	+	+	+	+			
Nawawia sp.						+		
Phalangispora constricta						Ĭ .	+	
Robillarda phragmitis	+			+	+			
Sopagraha sibika	+	+						
Seimatosporium sp.			+	+		+		+
Triscelophorus konajensis	+	+	+	+	+	+		
Triscelophorus monosporus	+	+	+		+			
Tripospermum myrti			+					
Tetrachaetum elegans	+	+		+				
Total no. of species	18	16	16	13	9	9	5	5

Table. 4.3.3. LIST OF FUNGI APPEARED AS ROOT ENDOPHYTE

Name of fungus	R1	R2	R3	R4	R5	R6	R7
Anguillospora crassa	+					+	
Beltraniella odinae		+	+				
Campylospora filicladia		+			+	+	
Corynespora sp.	+		+	+		+	
Curvularia lunata				+			+
Cylindrocladium sp.1		+			+		+
Cylindrocladium sp.2	+	+	+	+			
Dactylaria sp.1						+	
Dendrosporium lobatum		+			1	+	
Dictyochaeta assamica		+	+	+		+	+
Dichotomophthoropsis aquatica				+			
Flagellospora curvula	+	+				+	
Fusarium sp.1			+			+	
Fusarium sp.2		+		+			+
Gonatophragmium sp.	T		+				+
Gonytrichum sp.	+						+
Kumbhamaya jalapriya						+	
Lunulospora curvula	+	+		+			
Nectria sp.						+	
Pestalotiopsis sp.1		+	+				+
Ramichloridium sp.			+				
Robillarda sp.	+	+	+		+	+	+
Seimatosporium sp.	+		+			+	+
Triscelophorus monosporus		+	+		+	+	+
Undetermined ascomycete-1	+						
Undetermined ascomycete-2			+				
Undetermined sporodochial		+		+	+	+	
Total no of species	9	13	12	8	5	14	10
Non-sporulating morphotypes	6	16	23	5	4	18	24
Total	15	29	35	13	9	32	34

Table 4.3.4. Total number of endophytes (including nonsporulating types) isolated from different root parts:

Root type	R1	R2	R3	R4	R5	R6	R7
Primary	9	17	23	8	6	20	21
Secondary	4	8	9	5	3	7	7
Tertiary	2	4	3	-	_	5	6
Total	15	29	35	13	9	32	34

R1-Alstonia scolaris

R2-Calamus thwaitesii

R3-Dellinia indica

R4-Glaberus sp.

R5-Glochidion hohenackeri

R6-Hopea ponga

R7-Saraca asoca

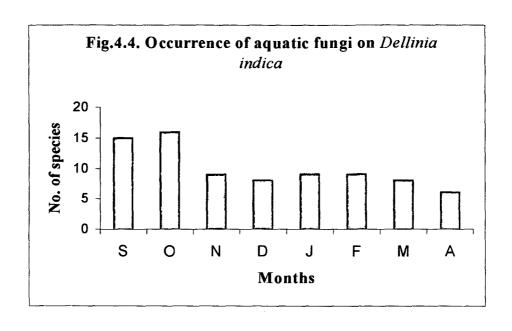
Table 4.3.5. Fungi on submerged leaf with different conc. of mining rejects

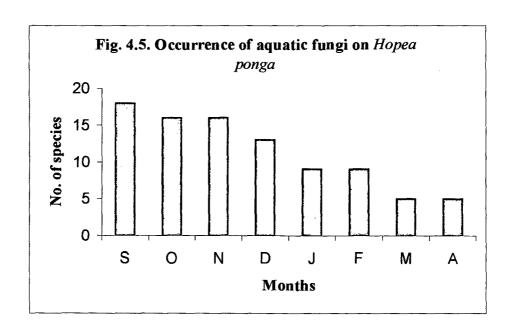
Name of fungus	5g	10g	15g	20g	25g
					_
Anguillospora crassa	+	+	-	-	-
Cylindrocladium sp.1	+	-	+	+	+
Dictyochaeta assamica	+	+	+	+	-
Flagellospora curvula	+	-	+	-	-
Fusarium sp.	+	+	-	+	+
Helicomyces roseus	-	+	+	-	-
Pestalotiopsis sp.	+	+	-	+	+
Robillarda phragmitis	+	-	+	+	+
Speiropsis pedatospora	-	+	-	-	-
Thozetella sp.	+	+	+	+	-
Triscelophorus monosporus	+	+	_	+	-
Weiseriomyces javanicus	-	-	+	-	+
Total no. of species	9	8	7	7	5

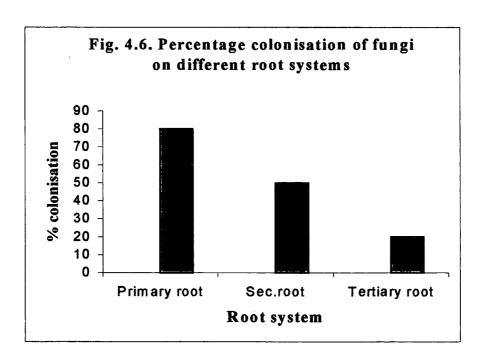
Table 4.3.6. Fungi on submerged leaf with different conc. of control soil.

Name of fungus	5g	10g	15g	20g	25g
Anguillospora crassa	-	+	+	-	+
Anguillospora longissima	+	-	+	+	-
Cylindrocladium sp.1	+	+	+	+	+
Dictyochaeta assamica	+	+	+	+	+
Flabellospora verticillata	+	+	-	-	-
Flagellospora curvula	+	+	+	+	-
Fusarium sp.	+	+	+	+	+
Helicomyces roseus	-	-	+	+	+
Pestalotiopsis sp.	+	-	+	+	+
Robillarda phragmitis	+	-	-	+	+
Speiropsis pedatospora	-	+	+	-	-
Thozutella sp.	+	-	-	+	+
Triscelophorus monosporus	+	+	+	+	+
Weiseriomyces javanicus	+	+	+	-	+
Total no. of species	11	9	11	10	10

pН	Alkal.	CO_2	O_2	Temp.	Na.	K.	Canopy cover
	(mg/L)	(mg/L)	(mg/L)	(°C)	(mg/L)	(mg/L)	(%)
5.8	106	8.2	11.3	24	18.6	20.4	50







PART IV: ABILITY OF AQUATIC FUNGI TO PRODUCE DIFFERENT ENZYMES

Of the nearly 800 isolates of fungi gathered from aquatic system and brought into pure culture during the course of present study, 60 randomly selected morphotypes were subjected to qualitative screening for a few enzymes, namely amylase, cellulase, esterase and pectinase. These are important enzymes of fungal origin having various applications in industry (Dreyfuss and Chapela, 1994; Rossman, 1994; Hawksworth et al. 1995).

The cultures subjected for screening were isolated from foam by single spore isolation technique. Majority were identified down to species level. A few non-sporulating isolates which appeared as root endophytes were also subjected for enzyme study and categorised here as NS-1 to NS-13.

Amongst the 60 isolates tested for production of different enzymes, except one (NS-1), all the others showed varying degrees of enzyme activity. Two isolates, namely *Dactylaria sp.2* and *Triscelophorus monosporus*, exhibited ability to secrete all the enzymes. The results of each of these qualitative assays are given in the Tables: 4.4.2-4.4.5.

Twentysix isolates were positive for amylase (Table 4.4.2). These included: Anguillospora longissima, Ardhachandra selenoides, Ardhachandra sp., Beltrania rhombica, Ceratosporium sp., Dactylaria aquatica, Dactylaria sp., Dendrospora erecta, Dendrosporium lobatum, Dichotomophtoropsis sp., Flabellospora verticillata, Fusarium sp.1, Fusarium sp.2, Helicosporium sp.3, Ingoldiella hamata, Kumbhamaya jalapriya, Robillarda phragmitis, Seimatosporium sp., Speiropsis pedatospora, Tricladium splendens, Triscelophorus monosporus, Tripospermum myrti, Triscelophorus acuminatus

The following 30 isolates showed positiveness for cellulase (Table 4.4.3):

Actinospora megaolospora, Anguillospora crassa, Ardhachandra selenoides,

Articulospora tetracladia, Beltrania rhombica, Camposporium pellucidum, Dactylaria

sp., Dendrosporium lobatum, Dichotomophtoropsis sp., Dictyochaeta assamica,

Diploospora indica, Flabellospora verticillata, Fusarium sp.1, Helicopsorium sp.2,

Ingoldiella hamata, Isthmotricladia laeensis, Lunulospora curvula, Robillarda

phragmitis, Seimatosporium sp., Subulispora sp.1, Tricladium splendens, Triscelophorus

acuminatus, Triscelophorus konajensis, Triscelophorus monosporus, NS-2, NS-4, NS-5,

NS-6, NS-10 and NS-11

The following 32 strains exhibited positive activity of esterase (Table 4.4.4): Alatospora acuminata, Anguillospora crassa, Anguillospora longissima, Ardhachandra Articulospora tetracladia, Beltraniella Sp., Camposporium pellucidum, sp., Campylospora chaetocladia, Ceratosporium sp., Condylosporda spumigena, Dactylaria sp. 1, Dactylaria Sp., Flabellospora multiradiata, Flabellospora verticillata, Flagellospora curvula, Fusarium sp.2, Helicosporium sp.1, Helicopsorium sp.2, Helicosporium sp.3, Isthmotricladia laeensis, Pestalotiopsis sp., Robillarda phragmitis, Trinacrium indica, Tripospermum myrti, Triscelophorus acuminatus, NS-3, NS-4, NS-5, NS-8, NS-10, NS-12 and NS-13

The following 29 morphotypes showed pectinase activity (Table 4.4.5):

Actinospora megaolospora, Anguillospora crassa, Anguillospora longissima,

Ardhachandra selenoides, Ceratosporium sp., Dactylaria sp.1, Dactylaria sp.2,

Dendrospora erecta, Flabellospora multiradiata, Flagellospora curvula, Fusarium sp.1,

Gonytrichum sp., Helicomyces roseus, Helicosporium sp.1, Helicopsorium sp.2,

Helicosporium sp.3, Kumbhamaya jalapriya, Lemonniera aquatica, Lunulospora curvula, Speiropsis hyalospora, Subulispora sp.1, Tricladium splendens, Triscelophorus monosporus, NS-4, NS-5, NS-7, NS-9, NS-10 and NS-13.

Seventeen of the tested isolates were found to be active only for one enzyme. These included Beltraniella odinae, Campylospora chaetocladia, Condylospora spumigena, Dictyochaeta assamica, Dilpoospora indica, Gonytrichum sp., Helicomyces roseus, Lemonniera aquatica, Pestalotiopsis sp., Speiropsis hyalospora, Trinacrium indica, Triscelophorus konajensis, NS-2, NS-3, NS-7, NS-9 and NS-12. Fifteen of the 60 test cultures, showed activity of any 3 enzymes assayed. These included: Anguillospora crassa, A. longissima, Ardhachandra selenoides, Ceratosporium sp. Dactylaria sp. 1, Flabellospora verticilata, Fusarium sp. 1, Helicosporium sp. 2, Helicopsporium sp. 3, Robillarda phragmitis, Tricladium splendens, Triscelophorus acuminatus, NS-4, NS-5 and NS-10.

Among the 60 isolates of fungi tested for enzyme activity, maximum number (32) exhibited positiveness for esterase activity, followed by cellulase (30), pectinase (29) and the minimum for amylase (26). According to the clearance zone, some of the isolates such as *Anguillospora longissima* (1.5 cm), *Dendrosporium lobatum* (1.8 cm) and NS-11 (1.5 cm) were powerful producers of amylase enzyme. *Dichotomophthoropsis aquatica* (1.3 cm), *Isthmotricladia laeensis* (1.4 cm) and NS-6 (1.4 cm) were powerful producers of cellulase. *Beltraniella odinae* (1cm) and *Triscelophorus monosporus* (1.2 cm) were found to be the active producers of esterase enzyme and *Flabellospora multiradiata* (0.8 cm) as producer of measurable quantity of pectinase.

Table: 4.4.1: Qualitative estimation of enzyme activity of fungi obtained from freshwater habitat:

Name of fungus	Amylase	Cellulase	Esterase	Pectinase
1				
Actinospora megaolospora	_	+		+
Alatospora acuminata	 			+
Anguillospora crassa	-	+		+
Anguillospora longissima	+		+	+
Ardhachandra selenoides	+	+		+
Ardhachandra sp.	+		+	
Articulospora tetracladia		+	+	
Beltrania rhombica	+	+		
Beltraniella odinae			+	
Camposporium pellucidum	_	+	+	
Campylospora chaetocladia	·		+	
Ceratosporium sp.	+		+	+
Condylosporda spumigena			+	
Dactylaria aquatica	+	_	+	+
Dactylaria sp.	+	+	+	+
Dendrospora erecta	+			+
Dendrosporium lobatum	+	+		
Dichotomophtoropsis aquatica	+	+		
Dictyochaeta assamica		+		
Diploospora indica		+		
Flabellospora multiradiata			+	+
Flabellospora verticillata	+	+	+	
Flagellospora curvula			+	+
Fusarium sp. l	+	+		+
Fusarium sp.2	+		+	
Gonytrichum sp.	1			+
Helicomyces roseus				+
Helicosporium sp. 1	 		+	+
Helicopsorium sp.2	† -	+	+	+
Helicosporium sp.3	+	·	+	+
Ingoldiella hamata	+	+		
Isthmotricladia laeensis	<u> </u>	+	+	
Kumbhamaya jalapriya	+			+
Lemonniera aquatica	 			+
Lunulospora curvula		+		+
Pestalotiopsis sp.		· ·	+	
Robillarda phragmitis	+	+	+	
Seimatosporium sp.	+	+		

Speiropsis hyalospora		_		+
Speiropsis pedatospora	+			
Subulispora sp. 1	_	+		+
Tricladium splendens	+	+		+
Trinacrium indica	_		+	
Tripospermum myrti	+	_	+	
Triscelophorus acuminatus	+	+	+	
Triscelophorus konajensis	<u>_</u>	+		
Triscelophorus monosporus	+	+	+	+
NS.1		_		
NS.2	_	+		
NS.3	_	_	+	_
NS.4		+	+	+
NS.5	_	+	+	+
NS.6	+	+		
NS.7	_	_	_	+
NS.8	+		+	
NS.9	_	_		+
NS.10		+	+	+
NS.11	+	+		
NS.12			+	
NS.13			+	+
Total of active cultures	26	30	32	29

Table: 4.4.2: AMYLOLYTIC ACTIVITY

Name of fungus	Amylase	Remarks
Actinospora megaolospora		No activity
Alatospora acuminata	_	No activity
Anguillospora crassa	<u>_</u>	No activity
Anguillospora longissima	+	1.5 cm wide clearance zone.
Ardhachandra selenoides	+	0.6 cm wide clearance zone
Ardhachandra sp.	+	1.0 cm wide clearance zone
Articulospora tetracladia		No activity
Beltrania rhombica	+	0.8 cm wide clearance zone
Beltraniella odinae	_	No activity
Camposporium pellucidum		No activity
Campylospora chaetocladia		No activity
Ceratosporium sp.	+	0.3 cm wide clearance zone
Condylosporda spumigena	_	No activity
Dactylaria aquatica	+	Colony bound clearance
Dactylaria sp.	+	0.5 cm wide clearance zone
Dendrospora erecta	+	0.5 cm wide clearance zone
Dendrosporium lobatum	+	1.8 cm wide clearance zone

Dichotomophtoropsis aquatica	+	Colony bound clearance
Dictyochaeta assamica		No activity
Diploospora indica		No activity
Flabellospora multiradiata		No activity
Flabellospora verticillata	+	0.4 cm wide clearance zone
Flagellospora curvula		No activity
Fusarium sp. 1	+	0.3 cm wide clearance zone
Fusarium sp.2	+	0.4 cm wide clearance zone
Gonytrichum sp.		No activity
Helicomyces roseus		No activity
Helicosporium sp. 1		No activity
Helicopsorium sp.2		No activity
Helicosporium sp.3	+	0.7 cm wide clearance zone
Ingoldiella hamata	+	0.2 cm wide clearance zone
Isthmotricladia laeensis		No activity
Kumbhamaya jalapriya	+	0.1 cm wide clearance zone
Lemonniera aquatica		No activity
Lunulospora curvula		No activity
Pestalotiopsis sp.		No activity
Robillarda phragmitis	+	0.4 cm wide clearance zone
Seimatosporium sp.	+	Colony bound clearance
Speiropsis hyalospora		No activity
Speiropsis pedatospora	+	0.3 cm wide clearance zone
Subulispora sp. 1		No activity
Tricladium splendens	+	0.5 cm wide clearance zone
Trinacrium indica	_	No activity
Tripospermum myrti	+	0.7 cm wide clearance zone
Triscelophorus acuminatus	+	0.4 cm wide clearance zone
Triscelophorus konajensis		No activity
Triscelophorus monosporus	+	0.5 cm wide clearance zone
NS-1	_	No activity
NS-2		No activity
NS-3		No activity
NS-4		No activity
NS-5		No activity
NS-6	+	0.3 cm wide clearance zone
NS-7		No activity
NS-8	+	1.3 cm wide clearance zone
NS-9		No activity
NS-10		No activity
NS-11	+	1.5 cm wide clearance zone
NS-12		No activity
NS-13		No activity

Table: 4.4.3: <u>CELLULOLYTIC ACTIVITY</u>

Name of fungus	Cellulase	Remarks
Actinospora megaolospora	+	0.3 cm wide clearance zone
Alatospora acuminata		No activity
Anguillospora crassa	+	0.2 cm wide clearance zone
Anguillospora longissima		No activity
Ardhachandra selenoides	+	0.6 cm wide clearance zone
Ardhachandra sp.		No activity
Articulospora tetracladia	+	1.1 cm wide clearance zone
Beltrania rhombica	+	0.8 cm wide clearance zone
Beltraniella odinae		No activity
Camposporium pellucidum	+	0.6 cm wide clearance zone
Campylospora chaetocladia		No activity
Ceratosporium sp.		No activity
Condylosporda spumigena		No activity
Dactylaria aquatica		No activity
Dactylaria sp.	+	0.9 cm wide clearance zone
Dendrospora erecta		No activity
Dendrosporium lobatum	+	0.7 cm wide clearance zone
Dichotomophtoropsis aquatica	+	1.3 cm wide clearance zone
Dictyochaeta assamica	+	0.4 cm wide clearance zone
Diploospora indica	+	0.9 cm wide clearance zone
Flabellospora multiradiata		No activity
Flabellospora verticillata	+	0.6 cm wide clearance zone
Flagellospora curvula		No activity
Fusarium sp. 1	+	0.7 cm wide clearance zone
Fusarium sp.2		No activity
Gonytrichum sp.		No activity
Helicomyces roseus		No activity
Helicosporium sp. 1		No activity
Helicopsorium sp.2	+	0.3 cm wide clearance zone
Helicosporium sp.3		No activity
Ingoldiella hamata	+	1.1 cm wide clearance zone
Isthmotricladia laeensis	+	1.4 cm wide clearance zone
Kumbhamaya jalapriya		No activity
Lemonniera aquatica		No activity
Lunulospora curvula	+	0.4 cm wide clearance zone
Pestalotiopsis sp.		No activity
Robillarda phragmitis	+	0.2 cm wide clearance zone
Seimatosporium sp.	+	0.3 cm wide clearance zone
Speiropsis hyalospora		No activity

Speiropsis pedatospora		No activity
Subulispora sp. 1	+	0.7 cm wide clearance zone
Tricladium splendens	+	0.1 cm wide clearance zone
Trinacrium indica		No activity
Tripospermum myrti		No activity
Triscelophorus acuminatus	. +	0.6 cm wide clearance zone
Triscelophorus konajensis	+	1.0 cm wide clearance zone
Triscelophorus monosporus	+	0.8 cm wide clearance zone
NS-1		No activity
NS-2	+	0.4 cm wide clearance zone
NS-3		No activity
NS-4	+	0.9 cm wide clearance zone
NS-5	+	0.3 cm wide clearance zone
NS-6	+	1.4 cm wide clearance zone
NS-7		No activity
NS-8		No activity
NS-9		No activity
NS-10	+	0.2 cm wide clearance zone
NS-11	+	0.5 cm wide clearance zone
NS-12		No activity
NS-13		No activity

Table: 4.4.4: ESTERASE ACTIVITY

Name of fungus	Esterase	Remarks
Actinospora megaolospora		No activity
Alatospora acuminata	+	Media turns dark purple
Anguillospora crassa	+	Media turns dark purple, crystals present
Anguillospora longissima	+	Media turns dark purple, 0.5 cm zone crystals
Ardhachandra selenoides	_	No activity
Ardhachandra sp.	+	Media turns dark purple, 0.3 cm zone crystals
Articulospora tetracladia	+	Media turns dark purple, 0.3 cm zone crystals
Beltrania rhombica	_	No activity
Beltraniella odinae	+	Media turns dark purple, 1cm zone crystals
Camposporium pellucidum	+	Media turns dark purple, 0.5 cm zone cystals
Campylospora chaetocladia	+	Media turns dark purple.
Ceratosporium sp.	+	Media turns dark purple.
Condylosporda spumigena	+	Media turns dark purple, crystals present.
Dactylaria aquatica	+	Media turns dark purple.
Dactylaria sp.	+	Media turns dark purple,0.6 cm zone crystals.
Dendrospora erecta		No activity
Dendrosporium lobatum	_	No activity
Dichotomophtoropsis sp.	_	No activity

Dictyochaeta assamica	-	No activity
Diploospora indica	· · · · · · · · · · · · · · · · · · ·	No activity
Flabellospora multiradiata	+	Media turns dark purple.
Flabellospora verticillata	+	Media turns dark purple, 0.5 cm zone.
Flagellospora curvula	+	Media turns dark purple.
Fusarium sp. 1		No activity
Fusarium sp.2	+	Media turns dark purple, 0.3 cm zone.
Gonytrichum sp.		No activity
Helicomyces roseus		No activity
Helicosporium sp. 1	+	Media turns dark purple.0.4 cm zone.
Helicopsorium sp.2	+	Media turns dark purple, 0.8 cm zone.
Helicosporium sp.3	+	Media turns dark purple.
Ingoldiella hamata		No activity
Isthmotricladia laeensis	. +	Media turns dark purple.
Kumbhamaya jalapriya		No activity
Lemonniera aquatica		No activity
Lunulospora curvula		No activity
Pestalotiopsis sp.	+	Media turns dark purple.
Robillarda phragmitis	+	Media turns dark purple, 0.2 cm zone.
Seimatosporium sp.		No activity
Speiropsis hyalospora	_	No activity
Speiropsis pedatospora		No activity
Subulispora sp. 1		No activity
Tricladium splendens		No activity
Trinacrium indica	+	Media turns dark purple, 0.3 cm zone.
Tripospermum myrti	+	Media turns dark purple, 0.3 cm zone crystals.
Triscelophorus acuminatus	+	Media turns dark purple.
Triscelophorus konajensis		No activity
Triscelophorus monosporus	+	Media turns dark purple, 1.2 cm zone crystals.
NS-1		No activity
NS-2		No activity
NS-3	+	Media turns dark purple.
NS-4	+	Media turns dark purple, 0.4 cm zone.
NS-5	+	Media turns dark purple, 0.5 cm zone.
NS-6		No activity
NS-7	····	No activity
NS-8	+	Media turns dark purple.
NS-9		No activity
NS-10	+	Media turns dark purple, 0.2 cm zone crystals.
NS-11		No activity
NS-12	+	Media turns dark purple.
NS-13	+	Media turns dark purple.

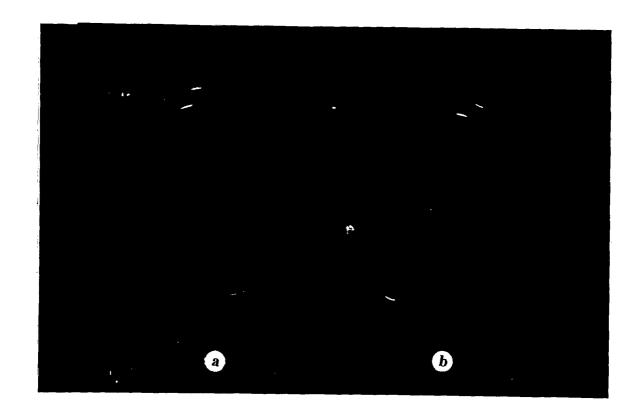
Table: 4.4.5: <u>PECTINOLYTIC ACTIVITY</u>

Name of fungus	Activity	Remarks
Actinospora megaolospora	+	Colony bound clearance
Alatospora acuminata	_	No activity
Anguillospora crassa	+	0.2 cm wide clearance zone
Anguillospora longissima	+	0.3 cm wide clearance zone
Ardhachandra selenoides	+	Colony bound clearance
Ardhachandra sp.		No activity
Articulospora tetracladia		No activity
Beltrania rhombica	_	No activity
Beltraniella odinae		No activity
Camposporium pellucidum	_	No activity
Campylospora chaetocladia		No activity
Ceratosporium sp.	+	0.5 cm wide clearance zone
Condylosporda spumigena		No activity
Dactylaria aquatica	+	0.3 cm wide clearance zone
Dactylaria sp.	+	0.4 cm wide clearance zone
Dendrospora erecta	+	0.3 cm wide clearance zone
Dendrosporium lobatum		No activity
Dichotomophtoropsis sp.		No activity
Dictyochaeta assamica		No activity
Diploospora indica		No activity
Flabellospora multiradiata	+	0.8 cm wide clearance zone
Flabellospora verticillata		No activity
Flagellospora curvula	+	Membrane bound pectin
Fusarium sp. 1	+	0.3 cm wide clearance zone
Fusarium sp.2		No activity
Gonytrichum sp.	+	Membrane bound pectin
Helicomyces roseus	+	0.3 cm wide clearance zone
Helicosporium sp.1	+	0.4 cm wide clearance zone
Helicopsorium sp.2	+	0.3 cm wide clearance zone
Helicosporium sp.3	+	0.5 cm wide clearance zone
Ingoldiella hamata	_	No activity
Isthmotricladia laeensis		No activity
Kumbhamaya jalapriya	+	0.2 cm wide clearance zone
Lemonniera aquatica	+	0.6 cm wide clearance zone
Lunulospora curvula	+	0.5 cm wide clearance zone
Pestalotiopsis sp.		No activity
Robillarda phragmitis		No activity
Seimatosporium sp.		No activity
Speiropsis hyalospora	+	0.4 cm wide clearance zone

Speiropsis pedatospora		No activity
Subulispora sp. 1	+	0.4 cm wide clearance zone
Tricladium splendens	+	0.3 cm wide clearance zone
Trinacrium indica		No activity
Tripospermum myrti	_	No activity
Triscelophorus acuminatus	_	No activity
Triscelophorus konajensis		No activity
Triscelophorus monosporus	+	0.5 cm wide clearance zone
NS-1		No activity
NS-2		No activity
NS-3		No activity
NS-4	+	0.5 cm wide clearance zone
NS-5	+	0.2 cm wide clearance zone
NS-6		No activity
NS-7	+	0.2 cm wide clearance zone
NS-8		No activity
NS-9	+	0.3 cm wide clearance zone
NS-10	+	0.2 cm wide clearance zone
NS-11		No activity
NS-12		No activity
NS-13	+	0.5 cm wide clearance zone

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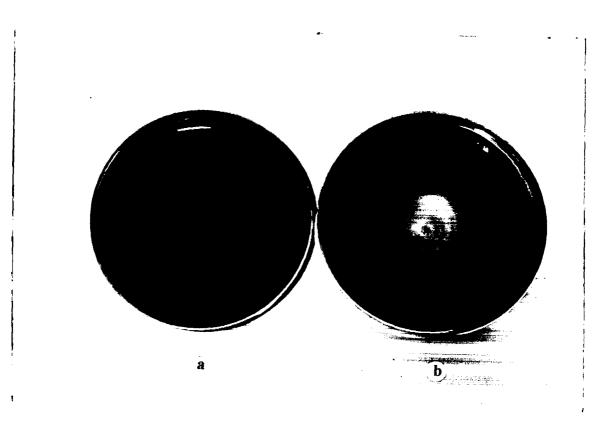
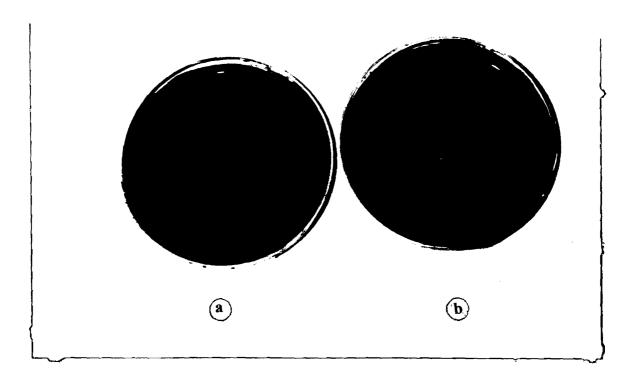


Plate. 13

<u>Plate: 14</u>

- 1. Cellulase assay a) control plate
 - b) test culture with clearance zone.
- 2. Esterase assay
- a) control plate
- b) media turned to dark purple.



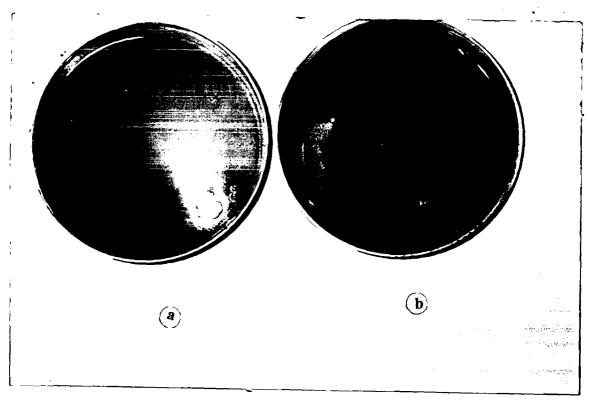


Plate. 14

DISCUSSION

The results presented in this thesis are based on an investigation carried out over a period of two and a half years, from January 1999 to May 2001, on the diversity, ecology and activity of aquatic fungi of the streams and rivers of Western Ghats forests in Goa State.

Seasonal occurrence and abundance of fungi, were studied by sampling aquatic fungi at regular intervals for two years during monsoon, pre-monsoon and post-monsoon from the streams of Bondla, Molem and Cotigao wildlife sanctuaries in the State. All the streams had dense riparian canopy. Studies on diversity and floristics were carried out based on samples randomly gathered from several more streams and rivers in Goa during monsoon and post-monsoon.

Floating foam (FF), submerged leaf-litter (SL) and submerged roots (SR) constituted the source of aquatic fungi dealt in this study. In all, 135 species of fungi belonging to 113 genera were recovered. Taxonomically, they belonged to Hyphomycetes (122), Coelomycetes (6) and Ascomycetes (7). The descriptions along with illustrations of 102 aquatic conidial fungi belonging to 80 genera are given in detail in Part I of this Chapter. The ascomycetous fungi recovered are described but only listed out (Tables 4.1.2 and 4.1.3).

Standard mycological diagnostic characters were used while describing the aquatic fungi. Herbarium specimens of the fungi described were deposited at the Herbarium of Goa University Botany Department (GUBH). Pure cultures of fungi recovered in this study were housed at the Goa University Fungus Culture Collection Unit (GUFCC), a

regional facility for fungi, available in the Department of Botany. International Code for Botanical Nomenclature(ICBN) has been followed in describing known and new fungi (Hawksworth, 1974).

1. Taxonomic diversity of aquatic fungi:

Of the total 135 species of microfungi which included Hyphomycetes (122), Coelomycetes (6) and Ascomycetes (7), recovered from foam, submerged leaf or submerged roots of freshwater streams and rivers of Goa, 102 taxa of conidial fungi are described in detail in this thesis. Those fungi regularly encountered and sporulated well when the leaf substrate or agar blocks containing mycelia immersed in sterile water and aerated continuously were recognised as 'strict aquatics'. The isolates occasionally encountered and not sporuated well under water but readily produced spores above water were treated as 'casual aquatics'. In a true sense, the latter are terrestrial forms but found their way to water by accident. By this new catagorization, a total of 63 taxa of strict aquatics and 61 species of causal aquatics were documented in this effort.

Terrestrial fungi recovered in culture from submerged litter and surface foam during the course of this study, the casual aquatics, were those either washed along with flood water from nearby terrestrial habitats or those got into the stream through leaves shed from riparian trees. Reproductive propagules such as conidia or ascospores of these fungi did not exhibit any special aquatic adaptive features such as branching, coiling or bearing appendages of varied types, as seen in strict aquatics.

Hitherto recognised terrestrial leaf litter fungi such as *Beltrania 'rhombica*, *Beltraniella odinae*, *Ardhachandra selenoides*, *Diploospora indica* etc. however produce conidia of interesting morphology. The spores are dorsiventrally flat and narrower at both ends and therefore may be considered ideal for aquatic floatation. Similarly, *Tetraploa aristata*, *Dictyochaeta assamica* and *Thozetella sp.* have distinct appendages which may be considered as an adaptation for accidental water inhabitation. It is therfore prudent to consider most of those causal aquatics occasionally finding their way to water as 'possible aquatics'. Further, in tropical moist deciduous forests such as the wildlife sanctuaries of Goa, a clear demarcation as 'aquatic' and 'terrestrial' amongst fungi will not be possible.

The survey carried out by Sridhar et al. (1992) indicated that 62 species of aquatic fungi are so far known from the streams of Western Ghats. Another 3 species were added from earlier work of this Department (Table 2.1.) Through the present study, although those reported earlier are not recovered fully, an additional of 21 species belonging to 19 genera were added to the aquatic fungal flora of Western Ghats. The following 21 species of strict aquatic fungi are new records from the forests of Western Ghats: Anguillospora furtiva, Articulospora sp., Bahusutrabeeja angularis, Camposporium pellucidum, Centrospora acerina, Dactylaria aquatica sp. nov., Dendrospora yessemreddia sp. nov., Dendrosporium lobatum, Dichotomophthoropsis aquatica sp. nov., Diplocladiella scalaroides, Diploospora indica sp. nov., Helicomyces roseus, Helicosporium sp.1-3, Isthmotricladia britanica, Kumbhamaya jalapriya sp. nov., Mycoleptodiscus indicus, Paratozetella aquatica gen. et sp. nov., Scutisporus brunneus and Trinacrium indica sp. nov.

The major contribution of this study is the discovery of seven new species and a ew genus of strict aquatics from the freshwaters of streams of Goa. These included Parathozetella Sreekala et Bhat gen. nov. (from foam), Dactylaria aquatica Sreekala et Bhat sp. nov. (a root endophyte), Dendrospora yessemreddia Sreekala et Bhat sp. nov. (a root endophyte), Diploospora indica Sreekala et Bhat sp. nov. (from foam), Kumbhamaya alapriya Sreekala et Bhat sp. nov. (a root endophyte), Parathozetella aquatica Sreekala et Bhat sp. nov. (from foam) and Trinacrium indica Soosamma, Lekha, Greekala et Bhat sp. nov. (from foam).

The study proved that the forests of Western Ghats in Goa region are very rich in heir aquatic fungal flora. The study also proved that serious and dedicated floristic effort such as this, if one intensively carries out, would always result with discovery of novel axa. Undoubtedly, isolation of seven new fungi from a small region such as Goa is a big and rewarding contribution to mycology.

During the course of this study, a few ascomycetes and many coelomycetes were ecovered. They are tentatively identified yet, undoubtedly add to the fungal wealth of our country. A sizable number of nonsporulating isolates (96) were recovered, especially as endophytes and they fell into 42 distinct morphotypes based on cultural characters. In the absence of any sporulation, their identity presently remained unknown. It will be possible to diagnose them to species level with certainty as and when they produce spores and spore-bearing structures. With increasing realization that fungi are sources of powerful metabolites of varied usage in agriculture, biotechnology, chemical, food and

pharmaceutical industries, the importance of this work and utility of these isolates cannot be underestimated. They are important additions to the fungal wealth of our nation.

Another landmark contribution of this effort is building up of a collection of pure cultures of freshwater fungi of the Western Ghats' forest streams, recovered through single spore isolation. Although other institutes and workers along the West Coast have worked on this group of fungi since long, no one has attempted to isolate, document and conserve the fungi in a repository or culture collection. The culture collection has been done with all sincerity during the course of this work. The fungi recovered, a total of 135 taxa (strict aquatics: 63; casual aquatics: 61; ascomycetes: 7; coelomycetes: 6; undetermined taxa: 10; nonsporulating forms: 96) are carefully maintained in MEA maintenance medium under mineral oil. Details of each taxon is documented in a specially constructed 'data-sheetwith information on taxonomy, habitat affinity and activity. This is the first time such as composite effort on aquatic fungi has been done.

Interesting points have emerged out from this study. Majority of aquatic fungi are now known to be nonphialidic, moniliaceous and with spores bearing adaptive features for floatation. This observation makes clear that freshwater fungi are a unique group of their own. Future studies in the future on molecular features of these fungi will prove if they have any more commonness in their biology.

A dichotomous taxonomic key has been constructed for fungi isolated from freshwater system and categorized as 'strict aquatics'. While preparing the key, it was observed that the species of isolated, mostly hyphomycetes, fell into two distinct morphotypes based on conidiogenisis - blastic and phialidic. Further details are in the key (Table 4.1.4).

Table 4.1.4: KEY TO GENERA AND SPECIES OF AQUATIC FUNGI RECORDED FROM GOA

I. Blastic type conidium:

1.	Each consists of a central large spherical body from which has straight divergent arms	Actinospora A. megalospora
2.	Conidia long, septate and curved with curvature more than at one plane	Anguillospora A. crassa A. furtiva A. longissima
3.	Tetraradiate with a stalk at the end of which there are 3-5 fat divergent arms	Articulospora
	a. Conidia with a stalk and 3 divergent arms.b. Conidia with a stalk and 2 divergent arms.	A. tetracladia Articulopsora sp.
	Conidia with proximal half triangular, distal half allantoid with 4 end cells from which appendages arise	Campylospra C. chaetocladia C. filicladia
5.	Conidia elongate, septate body, truncate at liberating end with a tail like projection	Centrospora C. acerina
6.	Conidia elongate, septate, cylindrical with a bend at then centre	Condylospora
	a. Conidia recurved at the mid-region.	C. spumigena
7.	a. Main axis erect, with 4-5 lateral branches	-
	which in turn shows branching.b. Main axis flexible.	D. erecta D. yessemredia
8. (Conidia three lobes on each side with a pedicillate stalk	Dendrosporium D. lobatum

9. (Conidio	ogenous cell dichotomoustly branched	Dichotomophtoro
	a.	Conidia large, multiseptate, worm like	
10.	The	short stalk ending in a small spherical, from	
·		which long straight arms diverge	Flabellospora
	a.	The stalk of the conidium is short, 3-4	r
		slightly fatter divergent arms.	F. crassa
	b.	Stalk small, with 25-32 rediating arms from	
	-,	the central bulbous tip of the stalk.	F. multiradiata
	c.	The stalk of the conidium is longer and	
	•	there are about seven arms.	F. verticillata
11.	Conid	lia spirally helicoid	Helicosporium
	a.	Conidia arising from a bulbous	•
		polyblastic conidiogenous cell.	Helicosporium sp.1
	b.	Conidiogenous cell monoblastic,	•
		conidia 15-20 µm in diam.	Helicosporium sp.2
	c.	Conidiogenous cell monoblastic,	•
		conidia very long.	Helicosporium sp.3
12.	Con	idium consists of thick stalk, at the end of	
		which there are three or more fatter	
		divergent arms	Isthmotricladia
	a.	Conidia has two arm initials, which	
		have shown dichotomy.	I. britanica
	b.	Conidium has 3 divergent arms.	I. laeensis
13. C	onidia	with clamp connections	Ingoldiella
	a.	Conidia with a main axis and 2-3 lateral	
		branches, all ends in a hooked tip.	I. hamata
14.		a with a central oval cell, from which	
		other cells arise laterally, each of which	r
	_	s out as a long pointed septa	Lateriramulosa
	a.	Water-borne conidial fungus, with 3	<i>r</i>
		pointed tip.	L. uniinflata
15. C		unicellular, crescent shaped to sigmoid with	
		achment scar and liberated by breakdown of a	Lamulagnara
•		separating cell	Lunuiospora L. curvula
a.	Cresc	ent shaped conidia.	L. Curvula
6. The		um consists of one or two basal cells and ivergent branches	Spaironsis
	5-7 U	evergent orangenes	pen opsis

16.

	a. Conidia hyaline with mainly 3 branches.b. Conidia pale brown, with 5-7 branches.	S. hyalospora S. pedatospora
	Comula pare of over, where it examples	2. F : 22F : .
	17. Conidium terminal, liberated by the breakdown	
	of a small separating cell	
	a. Tetraradiate, with very long branches.	T. elegans
	18. Conidia with a main axis with 2-4 lateral branches a. Lateral branch uniform in width, not	Tricladium
	tapering. b. Lateral branch 3-4 μm wide at the base,	T. gracile
	tapering to 1.5-2 µm near apex.	T. splendens
	19. Conidia multi-septate, 'Y' shaped	Trinarcium
	nature.	T. indica
	20. Tetraradiate, main axis in line with conidiophore	Triscelophorus
	a. Tetrarariate, arms septate.	T. acuminatus
	b. Tetraradiate, aseptate, smaller sized arms.c. Tetraradiate, aseptate, four arms arising	T. konajensis
	in succession.	T. monosporus
II	Phialidic conidium:	
	Conidium tetraradiate, consisting of a main axis and two laterals	Alatospora
	a. Main axis forming two arms with two	
	lateral arms.	A. acuminata
	2. Conidia slightly circular with filiform appendages	
	Conidia slightly circular with filiform appendages arising from its surface	Nawawia
	2. Conidia slightly circular with filiform appendages	
	Conidia slightly circular with filiform appendages arising from its surface	Nawawia N. filiformis
	Conidia slightly circular with filiform appendages arising from its surface	Nawawia N. filiformis
III.	Conidia slightly circular with filiform appendages arising from its surface	Nawawia N. filiformis Flagellospora
III.	 Conidia slightly circular with filiform appendages arising from its surface	Nawawia N. filiformis Flagellospora F. curvula
III.	 Conidia slightly circular with filiform appendages arising from its surface	Nawawia N. filiformisFlagellospora F. curvulaCamposporium
III.	 Conidia slightly circular with filiform appendages arising from its surface	Nawawia N. filiformisFlagellospora F. curvulaCamposporium
III.	 Conidia slightly circular with filiform appendages arising from its surface	Nawawia N. filiformisFlagellospora F. curvulaCamposporium C. pellucidum

	Conidia two armed, mid brown a. Two horned, brown conidia.	Diplocladiella D. scalaroides
	4. Conidia dark-brown, four-celled, with 4 flexible appendages arising from each cell a. Four equal sized cells from which 4	Scutisporus
	appendages arise.	S. brunneus
	5. Conidia dark-brown, four-celled, with 4 stiff setae arising from the four cells	Tetranloa
	a. Thick-walled conidium with four stiff, straight appendages.	T. aristata
	6. Condia, pyriform with an ellipsoidal stalk cell	Tripospermum T. myrti
IV.	Sporodochial or pycnidial:	
	Sporodochial, conidia cylindrical, unbranched a. Cylindrical conidia joined to form a conidial chain.	Weisneriomyces W. javanicus
		ů.
	 Sporodochial, conidia cylindrical, branched a. Conidia 'Y' shaped, lateral branch 	Phalangispora
	arising from the third basal cell.	P.constricta
	Pycnidial, conidia oblong and cylindrical a. Conidia 1-septate, with 2-3	Robillarda
	appendages at its apical region.	R. phragmitis
	4. Pycnidial, conidia cylindrical, oval a. Conidia hyaline, elliptical, with	Seimatosporium
	two filiform appendages on either ends.	Seimatosporium sp.

2. Seasonal appearance and abundance of aquatic fungi

The results showed that there is no much difference amongst the 3 sites considered for seasonal study in their fungal wealth. This is understandable because all three sites had dense riparian canopy and good flow of water in the streams during monsoon and post monsoon. Of the fungi recovered, 75.55% were foam-trapped, 56.29% associated with submerged leaf litter and 23.70% with roots. This clearly showed that aquatic foam from natural streams will continue to be the best source of diverse fungi, as described by Descals (1997).

It may be seen from Fig. 4.7 that, fungi with blastic type of conidiogenisis were of higher percentage (89.65) than phialidic (10.35) type. It may be said that from the evolutionary stand point (Hawksworth, 1995), the fungi of aquatic ecosystem need not possess phialidic conidiogenesis. This is an instrument largely used for conidiogenesis by fungi in terrestrial environment

The study also revealed (Fig. 4.8) that aquatic spora of branched and appendaged type were of higher percentage (29.31), followed by tetraradiate (24.14), sigmoid (15.52) and helicoid (1.72). These are adaptations for aquatic environment where the spora can remain afloat and get disseminated to a much larger distance. Conidial appendages in aquatic hyphomycetes have been a matter of study for long time (Ingold, 1975; Dix and Webster, 1995). It has been realised that fungi that are taxonomically unrelated while converging into water during the course of evolution exhibited similar morphology as an adaptation to aquatic system. As can be seen from the study, there is no such significance

in the abundance of different types of conidia. All the three to four types of conidia, appendaged, branched, sigmoid and tetraradiate were found in abundance in aquatic systems.

From Fig. 4.9 it can be deduced that moniliaceous fungi (87.93) were more than the pigmented type (12.07) in aquatic systems. In aquatic system one should realize that fungi do not require any pigmentation as a protective measure. In other words, dematiaceous fungi which are in abundance in terrestrial habitat, found in low density in water.

The study revealed that (Fig. 4.10) natural foam accumulated on the surface of water was the best source of aquatic fungi (75.55%) for isolation. Howver, when aerated, significantly high percentage of aquatic fungi was recovered from submerged leaves (56.29%). This justified our taking of submerged leaves as a substrate to evaluate the aquatic flora in the stream ecosystem. Twigs with a very low percentage (8.8%) occurrence of aquatic fungi proved to be a poor substrate for isolation of aquatic fungi.

From the investigation it is clear that (i) monsoon and post monsoon seasons are the best for recovery of freshwater fungi from the streams of forests of Western Ghats. (ii) Those streams with dense riparian tree canopy and abundant substrate availability would yield higher diversity of aquatic fungi. (iii) The physico-chemical factors of stream water have very little impact on the density and abundance of fungi.

3. Survival mechanism adapted by aquatic fungi for dry seasons

The comparitive investigation of microfungi recovered from litter bag experiment ogether with the aquatic fungi isolated as root endophytes provided an opportunity to compare and analyse the fate of aquatic fungi during summer months. The fungi such as Cylindrocladium sp.2, Dictyochaeta assamica, Fusarium sp.1, Pestalotiopsis sp, Robillarda sp., Seimatosporium sp. and Triscelophorus monosporus were recovered from substrate maintained in litter bags of Dellinia indica and Anguillospora crassa, Campylospora filicladia, Corynespora sp., Dendrosporium lobatum, Dictyochaeta assamica, Flagellospora curvula, Robillarda phragmitis., Seimatosporium sp. and Triscelophorus monosporus were recovered from Hopea ponga. Interestingly, all these fungi were also found as root endophytes of same plants. This clearly demonstrated that luring dry months, freshwater fungi largely take refuge in riparian tree root tissues.

Substrate availability for fungi to survive in the stream ecosystem, as the water lries up, is a chance occurrence. Therefore fungi probably look for alteratives and roots of iparian trees provide the much needed shelter. The results clearly indicated that primary oots are the major hosts or sanctuary for aquatic endophytes. Presence of more ascomycetes as root endophytes is also noteworthy.

4. Effect of mining rejects on aquatic fungi

Iron and manganese ore mining are the two major industries in the State of Goa.

Notable amount of rejects were washed into the stream and river during rain. The effect of mining on the growth and sporulation rate of aquatic fungi thus became an interest during this study.

Studies carried out by Sridhar & Kaveriappa (1986, 1989) showed that aquatic fungi are sensitive to herbicides, insecticides and fungicides. Sridhar et al. (2000) demonstrated that copper mining rejects have some impact on aquatic fungi. Although the results of the study carried out in this thesis indicated that mining rejects had some impact on the density and diversity of aquatic fungi, it was truly negligible. Therefore, at this stage, it will not be possible to use aquatic fungi as an indicator of mining pollution of water.

5. Ability of aquatic fungi to produce different enzymes

Of the tests conducted for enzyme activity of 60 isolates of fungi, maximum number exhibited positiveness for esterase activity, followed by cellulase, pectinase and amylase in the decreasing order. Some of the isolates such as *Anguillospora longissima*, *Dendrosporium lobatum* and NS-11 were powerful producers of amylase enzyme. *Dichotomophthoropsis aquatica*, *Isthmotricladia laeensis* and NS-6 were powerful

producers of cellulase. *Beltraniella* sp. and *Triscelophorus monosporus* were found to be the active producers of esterase enzyme and *Flabellospora multiradiata* as producer of measurable quantity of pectinase.

Tween 80 being the important indicator for esterase activity, the fungal cultures active with esterase enzyme can be used as a model for understanding the environmental pollution of Western Ghat streams (Dix and Webster, 1995). Degradation of cellulose and pectin of submerged leaf tissues gets enhanced by the cellulases and pectinases released by the fungi inhabiting leaf litter. A large number of isolates showing positive activity for these enzymes clearly supports the view that aquatic fungi are the major players in the functioning of freshwater ecosystem.

All these assay results indicate that fungi are armoured with a variety of enzymes aid them in their saprophytic mode of activities. Isolation of fungi in culture alone will not give any indication of their activities. The fungi will have to be tested for production of enzymes or any other activity as done in this work. Only on assaying individual plants and associated fungi, one can build a profile of their enzyme machinery. Although enzymes of fungi are for their own growth and sustenance, these can be used for our advantage. Most of the industrially produced or used enzymes are of fungal origin (Hawksworth et al., 1995)

Building up an 'enzyme profile catalogue' along with 'collection of pure cultures' and setting up of a 'taxonomy and biology database' are all positive indicators of an effort of documentation, conservation and utilization of fungal biodiversity of our country. Study such as this therefore will have a far-reaching positive implication in our efforts of building up of microbial biotechnology resource bank in India.

Epilogue

Investigations on freshwater fungi of the streams and rivers of forests of Western ihat in Karnataka State have been carried out by several students of mycology in the last 5 years. Their studies included elucidation of taxonomic diversity, seasonal abundance, npact of physico-chemical character of water on the occurrence of fungi and so on Sridhar et al., 1992). Although the forests of Goa have much in common in their stream cosystem with any other streams of other States along the Western Ghats, it was felt that renewed composite and comprehensive investigation on the diversity, ecology and ctivity of aquatic fungi of the streams of Goa would be worthwhile from the point of levelopment of mycology in this region of the country.

This thesis embodies results of such a study. This modest effort is resulted with locumentation of a large number of microfungi from freshwaters of Goa, elucidation of cological intricacies of the fungi in streams ecosystem and understanding of the enzyme activities of some of these fungi. The effort has also resulted with pure cultures of a number of aquatic fungi. The cultures are deposited and maintained in a state-of-the-art fungus culture collection' repository in Goa University, a unique facility in the country.

It is to be realised that if maximum recovery of fungi has to be achieved from a lifficult or extreme habitat such as the stream system, the method of sampling, single spore isolation technique, maintanence of cultures and above all, scientific documentation of the fungi, should be most efficient and ingenious. This was attempted with all sincerity n this work.

The streams and rivers of Goa during monsoons, were with rapid flow of water, abundant submerged litter and at intervals with enough floating and accumulated foam. The foam and litter gathered were brought to the laboratory and from which single spore isolation of aquatic fungi was done. This is a difficult process but resulted with pure cultures of a large number of wonderful aquatic fungi. This is to be realised that, for the first time in any part of our country, a large number of aquatic fungi were brought into pure culture. It is not only the strict aquatics, but also casuals were brought into culture. A comprehensive database has been prepared for all these fungi. Such documented information and culture facilities of aquatic fungi are very invaluable, especially for future biotechnology in our country.

Almost all the fungi isolated are identified down to the species level. Besides, the effort made on elucidating the ecology of these fungi has given a clue that (i) the best season of isolation of aquatic fungi could be the monsoon and/or early post-monsoon, where one could easily isolate spora of aquatic fungi from floating foam. (ii) we now know that in the off seasons, majority of these fungi take shelter in the roots of riparian trees. (iii) This also indicates that aquatic fungi have evolved a safety system to guard themselves in an extreme habitat.

The basic information gathered on enzymes of these fungi such as amylase, cellulase, esteraser and pecitnase production, indicates that these fungi produce all kinds of enzymes. Only one has to investigate. There is no doubt that aquatic fungi are no less capable than fungi from other habitats. Our unpublished efforts also proved that aquatic fungi are producers of extremly potent natural components of high therapeutic values. An isolate of *Ingoldiella hamata* gathered from streams of Molem has shown positive activity

against certan human ailment. It may be said that fungi inhabiting difficult ecological status could be very creative in their chemical mechinary.

The seven new taxa including one genus and 6 species described in this thesis (of which 4 alrealy published) are undoubtedly a major contribution from this work. In all, hitherto described species of aquatic fungi known from world literature are 330. Of these, 78 species have been so far recorded from India. Fifty eight species, that is 59.2% of Indian taxa are now freshly described in this thesis. Seven new species are added further. All these point out that streams of Western Ghats are immersely rich in their aquatic flora. It is not only a matter of pride for us to project this richness, but also to realise that Western Ghat is a 'gene rich' region as far as fungi are concerned. These fungi, now being accommodated in a culture collection as an *ex situ* repository in Goa University, are good for future biotechnology of our country.

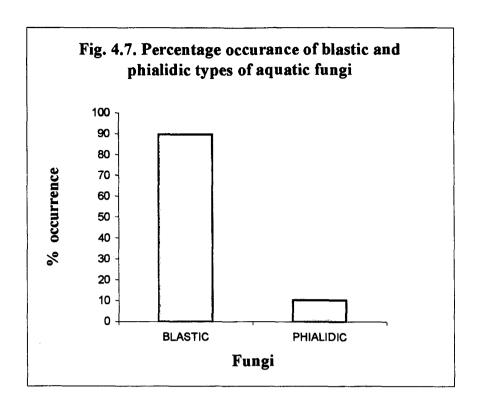
Table 4.1.2: <u>List of hyphomycetous fungi recovered</u>:

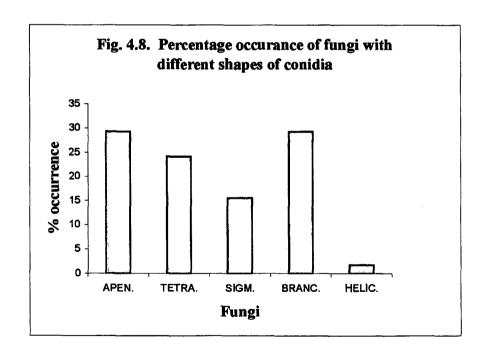
Strict aquatics	Casual aquatics
1. Actinospora megaolospora	1. Acremonium sp.
2. Alatospora acuminata	2. Alternaria alternata
3. Anguillospora crassa	3. Ardhachandra selenoides
4. Anguillospora furtiva	4. Ardhachandra sp.
5. Anguillospora longissima	5. Arxiella terrrestris
6. Articulospora tetracladia	6. Aspergillus niger
7. Articulospora sp.	7. Beltrania rhombica
8. Bahusutrabeeja angularis	8. Beltraniella odinae
9. Camposporium pellucidum	9. Brachysporiella laxa
10. Campylospora chaetocladia	10. Canalisporium caribense
11. Campylospora filicladia	11. Candelabrum brocchiatum
12. Centrospora acerina	12. Ceratosporium sp.
13. Chaetendophragmia triseptata	13. Cladosporium sp.
14. Condylospora spumigena	14. Chalara sp.
15. Dendrospora erecta	15. Colletotrichum dematium
16. Dendrospora yessemreddia sp. nov.	16. Corynespora sp.
17. Dendrosporium lobatum	17. Curvularia lunata
18. Dichotomophthoropsis aquatica sp. nov.	18. Cylindrocarpon sp.1
19. Dictyochaeta assamica	19. Cylindrocarpon sp.2
20. Diplocladiella scalaroides	20. Cylindrocarpon sp.3
21. Diploospora indica sp. nov.	21. Cylindrocladiopsis sp.
22. Flabellospora crassa	22. Cylindrocladium sp.1
23. Flabellospora multiradiata	23. Cylindrocladium sp.2
24. Flabellospora verticillata	24. Dactylaria aquatica sp. nov.
25. Flagellospora curvula	25. Dactylaria sp.
26. Helicomyces roseus	26. Dactylella ellipsospora
27. Helicosporium sp.1	27. Drechslera tripogonis
28. Helicopsorium sp.2	28. Endophragmia inaquiseptata
29. Helicosporium sp.3	29. Fusarium sp.1
30. Ingoldiella hamata	30. Fusarium sp.2
31. Isthmotricladia britanica	31. Gliomastrix murorum
32. Isthmotricladia laeensis	32. Gonatophragmium sp.
33. Lateriramulosa uniinflata	33. Gonytrichum macrocladium
34. Lemonniera aquatica	34. Hansfordia pulvinata
35. Lunulospora curvula	35. Helminthosporium sp.
36. Mycoleptodiscus indicus	36. Idriella angustispora

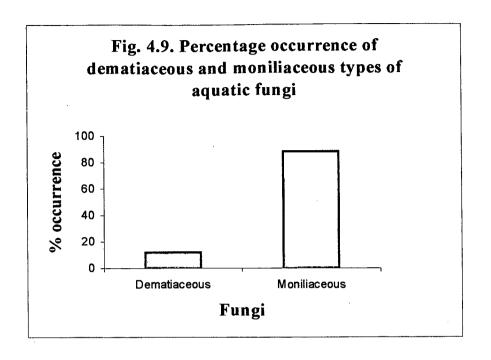
37. Nawawia filiformis	37. Idriella lunata
38. Paratozetella aquatica gen. et sp. nov.	38. Kumbhamaya jalapriya sp. nov.
39. Phalangispora constricta	39. Paecilomyces sp.
40. Robillarda phragmitis	40. Penicillium sp.
41. Scutisporus brunneus	41. Periconia sp.
42. Seimatosporium sp.	42. Pheoisaria sp.
43. Speiropsis hyalospora	43. Pithomyces chartarum
44. Speiropsis pedatospora	44. Pseudobotrytis terrestris
45. Subulispora sp.1	45. Pseudospiropes loratus
46. Subulispora sp.2	46. Ramichloridium fasciculatum
47. Tetrachaetum elegans	47. Scolecobasidium sp.
48. Tetraploa aristata	48. Septonema sp.
49. Tricladium angulatum	49. Sesquicillium sp.
50. Tricladium splendens	50. Sopagraha sibika
51. Trinacrium indica sp. nov.	51. Sporedesmium sp.
52. Trincarium subtile	52. Thozetella sp.
53. Tripospermum myrti	53. Trichoderma sp.
54. Triscelophorus acuminatus	54. Tubercularia vulgaris
55. Triscelophorus konajensis	55. Vermiculariopsiella sp.
56. Triscelophorus monosporus	56. Verticillium sp.
57. Weisneriomyces javanicus	57. Virgaria sp.
58. Undetermined sp.1	58. Undetermined sp.7
59. Undetermined sp.2	59. Undetermined sp.8
60. Undetermined sp.3	60. Undetermined sp.9
61. Undetermined sp.4	61. Undetermined sp.10
62. Undetermined sp.5	
63. Undetermined sp.6	

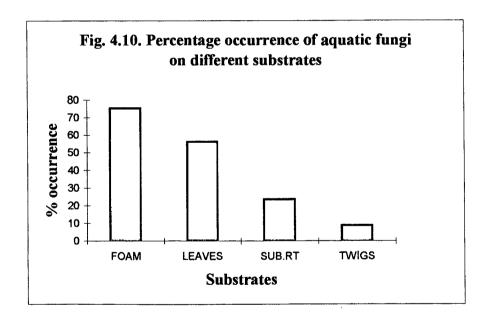
Table 4.1.3: <u>List of Ascomycetes and Coelomycetes recovered during the study</u>:

Ascomycetes	<u>Coelomycetes</u>	
Chaetomium nigricolor	Chaetomella sp.	
Emericella sp.	Robillarda phragmitis	
Nectria haematococca	Seimatosporium subulatum	
Xylaria sp.	Phoma sp.	
Undetermined sp.1	Pestalotiopsis sp.1	
Undetermined sp.2	Pestalotiopsis sp.2	
Undetermined sp.3		









CHAPTER 5:

SUMMARY.

SUMMARY

Our knowledge on freshwater fungi, especially the conidial fungi, has greatly increased by application of modern field and laboratory methods for recovery of pure cultures by single spore isolation techniques (Descals, 1997), mapping of submerged leaves (Shearer and Lane, 1983), biochemical studies (Chamier and Dixon, 1983), ecological experiments (Dix and Webster, 1995) and so on. While the more than two decade long earlier researches in India were directed mostly to exploratory surveys and ecology, studies on taxonomy, biology and physiology of aquatic fungi have remained largely untouched.

Of the so far known 330 species of freshwater fungi, approximately one fourth (78 taxa) has been reported from India. Of these, about 80% (62 species) were so far recorded from the streams and rivers of forests of Western Ghats of Karnataka, Kerala, Maharashtra and Tamil Nadu (Sridhar et al., 1992). Very little was known on diversity and abundance of these fungi from freshwater streams of Goa State. The present work was an attempt to fill this gap.

This thesis embodies the results of a detailed study carried out during the last two and a half years, on the diversity, abundance and activity of aquatic fungi in the streams of forests of Western Ghats in Goa State. Sampling was carried out in 12 streams and rivers, both seasonal and perennial, which included 3 streams of Bondla, Cotigao and Molem Wildlife Sanctuaries.

Floating foam, submerged and decaying leaf and twig litter and submerged live roots of riparian trees were collected from these sites at regular intervals in monsoon and post-monsoon seasons and brought to the laboratory. Where natural foam was not available, leaf litter was incubated in large glass jars and aerated in the laboratory to induce foam formation. Aquatic spores were individually isolated from the foam using simple techniques and pure cultures were established. As the cultures sporulated their identity was confirmed.

A total of 135 species of fungi belonging to 113 genera were recovered during the course of study. Based on their aquatic affinity, the fungi were categorised into 'strict aquatics' and 'casual aquatics'. The former are those which sporulated only when culture blocks maintained under water while the latter accommodated those sporulated above water. Taxonomically they belonged to Hyphomycetes (122), Coelomycetes (6) and Ascomycetes (7). The descriptions along with camera lucida illustrations of 102 aquatic conidial fungi belonging to 80 genera are given in detail in Chapter IV, Part I. In all, 63 species of strict aquatics and 61 species of casual aquatics were recovered from the the streams of Goa. These are listed under different headings in the discussion part. A dichotomous taxonomic key has been provided for the strict aquatics of Goan streams.

The major contribution from this work is the discovery of seven new species and a new genus of aquatic fungi from the treams of Goa. These are *Parathozetella* Sreekala & Bhat Gen. nov., *Dactylaria aquatica* Sreekala & Bhat sp. nov. (a root endophyte), *Dendrospora yessemreddia* Sreekala & Bhat sp. nov. (from foam), *Dichotomo-phthoropsis aquatica* Sreekala & Bhat sp. nov. (a root endophyte), *Diploospora indica* Sreekala & Bhat sp. nov. (from foam), *Kumbhamaya jalapriya* Sreekala & Bhat sp. nov.

(a root endophyte), *Parathozetella aquatica* Sreekala & Bhat sp. nov. (from foam) and *Trinacrium indica* Soosamma, Lekha, Sreekala & Bhat sp. nov. (from foam). The study proved that the forests of Western Ghats in Goa region are rich in their aquatic fungal flora.

The abundance and periodicity of occurrence of aquatic fungi on submerged leaf litter in different seasons studied in one stream each in three wildlife sanctuaries of Goa -Bondla, Cotigao and Molem - were described and discussed in Part II of the Chapter IV. The percentage relative abundance of the density of fungi on leaf litter, as expressed in water by aquatic spora, during the three different seasons, monsoon (M), post-monsoon (PM) and summer (S) was calculated and their significance of occurrence was statistically analysed using 'Analysis of Varience' (ANOVA) test. This study was done for two years, in 1999-2000 and 2000-2001. The tests showed very high significance level between the three seasons, and very low significance rate between the three sites. This can be attributed to the similar vegetation in the three sampling sites. The results indicated that in 1999-2000, during monsoon, highest number of species was at Bondla, followed by Cotigao and Molem. During post-monsoon the species richness remained in the same order of abundance in three sites. However, the abundance was in different order in summer; highest number of species being at Cotigao, followed by Bondla and Molem. Similar study in the year 2000-2001 showed that during monsoon, the highest number of species was at Cotigao, followed by Bondla and Molem. During post-monsoon, the species richness remained in the same order of abundance at the three sites. The order of abundance remained the same also in summer.

A total of 65 species of freshwater fungi were encountered on randomly sampled leaves. In both years (July 1999-May 2000 and July 2000-May 2001), highest number of species observed were during monsoon (from June to September) and the lowest during summer season (from February to May). Nevertheless, *Anguillospora crassa*, *Campylospora chaetocladia*, *C. filicladia*, *Lunulospora curvula* and *Triscelophorus monosporus* were found to be the major colonizers of leaf material round the year. Besides, *Chaetendophragmia triseptata*, *Dictyochaeta asamica*, *Helicomyces roseus*, *Pestalotiopsis sp.* and *Speiropsis pedatospora* which occurred throughout the year, can be treated as predominant species in the regional aquatic system. It may also be said that presence of aquatic hyphomycetes is primarily dependent on the availability of stream water and suitable submerged leaf litter in the stream.

The distribution and abundance of aquatic fungi at 12 sampling sites of Goa, as represented by conidia in water were significantly correlated with the sampling sites. The mean number of conidia was higher at the sampling sites of Bondla, Cotigao and Molem where the streams were covered by dense riparian canopy round the year. Less number of conidia and species of fungi were observed at the sites of Taleigao and Harvalem wherein very few riparian plants were observed overhanging the streams. This clearly showed that distribution of fungi is directly related with the vegetation around.

No clear-cut distribution pattern for the most abundant species was recognised in relation to stream water chemistry (pH and/or nutrients). The pattern of species richness seemed to be independent of the water chemistry. The distribution of fungi was more consistent in the presence of dense riparian canopy. Species richness was higher at all the three sites, which are endowed with rich riparian canopy. These variations are clearly

visible in rarefaction curves plotted with the sample size collected from each site against the expected number of species. Although higher concentration of individuals was found in Molem, as shown in the rarefaction curve, when the sample size was 150, the expected number of species for Cotigao sample was 32 and that of Molem 22. The diversity index slided from Cotigao to Molem. This interesting result may be attributed to the continuous (100%) tree canopy found along the stream of Cotigao.

every year was studied by setting up a simple in situ experiment labeled 'litter bag experiment' for a period of 8 months. Leaf litter of two riparian plant species, Hopea ponga and Dellinia indica, collected from adjoining terrestrial habitat, were placed in separate A4 size nylon mesh bags with # 3 mm mesh size and immersed in a stream at Molem wildlife sanctuary. In all, 24 species of aquaitc fungi belonging to 21 genera were recovered from the leaf litter of Dillenia indica during the 8 month litter-bag study period. From the leaf litter of Hopea ponga, 28 species belonging to 25 genera of aquatic fungi were recovered. Of these, Anguillospora crassa, Angulospora aquatica and Triscelophorus monosporus appeared in more frequently, in the litter bag of Dillenia indica. Fungi such as Ardhachandra selenoides, Flagellospora curvula, Lunulospora curvula and Triscelophorus konajensis appeared more frequently in the litter bag of Hopea ponga. The appearance of remaining species ranged from once to 4 times in both plant litter.

The results indicated that sizable number of the aquatic fungi were common to both plant litter. Fourteen out of 28 species were common to both plants, whereas the following species occurred only on *Dellinia indica*: *Cylindrocladium* sp.2, *Helicomyces*

sp., Thozetella sp. Speiropsis pedatospora, Trinacrium gracile and Vermispora sp. and Camposporium pellucidum, Lunulospora curvula, Ingoldiella hamata, Triscilosporus konajensis, Tripospermum myrti, Phalangospira constricta only on Hopea ponga

The study also pointed out that in the summer months, from February to April, while the litter bags were exposed to dryness due to drying of stream bed, on incubation several of the otherwise terrestrial forms were recovered from leaf litter. In the litter of Dellinia indica, species such as Ardhachandra selenoides, Cylindrocladium sp.1, Dactylella ellipsospora, Pestalotiopsis sp., Sporoschisma uniseptata and Weiseriomyces javanicus were found in the months of January to April. In Hopea ponga, Ardhachandra selenoides, Cylindrocladium sp.1, Corynespora sp., Beltrania rhombica, Dactylella ellipsospora, Dictyochaeta assamica and Fusarium sp were found.

The root endophytic association of aquatic fungi was studied by examining submerged roots of 7 riparian plant species, namely Alstonia scolaris, Calamus thwaitesii, Dellinia indica, Glaberus sp., Glochidion hohenackeri, Hopea ponga and Saraca asoca. The study offered an opportunity to compare and analyse the fate of aquatic fungi during summer months. The fungi such as Cylindrocladium sp.2, Dictyochaeta assamica, Fusarium sp.1, Pestalotiopsis sp, Robillarda sp., Seimatosporium sp., Triscelophorus monosporus recovered from litter bags of Dellinia indica and Anguillospora crassa, Campylospora filicladia, Corynespora sp., Dendrosporium lobatum, Dictyochaeta assamica, Flagellospora curvula, Robillarda phragmitis., Seimatosporium sp., Triscelophorus monosporus recovered from Hopea ponga were also found as root endophytes of same plants.

The root endophytes were isolated from primary, secondary and tertiary roots of all the 7 plants. In the order of abundance, the highest percent colonisation was observed in primary roots (80%), followed by secondary (50%) and tertiary root tissues (20%). The results indicated that primary roots are the major hosts or sanctuary for aquatic endophytes in difficult times.

In all, 96 nonsporulating morphotypes were isolated as root endophytes. Amongst these, maximum numbers were recovered from *Saraca asoca* (24), followed by *Dellinia indica* (23), *Hopea ponga* (18) and *Calamus thwaitesii* (16).

A priliminary study was carried out to examine the effect of mining rejects on growth and sporulation of aquatic fungi in laboratory condition. Foam and submerged leaf samples were collected from two perennial streams of Cudnem Iron Ore Mines, Goa. Single spore isolation was carried out to recover fungi from foam and incubated leaf litter. In all, 14 species of fungi belonging to 12 genera were observed from aerated leaf litter. When the same leaf litter was aerated with mining rejects, 12 species of fungi were recovered. It was also observed that there was a gradual decrease in the number (from 9 to 5) of species as the concentration of mining reject was increased.

Amoung the 800 plus isolates of fungi brought into pure culture during the course of present study, 60 randomly selected morphotypes were subjected to qualitative screening for a few enzymes, namely amylase, cellulase, esterase and pectinase. A few non-sporulating isolates (categorised as NS-1 to NS-13) which appeared as root endophytes were also subjected for enzyme screening.

Amongst the 60 isolates tested for production of different enzymes, except one (NS-1), all others showed varying degrees of enzyme activity. Two isolates, namely

Dactylaria sp.2 and Triscelophorus monosporus, exhibited ability to secrete all the enzymes. Maximum number (32) of isolates exhibited positiveness for esterase activity, followed by cellulase (30), pectinase (29) and the minimum for amylase (26). According to the clearance zone, some of the fungi such as Anguillospora longissima, Dendrosporium lobatum and NS-11 were found to be powerful producers of amylase. Dichotomophthoropsis aquatica, Isthmotricladia laeensis and NS-6 were powerful producers of cellulase. Beltraniella odinae and Triscelophorus monosporus were found to be active producers of esterase enzyme and Flabellospora multiradiata as producer of measurable quantity of pectinase.

Study of taxonomy and abundance, building up of a collection of pure cultures, preparation of an enzyme profile catalogue, setting up of a taxonomy and biology database for aquatic fungi from the streams and rivers of forests of Western Ghats, all should be considered as an honest and positive effort of documentation, conservation and utilization of fungal biodiversity in our country. The streams of Western Ghats are immersely rich in their aquatic flora. It is not only a matter of pride to unravel and project this invaluable biological wealth, but also to realize that Western Ghats are a 'gene rich' zone as far as aquatic microfungi are concerned. These fungi, now being accommodated in a culture collection as an *ex situ* repository in Goa University, are good for future utilization efforts. Study such as this will undoubtedly have a far-reaching positive impact not only in the development of fungal biotechnology but also for generating wealth from fungal resources in the future in our country.

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APPENDIX

List of publications:

- 1. Soosamma, M., G. Lekha, Sreekala, K. N. and D. J. Bhat. 2001. A new species of *Trinacrium* from submerged leaves from India. *Mycologia*. **93(6)**: 1200-1202.
- 2. Sreekala, K. Nair. and D. J. Bhat, 2001. *Diploospora indica*, a new species of hyphomycetes. *Mycotaxon*. **80**: 101-104.
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Mycologia, 93(6), 2001, pp. 1200-1202. O 2001 by The Mycological Society of America, Lawrence, KS 66044-8897

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A new species of Trinacrium from submerged leaves from India

M. Soosamma

G. Lekha

I have checked this proof. I have marked all changes or

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corrections

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Department of Botany, Mount Carmel College, Bangalore-560 052, India

K. N. Sreekala

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Abstract: Trinacrium indica sp. nov., isolated from submerged coffee leaves from India, is described and illustrated. The new taxon is compared with previously described species of Trinectium.

Key Words: Aquatic hyphomycetes, biodiversity, India, foam, submerged coffee leaves, taxonomy, triradiate conidia

.Freshwater hyphomycetes (Ingoldian Hyphomycetes) constitute an ecologically specialized group of microfungi which grow on fallen and decomposing plant litter submerged in stream and river waters. The conidia of these fungi are distinctly shaped in that they are tri- or tetraradiate, branched, sigmoid, curved or appendaged (Ingold 1975). In the last two decades, mountain streams and rivers traversing the Western and Eastern Chats in southern India and the central and eastern Himalayas in the northern India have been surveyed, and efforts made to document the distribution of these fungi (Subramanian and Bhat 1981, Bhat and Chien 1990, Sridhar et al 1992, Sati and Tiwari 1997). Nearly one third of the total 318 species of freshwater hyphomycetes recorded from all over the world are documented from India (Sati and Tiwari 1997).

During a survey of freshwater hyphomycetes of Western Ghat forests, triradiate conidia with a recurved arm of an aquatic fungus were gathered in foam and submerged coffee leaves from a stream in a coffee plantation in Somwarpet, Kodagu District, Karnataka State, India. The fungus is undescribed. We describe it here as a new taxon of Trinacrium.

Single conidia of the fungus were transferred to 2% malt extract agar medium supplemented with antibiotic (Streptomycin sulfate 0.04 g and Penicillin G 0.04 g dissolved in 10 mL of sterile distilled water and added to 1 Lof cooling sterile medium). The fungus grew slowly on the culture medium but submerging 14-d-old culture strips in sterile distilled water yielded abundant conidia and conidiogenous structures at various stages of development within 3-4 d. Leaf litter of Coffea arabica L., gathered from the same locality and incubated in sterile distilled water in a vertical glass jar with agitator by a continuous flow of air from a fish-aerator also yielded abundant conidia after 2 d. The fungus sporulated in malt extract agar medium only after 6 wk of incubation.

Trinacrium indica Soosamma, Lekha, Sreekala and Bhat, sp. nov.

Coloniae tardae crescentes in ME agar, cum marginis circinatis, albidae ad ochraceae, aequusae, 1.2-1.5 cm diam in 10 ad 12 d inter 25 C. Mycelium immersum ramosum, septatum, liyalina hyphae 1-3 µm latum. Conidiophora micronematosa, mononematosa, formo lateralia on hyphae, simplicia, hyalina, laevis, 1.5-2.5 × 1.5-2.5 µm. Cellulae conidiogenae monoblasticae, laeviae, determinatae ad sympodiales, hyalinae, apicalae truncatae. Conidia solitaria, triradiata, Y-formia, bifurcatus asymetricalae, septata, hyalina, laevia, ad leniter constricta ac septata a punctatis bifurcata, pallude brunnea in massa, originata in massa 1-10 ab loci, principalis axis cylindrica, recta vel flexuosa, 1-3-septata, 28-56 \times 1.2-2.5 μ m, cum 2 cylindricale ac divergenta brachium originata synchronous ad apicem principalis axis; brachia parvus recta vel flexuosa, rotundata in finibus, 1-2septata, 23-35 × 1.2-2.5 μm; brachia longa 1-3-septata, recurvata distincta in medius inter, 46-65 \times 1.2-2.5 μm .

Colonies on malt' extract agar slow growing, with circular margin, white to gray, flat, 1.2-1.5 cm diam in 10-12 d at 25 C. Mycelium mostly immersed, composed of branched, septate, hyaline hyphae 1-3 µm wide. Conidiophores micronematous, mononematous, formed laterally on vegetative hyphae, simple, hyaline, smooth, $1.5-2.5 \times 1.5-2.5 \mu m$. Conidiogenous cells monoblastic, simple, determinate to sympodial, hyaline, truncate at the apex after conidial secession. Conidia solitary, triradiate, asymetrically Y-shaped, septate, hyaline, smooth, thin-walled, slightly constricted and septate at bifurcation point, pale brown in mass, developing in groups of 1-10 from one locus, with main axis cylindrical, erect or flexuous, 1-3-septate, $28-56 \times 1.2-2.5 \mu m$, with two cylindrical,

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SOOSAMMA ET AL: TRUNACRUUM FROM INDIA

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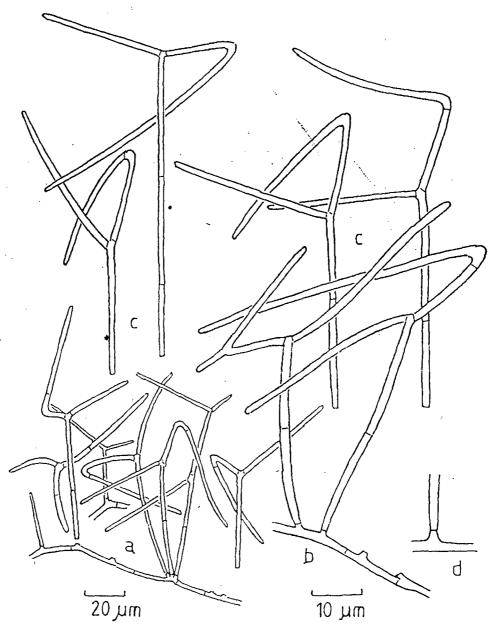


Fig. 1. Trinacrium indica, a. Conidiogenesis (in lower magnification), b. Conidiogenesis (in higher magnification); c. Conidia; d. Conidiogeneous locus (note the holoblastic conidiogenesis).

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synchronously developing and divergent arms at the apex of the main axis; short arm straight or slightly flexuous, rounded at the tip, 1-2-septate, $23-35 \times 1.2-2.5 \mu m$; long-arm 1-3-septate, distinctly recurved at midpoint, $46-65 \times 1.2-2.5 \mu m$.

Specimen examined. INDIA, KARNATAKA: Kodagu, Somwarpet. On decaying and submerged leaves of Coffea arabica, Nov 1998, [MI 385464] HOLOTYPE). Soosamma Mathews, GUFCC 168 (ISOTYPE). Dried MEA culture mat established from single conidium isolated from decaying and submerged leaves of Coffea arabica, 18 Nov 1998, Sreekala Nair, GUFCC 169 (PARATYPE).

The genus Trinacrium Riess, typified by T. subtile Riess, is characterized by production of triradiate conidia, consisting of a main axis bearing two divergent radiate arms (Saccardo 1886). The conidia in general are hyaline, smooth-walled, septate and Y-shaped. Depending upon the species, the main axis and arms of the conidia may be thin and delicate, clavate or further branched. Based on conidiophore and conidiogenous cell morphology and conidium morphology and ontogeny, seven species of Trinacrium have been described. These include T. angamosense Matsushima (1995), T. gracile Massushima (1975), T. inacquiramiserum Matsushima (1989), T. parvisporum Matsushima (1987), T. robustum Tzean & Chen (1989), T. subtile Riess (Saccardo 1886, Matsushima 1975) and T. torulosum Sacc. & Malbr. (Saccardo 1886). Species of Trinacrium generally inhabit decaying leaf litter (Matsushima 1975, 1987, 1989, 1995) but are also reported as parasitizing oospores of Pythium (Drechsler 1938). Ando (1992) noted that T. subtile has an affinity for aquatic environments. Tzean and Chen (1989) compared the species of Trinacrium and concluded that these can be distinguished by length and width of main axis and arms, or the shape of the conidia. The conidium of T. indica shows some similarity to T. subtile and T. gracile in its overall dimensions but differs markedly by its distinctly recurved long arm.

ACKNOWLEDGMENT

Two of us (S.M. and L.C.) sincerely thank Dr. (Sr) M. Genevieue, Principal, Mt Carmel College, Bangalore, for constant encouragement. This work is supported by a research grant to D.J. Bhat from the Department of Science & Technology, Government of India, New Delhi.

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DIPLOOSPORA INDICA, A NEW SPECIES OF HYPHOMYCETES

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ABSTRACT

A new species of Hyphomycetes, *Diploospora indica* Sreekala & Bhat, isolated from freshwater foam and submerged leaf litter of *Dipterocarpus laevis*, is described from India. The new taxon is compared with known species of the genus and other similar genera.

KEY WORDS: limnology, streams, taxonomy, India.

INTRODUCTION i

During a taxonomic survey of aquatic fungi of the forests of Western Ghat mountain ranges in southern India, a number of elongate, aseptate, fusiform, smooth and dorsi-ventrally flat conidia were encountered in foam and on submerged leaf litter of *Dipterocarpus laevis* Buch. (Dipterocarpae) gathered from a freshwater stream in Cotigao Wildlife Sanctuary in the State of Goa. The fungus was brought to culture and is described here as a new species of the genus *Diploospora* Grove.

METERIALS AND METHODS

Fresh foam was gathered by gently shoveling a glass petri plate lid through a foam cake found accumulated along the edge of a fast flowing stream in Cotigao Wildlife Sanctuary, Goa. A few drops of the liquid foam were flooded over 2% malt extract agar (MEA) plates mixed with a cocktail of antibiotics (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 sterile distilled water and added to 1 L of cooling sterile MEA medium). The foam-flooded agar plates were rotated orbitally so as to have an uniform spread of the spores. The foam-spora plates were transported to the laboratory in an ice box maintained at 4°C.

The submerged leaf litter collected from the stream was thoroughly rinsed, initially in stream water and later with deionized tap water after transporting to the laboratory in fresh polythene bags. After incubation in sterile distilled water in a vertical glass jar and agitation at 20-22°C in a continuous flow of sterile air, conidia laden bubbles were observed on the surface after five days. They were transferred to MEA plates by gently scooping the surface water from the jar and spreading it over the agar surface. Single germinating conidia were detected under the dissecting microscope and aseptically isolated from both foam and leaves onto fresh MEA plates using a sterile flattened needle. The plates, sealed with parafilm in order to protect them from mites and other contaminants, were incubated in a day light growth chamber at 20-22°C. The fungus grew well reaching up to 10 mm diam in 10 days but sporulated sparsely on the agar surface even after three weeks of incubation.

DESCRIPTION

Diploospora indica Sreekala et Bhat anam. sp. nov.

(Fig. 1)

Fungi conidiali, Hyphomycetes. Coloniae concentricae, effusae, albae, margine laeviae; reversione concoloriae vel pallide brunneae. Mycelium superficiale et immersum, ex hyphis ramosis, septatis, laevibus, incoloris, 1.5-2 μm crassis compositum. Conidiophora terminali vel laterali, mononematica, recta, flexuosa, laevia, 2-7-septata, ramosa, pallide brunnea, tenue crassitunicata, 75-125 x 3-5 μm. Cellulae conidiogenae integratae, polyblasticae, apicales, post secessionem conidiorum ad apicem, truncatae, 10-22 x 3-4 μm. Conidia in pseudocatenas acropetas longas, ad basim ramosas, fusoidea, utrinque truncata, laevia, aseptata, dorsi-ventrale leviter applanata, incolorata vel subhyalina, pallide brunnea in massa, 12-33 x 3-4 μm; ramoconidia aseptata, longa, basi truncata; cicatrices terminales. 2-5.

HOLOTYPE, dried culture mat derived from single spore isolate IMI 384550 (1); ex Herb. No. GUFCC-161 (Isotype); Cotigao Wild Life Sanctuary, South Goa, India, Sreekala, K. Nair; 19.VII.1999.

Conidial fungus, Hyphomycete. Colonies consisting of concentrically zonate hyphae alternating with conidial mass, effuse, white, with smooth margin, slow growing, reaching 0.5-1.0 cm diam. in 10 days, reverse of the colony colourless to pale brown. Mycelium partly superficial, composed of smooth, colourless hyphae 1.5-2 µm wide. Conidiophores terminal or lateral, mononematous, erect, flexuous, smooth, moderately thick-walled, 2-7-septate, branched, pale brown in mass, 75-125 x 3-5 µm. Conidiogenous cells integrated, polyblastic, 10-22 x 3-4 µm, apically truncate after secession of conidia. Conidia in long, acropetal, simple to proximally branched, false chains, fusiform, narrow and truncate at both ends, dorsi-ventrally flat, smooth-walled, aseptate, hyaline to very slightly pigmented, pale brown in mass, 12-33 x 3-4 µm; ramoconidia aseptate, truncate at the base, with 2-5 terminal flat scars.

Hughes (1968) examined and redescribed *Diploospora rosea* Grove, the type species of the genus *Diploospora* Grove, from the type collection maintained in W.B. Grove's herbarium in Herb. BM. The genus *Diploospora* is characterised by its catenate, hyaline but in mass dull pink-coloured, 1-2-septate, conidia in acropetal,

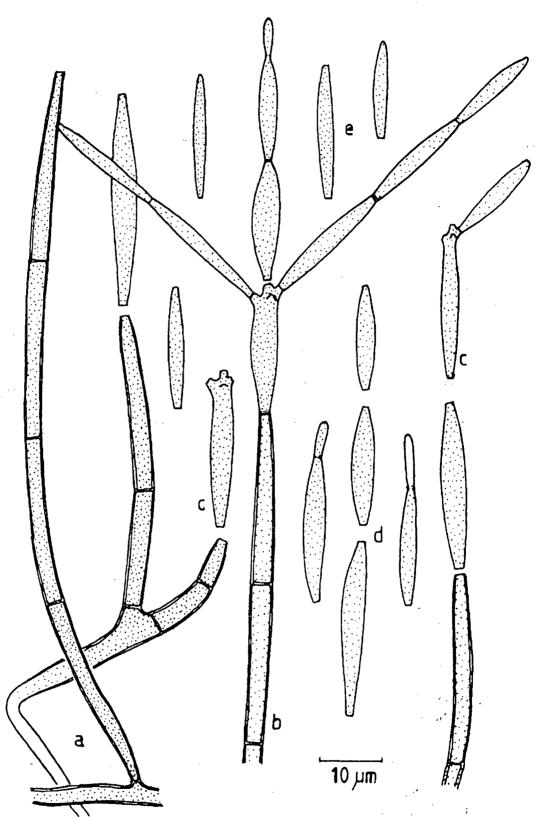


Fig.1. Diploospora indica. a. conidiophores, b. conidiophore with ramoconidia and conidia, c. ramoconidia, d. mature conidium showing acropetal development of conidia, e. conidia. (All drawn from pure culture).

branched chains developing on hyaline, simple, branched conidiophores. The conidia of D. rosea are cylindrical to broadly ellipsoidal and truncate at each slightly denticulate raised end (Hughes, 1968). The conidia of the second species known in the genus, D. longispora Matsushima (1975), are 1-3 septate and cylindrical. Those of D. indica are however aseptate and fusiform. Diploospora rosea and D. longispora are terrestrial whereas the conidia of D. indica were collected in an aquatic habitat although they may have been washed in from nearby litter.

The genus *Diploospora* shows some similarity with dematiaceous genera such as *Cladosporium* Link, Lylea Morgan-Jones, *Ramularia* Unger, *Septocylindrium* Bonord. ex Sacc. and *Septonema* Corda wherein the conidiophores are robust and stiff and the catenate conidia are moderately to dark-coloured and of varying shapes (Carmichael et al., 1980; Matsushima, 1975).

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DENDROSPORA YESSEMREDDEA SP. NOV. FROM FRESHWATER FOAM

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ABSTRACT

A new species of the Hyphomycetes. *Dendrospora yessemreddea* Nair et Bhat, isolated from a freshwater stream of the Western Ghat forests of Goa, India, is described and illustrated. The new taxon is compared with the so far known species in the genus.

Key words: Branched conidia. *Dendrospora* sp., Hyphomycetes, taxonomy, freshwater forest streams, Western Ghats, India.

INTRODUCTION

We deem it a privilege to have been invited to submit this paper to a volume being brought out in honour of Professor S.M. Reddy, Warangal, Andra Pradesh, India, who made an indelible mark in the field of mycology and plant pathology in India by his invaluable contributions.

During a taxonomic survey of aquatic fungi of the Western Ghat mountain forest streams in southern India, a few distinctly branched, septate, smooth and hyaline conidia were encountered in foam gathered from a freshwater stream in Bondla Wildlife Sanctuary in the State of Goa. The fungus was brought into pure culture by single

spore isolation. It was found to be hitherto unknown and is described here as a new species of the genus *Dendrospora*.

METERIALS AND METHODS

The methods described by Descals (1997) for isolation of aquatic fungi were followed in this work. A few drops of freshly gathered foam were spread over 2% malt extract agar(MEA) plates incorporated with a cocktail of antibiotics (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

lled water and added to 1 L of MEA). foam-inoculated plates were sported to the laboratory in an ice box . Single germinating conidia were tically isolated into fresh MEA plates g a sterile needle. The fungus grew erately well at 20-23°C and sporulated agar surface after two weeks of pation.

!rospora yessemreddea Sreekala et anam. sp. nov. (Fig. 1)

1. In honour of Professor S.M. Reddy) ungi conidiali, Hyphomycetes. iiae effusae, albae, modice tarda cresente ad MEA, 2.3 cm diam, in 10 coloniae inverter hyaliniae vel albae. lium immersum, ex hyphis levis, tis, ramosis, hyalinis composita. liophora mononematosa, non-ramosa, i, recta vel flexuosa, levia, 0-3 septata, 8-40 х 2-3 m. Cellulae ogenae incorporatae. terminalis, blasticae. truncata ad apicem. ices conidialis nonincrassatae: solitaria. holoblastica. a, lateriter ramosa usque ad 15, axis elongata, flexuosa, 2-24 septata. a, 30-280 x 2-4 m; ad lateralis usque Im longa.

olotype: Dried culture mat derived single spore isolation of the fungus; a Wild Life Sanctuary, Goa, India; ila, K. Nair; 15.8.1999; Herb. No. C-159.

nidial fungus. Hyphomycete. 2s effuse, white, moderately slow g, attaining a diam. of 2.3 cm in MEA 10 days: reverse of the colony ess to white. Mycelium immersed, sed of smooth, septate, branched, hyphae 1-2 m , wide. ophores mononematous. simple. flexuous. hyaline, smooth. state, unbranched, 8-40 x 2-3 m.

Conidiogenous cells monoblastic, integrated, terminal, truncate at the tip after secession of conidium. Conidia solitary, hyaline, smooth, septate, with several (up to 25) lateral branches developing closely from a flexible main axis and resulting into a flat fan-shaped structure, generally seen afloat on surface of water; main axis elongated, flexible, 2-24-septate, 30-280 x 2-4 m; laterals up to 200 m long, sometimes branching further into secondary laterals, inserted to the main through a narrow constriction.

The fungus when grown on potato dextrose agar (PDA) and in corn meal agar (CMA) media showed a slightly faster growth rate, i.e. 3.7 cm diam in 10 days. In PDA, the colony was flat and did not sporulate whereas the sporulation was moderately good both in MEA and CMA after 10 days of incubation.

Dendrospora Ingold, typified by D. erecta Ingold (1943), has in addition 7 species so far described in the genus (Descals, 1997; Ingold, 1975; Regelsberger et al., 1987). These include D. fastuosa Descals & Webster, D. fusca Descals & Webster, D. juncicola Iqbal (1972), D. nana Descals & Webster, D. polymorpha Descals. D. tenella Descals & Webster and D. torulosa Descals & Webster. Amongst them, D. erecta shows some similarity to D. yessemreddea in its distinct main axis, several closely laid lateral and secondary branches and overall dimensions. However, the main axis in D. erecta is straight whereas in D. yessemreddea it is flexuous.

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This work was supported by a research grant to D.J. Bhat from the Department of Science & Technology, Government of India.

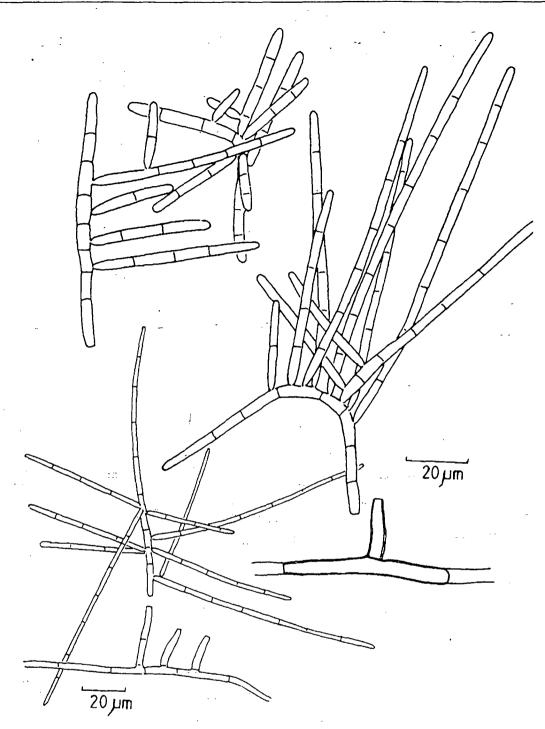


Fig. 1. Dendrospora sp., conidiophores and conidia.

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