



Original Research

Diversity of aquatic hyphomycetes in two coastal streams of southwest India

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Abstract

Aquatic hyphomycetes (or Ingoldian fungi) are widely distributed mycota in streams involved in the breakdown of organic matter and energy flow. In this study, diversity of aquatic hyphomycetes has been compared in two tropical coastal streams in southwest India. Water, foam, and litter samples (leaves and woody debris) in streams were assessed for aquatic hyphomycetes in relation to physicochemical characteristics. Water and foam samples were directly examined for the presence of conidia, while the leaf and woody litters were assessed by aeration in bubble chambers to release conidia. The species richness and diversity were higher in foam samples compared to other samples. Many species occurring in fairly cold climatic conditions in the Western Ghats were represented in the present study (*Clavospora tentacula*, *Condylospora spumigena*, *Flabellospora crassa*, *F. multiradiata*, *F. verticillata*, *Isthmotricladia gombakiensis*, *Lunulospora cymbiformis*, *Synnematophora constricta*, and *Varicosporium elodeae*). This study also revealed a new record of eight species on the Indian subcontinent (*Campylospora leptosoma*, *Condylospora vietnamensis*, *Helicoma atroseptatum*, *Helicomyces collegatus*, *H. hyderabadensis*, *Helicosporium aureum*, *Hydrometrospora symmetrica*, and *Setosynnema isthmosporum*). Further studies are warranted on the diversity, substrate preference, and ecological roles of aquatic hyphomycetes in coastal streams.

Keywords: Diversity, Foam, Incubation, Leaf litter, Wood

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1. Introduction

Freshwater mycota constitute phylogenetically variable assemblages of fungi (traditional Zygomycota, Cryptomycota, Chytridiomycota, and Ascomycota) in diverse habitats (Hyde &



Jeewon, 2003; Vijaykrishna et al., 2005; Raja et al., 2018; Luo et al., 2019). However, the distribution of fungi in nature is largely unknown, as the ecological setup shapes the distributions (Peay et al., 2016). Their lifestyles differ widely as endophytic, pathogenic, saprophytic, parasitic, and planktonic, which are dependent on the ecological niches of nature (Zhang et al., 2008; Huang et al., 2018a, 2018b). Freshwater hyphomycetes have morphologically distinguishable multiradiate (staurosporus), sigmoid (scolecosporus), and coiled (helicosporus) conidia (Ingold, 1975; Subramanian & Bhat, 1981; Marvanová, 1997; Bärlocher & Marvanová, 2010; Zhao et al., 2007; Gulis et al., 2020; Seena et al., 2022). However, the conidia of some species have conventional shapes like fusiform, oval, or spherical (e.g., *Cylindrocarpon*, *Dimorphospora*, *Tumularia*, and *Vermispora*). The blizzard conidial shapes indicate their functional roles in aquatic habitats for sedimentation, flotation, impaction, colonization, and dissemination. The community structure as well as the functional diversity of aquatic fungi have been correlated to the water quality in a coastal watershed based on environmental DNA studies (French et al., 2024).

Freshwater hyphomycetes constitute up to 335 morphospecies with a worldwide geographic distribution with the highest richness and diversity in middle latitudes (Duarte et al., 2016; Friggens et al., 2017). Although they prefer lotic habitats, they occur outside their usual habitats (stream banks, forest floors, canopy, and tree holes) throughout the world (Sridhar, 2009; Chauvet et al., 2016; Magyar et al., 2021; Sharathchandra & Sridhar, 2022). The Western Ghats of India, as a hotspot of diversity, represent up to 100 morphospecies of aquatic hyphomycetes, which is equivalent to 30 % of those reported worldwide (Sridhar, 2021).

Earlier studies on aquatic hyphomycetes in the southwest coastal region include samples from streams, terrestrial litter, tree canopies, tree holes, and epiphytic ferns (Sridhar et al., 2006, 2013, 2020; Sharathchandra & Sridhar, 2021, 2022). Although the aquatic hyphomycete richness of coastal streams is not comparable to the streams of the Western Ghats, the streams of the west coast represent attractive species. However, there is no comparative account of the diversity of aquatic hyphomycetes occurring on different substrates in coastal streams (Sridhar 2021). Hence, this study extends the exploration of freshwater hyphomycetes in two coastal streams on the west coast of India possessing contrasting water qualities and riparian vegetation.

2. Materials and Methods

Samples were collected from two freshwater streams flowing through scrub jungles in southwest India with differing water qualities and riparian vegetation. Drift conidia of aquatic hyphomycetes in water and accumulated conidia in foam samples were assessed. Colonized aquatic hyphomycetes were assessed in submerged leaf litter and wood (bark and cambium).

2.1 Study Areas and Sampling Sites

Schematic experimental strategies to study aquatic hyphomycetes are presented in Fig. 1. The occurrence of aquatic hyphomycetes was assessed and compared in Konaje and Kallimaru streams. The survey was performed monthly during the post-monsoon period (October through December, 2021).

The location selected in Konaje stream is a third-order reach passing through the laterite terrain with scrub jungle along with plantations about 1 km from Mangalore University Campus (12°48' N, 74°55' E; 90 m asl) (Fig. 2). The stream bed is largely lateritic, with red soil mixed with humus. The major tree species found were *Acrocarpus fraxinifolius*, *Adenanthera*



pavonina, *Anacardium occidentale*, *Artocarpus heterophyllus*, *Carallia brachiata*, *Careya arborea*, *Cycas circinalis*, *Ficus benghalensis*, *Ficus religiosa*, *Garcinia indica*, *Hopea parviflora*, *Holigarna arnottiana*, *Macaranga peltata*, *Mangifera indica*, *Morinda citrifolia*, *Psidium guajava*, *Pterocarpus marsupium*, *Rhizophora racemosa*, *Sapindus trifoliatus*, *Spondias mombin*, *Tamarindus indica*, and *Tectona grandis*.

Kallimaru stream is located about 1.5 km from Mangalore University Campus, passing through mixed vegetation, *Myristica* swamp, scrub jungle, and paddy fields (12.82°49' N, 74.92°55' E; 28 m asl). The soil texture in the stream bed is largely sandy loam, with humus in some stretches. The major tree species found were *Cycas circinalis*, *Holigarna arnottiana*, *Hopea parviflora*, *Macaranga peltata*, *Myristica malabarica*, *Psidium guajava*, *Syzygium cumini*, *Theobroma cacao*, *Tectona grandis*, and *Vateria indica*.

Humidity and air temperature were assessed under the canopy of riparian vegetation using a Mextech Digital Thermo Hygrometer (Model # M288CTHW, Mextech Technologies India Private Limited, Mumbai, India).

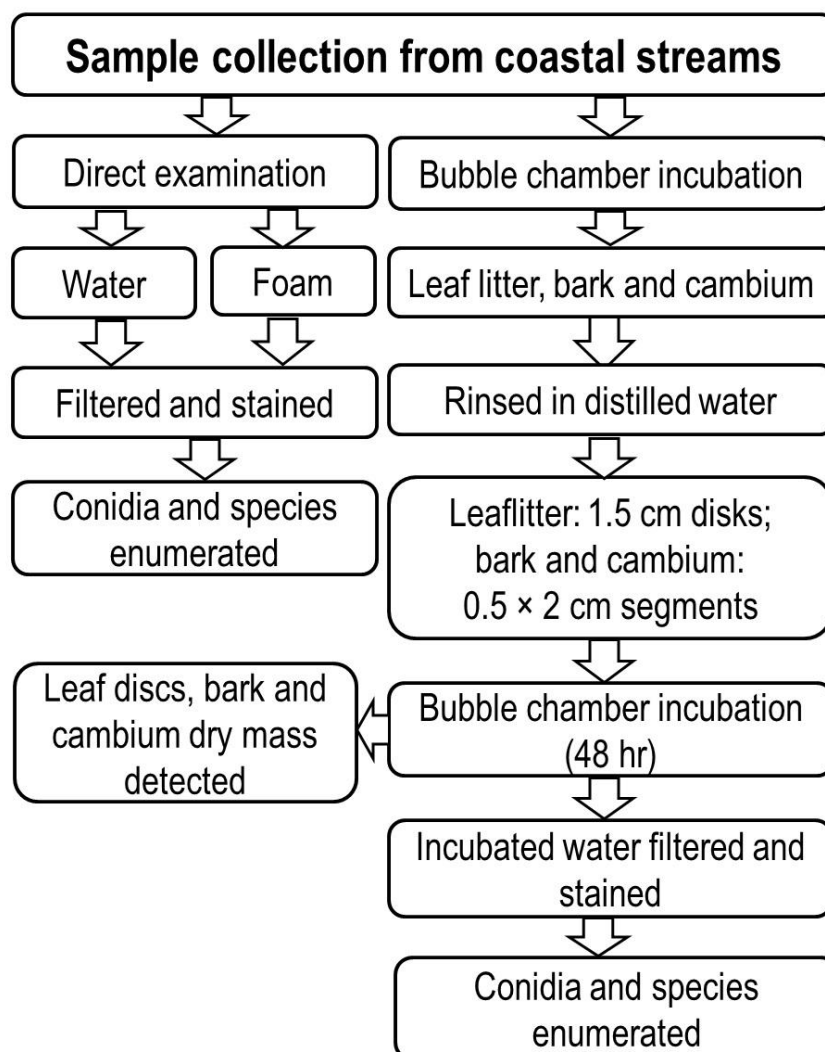


Fig. 1 - Schematic presentation of an experimental design to assess the occurrence of aquatic hyphomycetes in two coastal streams



Fig. 2 - Water flowing through the lateritic base (a), granite boulders (b), mini water fall (c), and accumulation of foam amidst granite boulders (d) in Konaje stream

2.2 Water Quality

The stream water temperature was assessed by a mercury thermometer (N.S. Dimple Thermometer, Model #17873, New Delhi, India). Water pH, conductivity, and total dissolved solids (TDS) were analyzed by a water analysis kit (Water Analyzer # 371, Systronics India Ltd., Ahmedabad, India). The water samples were fixed on the sampling sites to determine the dissolved oxygen by Winkler's method (APHA, 2017). The total alkalinity (titration using strong acid, methyl orange, and phenolphthalein indicators), total hardness (ethylene diamine tetraacetic acid titration using eriochrome black-T and murexide indicators), chloride (argentometric method), and contents were determined based on APHA (2017). The contents of inorganic phosphate (stannous chloride method), nitrate (brucine sulfanilic acid method), silicate (molybdosilicate method), and sulfate (turbidometric method) were assessed by APHA (2017). Magnesium has been estimated by the difference between hardness and calcium as CaCO_3 (APHA 2017). Water chemistry was evaluated using a NanoPhotometer (18V DC, 50VA) (Serial #1769, IMPLIN GmbH, Munich, Germany).

2.3 Drift and Accumulated Conidia

Stream water (25 ml from four locations per stream) was passed through filters (MF-Millipore Membrane Filter #SMWP02500, mixed cellulose esters; diam., 25 mm; porosity, 5 μm). The filters were stained with 1 % lactophenol aniline blue on the sampling spot and preserved in boxes to screen using a Nikon microscope (Model #ECLIPSE Ni-U 941966, Nikon Corporation, Tokyo, Japan) for qualitative and quantitative assessment of conidia by mounting filters with a few drops of lactic acid.



Freshly accumulated 2–3 foam samples were collected from the streams in a wide-mouthed container using a spoon, brought to the laboratory, and processed within 30 min. Foam samples were diluted with distilled water. Aliquots of diluted foam were filtered through Millipore filters, stained with 1 % aniline blue, and preserved in the dark. The filters were screened as detailed in water samples, and 300 conidia were scored for quantitative and qualitative assessment of conidia lodged on the filters.

2.4 Colonized Fungi

Naturally submerged leaf litters were collected from the streams and brought to the laboratory. Four to five different leaves were rinsed in water, piled and pressed into 1.5 cm discs using a cork-borer, and mixed. Five to six leaf disks were transferred to 250 ml Erlenmeyer flasks containing 150 ml of sterile distilled water (Iqbal & Webster, 1973; Bärlocher, 2020). The discs were aerated using Pasteur pipettes for up to 48 hr to allow the mycelia in the leaf discs to release the conidia. The aerated water was passed through Millipore filters (porosity 5 μm ; diam., 4.7 cm), stained with aniline blue, and screened for the presence of conidia as described for water and foam samples. Average conidia in three replicate leaf samples on a dry weight basis were compiled (the dry mass of leaf discs was gravimetrically determined by drying at 80 °C for 24 hr in a hot-air oven).

Similar to leaf litter, submerged woody litter was collected. Bark and cambium were separated and cut into 2 \times 0.5 cm segments. Five to six bark and cambium segments were separately aerated in bubble chambers as detailed for leaf litter to express the release of conidia in three replicate samples on a dry mass basis.

2.5 Fungal Identification

Conidia of aquatic hyphomycetes found in water, foam, leaf litter, bark, and cambium were identified based on morphology and measurements using a Nikon microscope (Ingold, 1975; Subramanian & Bhat, 1981; Nawawi, 1985; Marvanová, 1997; Santos-Flores & Betancourt-Lopez, 1997; Zhao et al., 2007; Bärlocher & Marvanová, 2010; Gulis et al., 2020).

2.6 Data Analysis

The physicochemical characters of stream location and water samples were compared among the two streams using a *t*-test (StatSoft Inc, 2008). The percent relative abundance (RA) of each species in water, foam, leaf litter, bark, and cambium were calculated considering the frequency of occurrence of a specific species and the sum of the frequency of occurrence of all species found. The Simpson's and Shannon's diversities with Pielou's equitability of aquatic hyphomycetes in water, foam, leaf litter, bark, and cambium were calculated (Pielou, 1975; Magurran, 1988).

3. Results and Discussion

3.1 Water Quality

The air temperature was in the mesophilic range, significantly higher in Kallimaru than Konaje stream ($p < 0.05$) (Table 1). The stream water temperature was in the mesophilic range, which was significantly higher in Kallimaru stream than Konaje stream ($p < 0.001$). Both streams were in the alkaline range, which was significantly higher in Kallimaru than Konaje stream ($p < 0.01$). Conductivity as well as dissolved solids were higher in Konaje than Kallimaru stream ($p < 0.001$). Dissolved oxygen, total alkalinity, and phosphate were not significantly different between the streams ($p > 0.05$). Sulfate, nitrate, silicate, chloride, and magnesium were



significantly higher in the Konaje stream than the Kallimaru stream ($p < 0.01$). Atmospheric humidity was similar in both streams.

Table 1. Physicochemical characteristics of two coastal streams ($n=9 \pm \text{SD}$; t -test, *, 0.05; **, $p < 0.01$; ***, $p < 0.001$)

	Konaje stream	Kallimaru stream
Air temperature (°C)	29.6 \pm 1.2	31.1 \pm 1.3*
Humidity (%)	69.0 \pm 2.3	69.0 \pm 4.0
Water temperature (°C)	24.2 \pm 0.3	26.0 \pm 0.6***
pH	8.0 \pm 0.3	8.4 \pm 0.2**
Conductivity ($\mu\text{S}/\text{cm}$)	135.0 \pm 19.9***	112.6 \pm 18.7
Total dissolved solids (mg/l)	89.5 \pm 16.2***	64.6 \pm 25.0
Dissolved oxygen (mg/l)	8.1 \pm 1.0	7.8 \pm 0.8
Total hardness (mg/l)	43.8 \pm 20.7	93.2 \pm 13.4***
Total alkalinity (mg/l)	87.4 \pm 19.3	90.8 \pm 12.0
Phosphate (mg/l)	2.6 \pm 0.5	2.3 \pm 0.7
Sulfate (mg/l)	4.4 \pm 1.8***	1.8 \pm 0.9
Nitrate (mg/l)	2.6 \pm 0.3**	1.8 \pm 0.8
Silicate (mg/l)	29.3 \pm 3.8***	20.8 \pm 5.2
Chloride (mg/l)	46.8 \pm 3.3***	37.0 \pm 6.7
Magnesium (mg/l)	2.3 \pm 0.4**	1.7 \pm 0.4

3.2 Drift and Accumulated Conidia

Water filtration resulted in 11 and 10 species of aquatic hyphomycetes in Konaje and Kallimaru streams, respectively (Table 2), with an overlap of six species (Fig. 3a). The conidial score was higher in Kallimaru than in the Koanje stream (53 vs. 46). *Anguillospora longissima*, *Lunulospora curvula*, and *L. cymbiformis* were common among the top five species in two streams. Filtration of foam resulted in scoring 25 and 27 species in Konaje and Kallimaru streams, respectively (Table 3), with an overlap of 12 species (Fig. 3b). *Anguillospora longissima*, *Helicoma atroseptum*, *Lunulospora curvula*, and *Triscelophorus acuminatus* were common among the top five species. A greater number of species were found in foam samples compared to water, leaf litter, bark, and cambium (25–27 vs. 7–15). In fact, conidial richness in foam samples reveals qualitative rather than quantitative differences in species, as the rate of production of conidia differs from species to species. However, in this study a random count of 300 conidia in foam from two streams was compared to assess qualitative and quantitative differences.

Water filtration usually results in a smaller number of species as well as conidia, depending on the availability of substrates in the streams (leaf and woody litter) (Sridhar & Kaveriappa, 1984, 1989; Sridhar et al., 2013). Foam represents the maximum number of conidia, as they were trapped by air bubbles. Earlier studies in coastal and Western Ghat streams also represented the highest number of species in foam samples (Sridhar et al., 1992). Such species and conidial abundance in foam samples are probably due to surface runoff and tree canopy washdown. Aquatic hyphomycetes were reported from throughfall, stemflow, tree holes, and damp leaf litter on the ground (Karamchand & Sridhar, 2008; Sridhar et al., 2013, 2020; Magyar et al., 2021; Sharathchandra & Sridhar, 2022). Such surface runoff during the rainy season is one of the potential reasons for heavy conidial deposition in foam. Such enormous conidia have been considered carriers of essential nutrients across the aquatic food web (Seena et al., 2022).



3.3 Colonized Fungi

Naturally submerged leaf litter on bubble chamber incubation showed colonization of 7 and 15 species in Konaje and Kallimaru streams, respectively (Table 4; Fig. 4). However, conidia per gram of dry leaf litter was higher in Konaje than Kallimaru stream (1001 vs. 678). Possibly, the rate of production of conidia differs from species to species; that is reflected in Table 4. In fact, extent colonization of substrates may also be dependent on the conidial richness of a specific species in water. *Anguillospora longissima*, *Flagellospora curvula*, *Lunulospora curvula*, and *Synnematophora constricta* were common in two streams among the five top species. Such dominance leads to competition with each other to exploit the available leaf litter. The number of species found in leaf litter in the Konaje stream is comparable to the earlier studies (Sridhar & Kaveriappa, 1984, 1989; Sridhar et al., 2013). The availability of leaf litter and surface area is higher than the woody litter in these streams, which is a possible reason for the higher species richness and conidial output.

Table 2. Conidia of aquatic hyphomycetes occurring in water samples (*, n=3, mean conidia/100 ml) and their relative abundance (RA) in two coastal streams

Konaje stream	Conidia/ 100 ml*	RA%	Kallimaru stream	Conidia/ 100 ml	RA%
<i>Lunulospora curvula</i>	18.3	39.6	<i>Helicomyces colligatus</i>	19.6	37.3
<i>Flagellospora curvula</i>	11.0	23.8	<i>Lunulospora cymbiformis</i>	15.0	28.6
<i>Anguillospora longissima</i>	7.7	16.7	<i>Anguillospora longissima</i>	7.3	13.9
<i>Flagellospora penicillioides</i>	5.3	11.5	<i>Lunulospora curvula</i>	4.9	9.3
<i>Lunulospora cymbiformis</i>	1.0	2.1	<i>Synnematophora constricta</i>	2.0	3.8
<i>Triscelophorus acuminatus</i>	1.0	2.1	<i>Triscelophorus monosporus</i>	1.7	3.2
<i>Helicomyces colligatus</i>	0.7	1.5	<i>Anguillospora crassa</i>	0.7	1.3
<i>Cylindrocarpon</i> sp.	0.3	0.7	<i>Wiesenriomyces laurinus</i>	0.7	1.3
<i>Phalangispora constricta</i>	0.3	0.7	<i>Alatospora acuminata</i>	0.3	0.8
<i>Tricladium</i> sp.	0.3	0.7	<i>Triscelophorus acuminatus</i>	0.3	0.8
<i>Triscelophorus monosporus</i>	0.3	0.7			
Species richness	11		Species richness	10	
Conidial richness	46.2		Conidial richness	52.5	

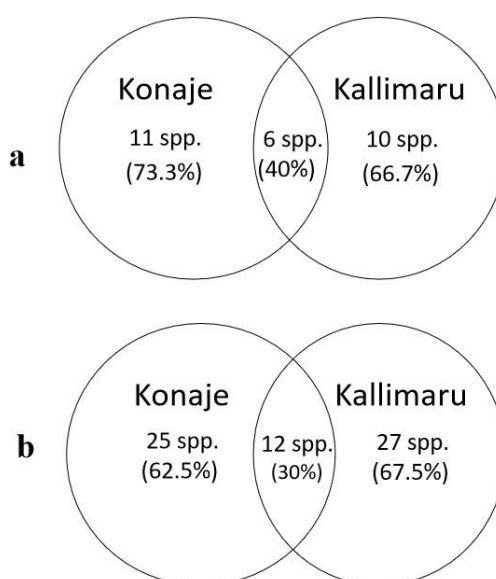


Fig. 3 - Venn diagram of species richness of aquatic hyphomycetes in water (a) and foam (b) samples in two coastal streams



Table 3. Conidia of aquatic hyphomycetes occurring in foam samples (*, among 300 conidia) and their relative abundance (RA) in two coastal streams

Konaje stream	Number of conidia*	RA%	Kallimaru stream	Number of conidia*	RA%
<i>Triscelophorus acuminatus</i>	102	34.0	<i>Lunulospora curvula</i>	33	11.0
<i>Anguillospora longissima</i>	77	25.6	<i>Flagellospora curvula</i>	27	9.0
<i>Lunulospora curvula</i>	35	11.7	<i>Anguillospora longissima</i>	23	7.7
<i>Helicomyces colligatus</i>	18	6.0	<i>Helicoma atroseptatum</i>	21	7.0
<i>Helicoma atroseptatum</i>	10	3.3	<i>Triscelophorus acuminatus</i>	20	6.7
<i>Triscelophorus monosporus</i>	9	3.0	<i>Anguillospora crassa</i>	18	6.0
<i>Anguillospora crassa</i>	5	0.7	<i>Helicomyces hyderabadensis</i>	18	6.0
<i>Campylospora chaetoclada</i>	5	1.7	<i>Triscelophorus monosporus</i>	18	6.0
<i>Condylospora spumigena</i>	5	1.7	<i>Flabellospora verticillata</i>	16	5.3
<i>Flabellospora crassa</i>	5	1.7	<i>Tricladium splendens</i>	15	5.0
<i>Helicosporium</i> sp.	5	1.7	<i>Isthmotricladia gombakiensis</i>	13	4.3
<i>Condylospora vietnamensis</i>	4	1.3	<i>Alatospora acuminata</i>	11	3.7
<i>Flabellospora</i> sp.	3	1.0	<i>Synnematophora constricta</i>	10	3.3
<i>Flagellospora penicillioides</i>	2	0.7	<i>Triscelophorus konajensis</i>	10	3.3
<i>Isthmotricladia gombakiensis</i>	2	0.7	<i>Wiesenriomyces laurinus</i>	10	3.3
<i>Tetracladium marchalianum</i>	2	0.7	<i>Varicosporium elodeae</i>	9	3.0
<i>Triscelophorus konajensis</i>	2	0.7	<i>Condylospora spumigena</i>	8	2.7
<i>Wiesenriomyces laurinus</i>	2	0.7	<i>Retarius</i> sp.	6	2.0
<i>Clavaria aquatica</i>	1	0.3	<i>Speiropsis pedatospora</i>	3	1.0
<i>Clavospora tentacula</i>	1	0.3	<i>Campylospora leptosoma</i>	2	0.7
<i>Cylindrocarpon</i> sp.	1	0.3	<i>Clavospora tentacula</i>	2	0.7
<i>Campylospora</i> sp. 1	1	0.3	<i>Helicosporium aureum</i>	2	0.7
<i>Lunulospora cymbiformis</i>	1	0.3	<i>Condylospora vietnamensis</i>	1	0.3
<i>Phalangispora constricta</i>	1	0.3	<i>Dwayaangam cornuta</i>	1	0.3
<i>Speiropsis pedatospora</i>	1	0.3	<i>Flabellospora multiradiata</i>	1	0.3
			<i>Helicoma</i> sp.	1	0.3
			<i>Hydrometrospora symmetrica</i>	1	0.3
Species richness	25		Species richness	27	

Table 4 Conidia of aquatic hyphomycetes released from leaf litter on bubble chamber incubation (*, n=3, mean conidia/g dry mass) and their relative abundance (RA) in two coastal streams

Konaje stream	Conidia/g*	RA%	Kallimaru stream	Conidia/g*	RA%
<i>Lunulospora curvula</i>	385	38.5	<i>Lunulospora curvula</i>	165	24.3
<i>Anguillospora longissima</i>	168	16.8	<i>Flagellospora curvula</i>	159	23.5
<i>Flagellospora curvula</i>	136	13.6	<i>Anguillospora longissima</i>	108	15.9
<i>Condylospora vietnamensis</i>	101	10.1	<i>Synnematophora constricta</i>	69	10.2
<i>Synnematophora constricta</i>	84	8.4	<i>Triscelophorus acuminatus</i>	55	8.1
<i>Triscelophorus monosporus</i>	76	7.6	<i>Setosynnema isthmosporem</i>	37	5.5
<i>Flagellospora penicillioides</i>	51	5.1	<i>Flagellospora penicillioides</i>	21	3.1
			<i>Anguillospora crassa</i>	20	3.0
			<i>Triscelophorus konajensis</i>	16	2.4
			<i>Triscelophorus monosporus</i>	9	3.0
			<i>Lunulospora cymbiformis</i>	7	1.0
			<i>Tripospermum myrti</i>	7	1.0
			<i>Cylindrocarpon</i> sp.	3	0.4
			<i>Clavospora tentacula</i>	1	0.2
			<i>Condylospora spumigena</i>	1	0.2
Species richness	7		Species richness	15	
Conidial richness	1001		Conidial richness	678	

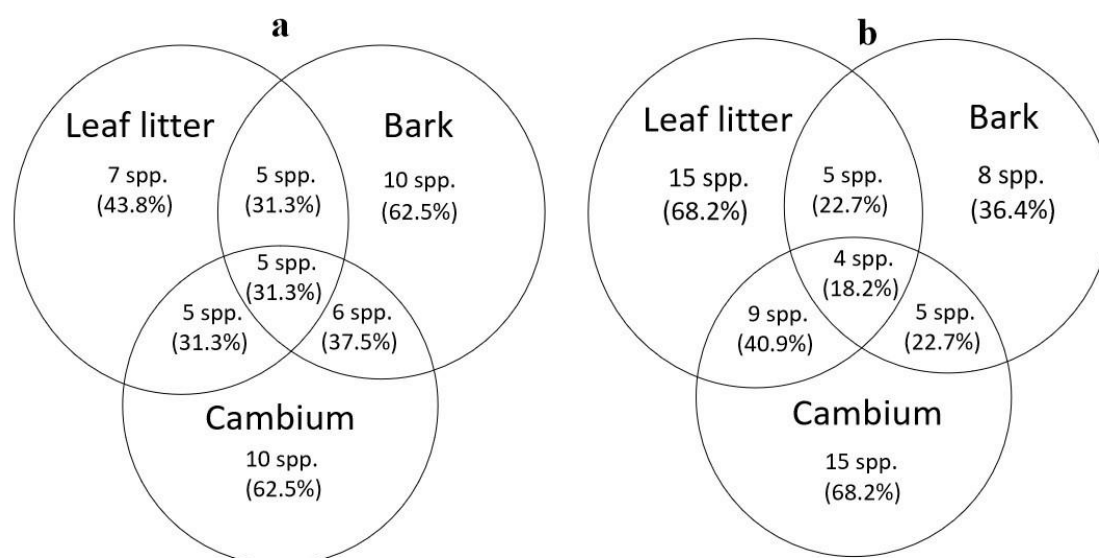


Fig. 4 - Venn diagram of the species richness of aquatic hyphomycetes in leaf litter, bark, and cambium samples in Konaje stream (a) and Kallimaru stream (b)

The bark of naturally submerged woody litter in bubble chamber incubation showed colonization of 10 and 8 species in Konaje and Kallimaru streams, respectively (Table 5; Fig. 4). Conidial release per gram dry mass of bark was higher in Konaje than in Kallimaru stream (172 vs. 150). *Anguillospora longissima*, *Cylindrocarpon* sp., *Flabelliospora crassa*, and *Flagellospora curvula* were common in two streams among the five top species. The epidermis and periderm of bark consist of cell-wall macromolecules like cutin, lignin, and suberin, which have protective functions (Lewis et al., 1987; Bernards, 2002; Graça, 2015). Suberin is a complex biopolymer that is lipophilic and composed of long chain fatty acids (glycerol and suberin acids). Such biopolymers may be inhibitory to aquatic hyphomycetes or cause low colonization, which needs further understanding.

The cambium part of the natural submerged woody litter in bubble chamber incubation showed colonization of 10 and 15 species in Konaje and Kallimaru streams, respectively (Table 6; Fig. 4). Release of conidia per gram dry mass of cambium was higher in Kallimaru than Konaje stream (65 vs. 30). *Anguillospora crassa*, *Flagellospora curvula*, and *Lunulospora curvula* were common among the five top species in two streams. Aquatic hyphomycetes are capable of colonizing woody litter in aquatic as well as semi-aquatic conditions (Sridhar & Sudheep, 2011; Sudheep & Sridhar, 2011, 2013; Sharathchandra & Sridhar, 2020). According to Bärlocher (2009), aquatic hyphomycetes will not completely exhaust their mycelial biomass through the release of conidia; instead, a small amount of mycelial biomass will be retained in the substrates to produce the teleomorph stage. The woody litter has been considered an ideal, long-lasting ecological niche to induce the perfect state of aquatic hyphomycetes (Webster, 1992; Sivichai & Jones, 2003; Hu et al., 2014), and such recent studies are lacking.

3.4 Species Richness and Diversity

A wide variety of aquatic hyphomycetes were represented in water, foam, leaf litter, bark, and cambium in Konaje and Kallimaru streams. Among the species represented in Figs. 5 and 6, *Campylospora leptosoma*, *Clavariana aquatica*, *Condylospora vietnamensis*, *Helicomycetes colligatus*, *H. hyderabadensis*, *Helicosporium aureum*, *Lunulospora cymbiformis*, and *Synnematophora constricta* are rare and not reported previously (Sridhar and Kaveriappa 1982,



1984, 1989; Sridhar et al. 2013). The overlapping species on substrates in Konaje stream ranged between 5 and 6 species, while it was between 4 and 9 species in Kallimaru stream (Fig. 4).

In both streams, Shannon diversity was higher than Simpson diversity in water as well as foam samples (Fig. 7). The Pielou's equitability in water samples was not significantly different between the streams, while it was significantly higher in the Kallimaru stream. An earlier study also showed the occurrence of more species in foam, followed by leaf litter and water samples in the Konaje stream (Sridhar & Kaveriappa, 1982, 1984, 1989). Foam represents a higher number of species richness owing to surface runoff from aquatic, semi-aquatic, and terrestrial niches in catchments of the streams. Simpson diversity in leaf litter, bark, and cambium in streams was similar to that in water and foam samples (Fig. 7). Shannon diversity was significantly higher in leaf litter and cambium of Kallimaru stream, while in bark of Konaje stream (Fig. 7).

Table 5 Conidia of aquatic hyphomycetes released from bark on bubble chamber incubation (*, n=3, mean conidia/g dry mass) and their relative abundance (RA) in two coastal streams

Konaje stream	Conidia/g*	RA%	Kallimaru stream	Conidia/g*	RA%
<i>Flabellospora crassa</i>	43	25.0	<i>Lunulospora curvula</i>	41	27.3
<i>Cylindrocarpon</i> sp.	26	15.1	<i>Flabellospora crassa</i>	29	19.3
<i>Triscelophorus monosporus</i>	26	15.1	<i>Cylindrocarpon</i> sp.	26	17.3
<i>Flagellospora curvula</i>	22	12.8	<i>Flagellospora curvula</i>	24	16.7
<i>Anguillospora longissima</i>	16	9.3	<i>Anguillospora longissima</i>	20	13.3
<i>Triscelophorus acuminatus</i>	13	7.6	<i>Clavariana aquatica</i>	7	4.7
<i>Lunulospora curvula</i>	11	6.4	<i>Clavatospora tentacula</i>	2	1.3
<i>Triscelophorus konajensis</i>	9	5.2	<i>Triscelophorus konajensis</i>	1	0.7
<i>Alatospora acuminata</i>	3	1.7			
<i>Flagellospora penicillioides</i>	3	1.7			
Species richness	10		Species richness	8	
Conidial richness	172		Conidial richness	150	

Table 6 Conidia of aquatic hyphomycetes released from cambium on bubble chamber incubation (*, n=3, mean conidia/g dry mass) and their relative abundance (RA) in two coastal streams

Konaje stream	Conidia/g*	RA%	Kallimaru stream	Conidia/g*	RA%
<i>Anguillospora longissima</i>	10	33.8	<i>Anguillospora crassa</i>	21	32.5
<i>Lunulospora curvula</i>	6	20.3	<i>Lunulospora curvula</i>	18	27.8
<i>Anguillospora crassa</i>	5	16.9	<i>Triscelophorus acuminatus</i>	8	12.4
<i>Flagellospora curvula</i>	5	16.9	<i>Flagellospora curvula</i>	5	7.7
<i>Flagellospora penicillioides</i>	1	3.4	<i>Triscelophorus konajensis</i>	3	4.6
<i>Wiesenriomyces laurinus</i>	1	3.4	<i>Flabellospora crassa</i>	2	3.1
<i>Alatospora acuminata</i>	1	3.4	<i>Anguillospora longissima</i>	2	3.1
<i>Condylospora spumigena</i>	0.3	1.0	<i>Campylospora</i> sp. 2	1	1.6
<i>Mycocentrospora acerina</i>	0.2	0.7	<i>Lunulospora cymbiformis</i>	1	1.6
<i>Triscelophorus monosporus</i>	0.1	0.3	<i>Flagellospora penicillioides</i>	1	1.6
			<i>Mycocentrospora acerina</i>	1	1.6
			<i>Alatospora acuminata</i>	1	1.6
			<i>Condylospora spumigena</i>	0.3	0.5
			<i>Ingoldiella hamata</i>	0.2	0.3
			<i>Wiesenriomyces laurinus</i>	0.2	0.3
Species richness/g dry mass	10		Species richness/g dry mass	15	
Conidial richness/g dry mass	29.6		Conidial richness/g dry mass	64.7	

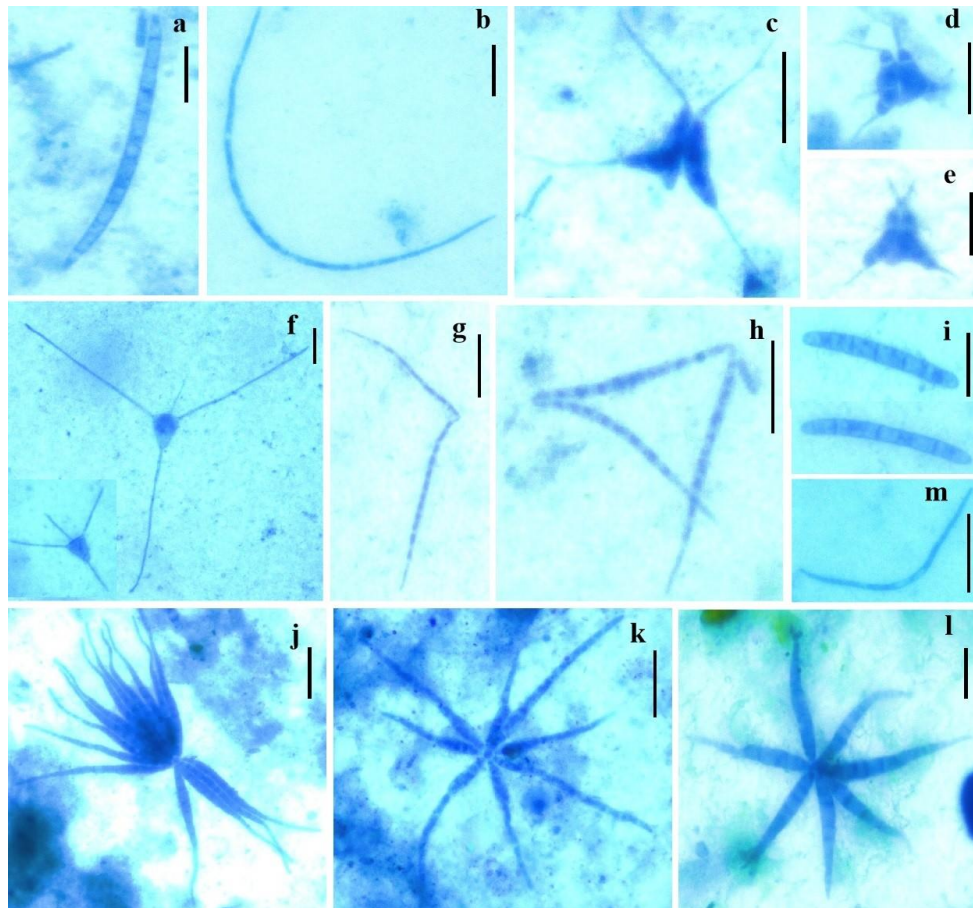


Fig. 5 - Conidia of *Anguillospora crassa* (a), *A. longissima* (b), *Campylospora leptosoma* (c), *Campylospora* sp. 1 (d), *Campylospora* sp. 2 (e), *Clavariana aquatica* (f), *Condyllospora spumigena* (g), *C. vietnamensis* (h), *Cylindrocarpon* sp. (i), *Flabellospora multiradiata* (j), *F. verticillata* (k), *Flabellospora* sp. (l), and *Flagellospora curvula* (m) (scale bar: 20 μ m)

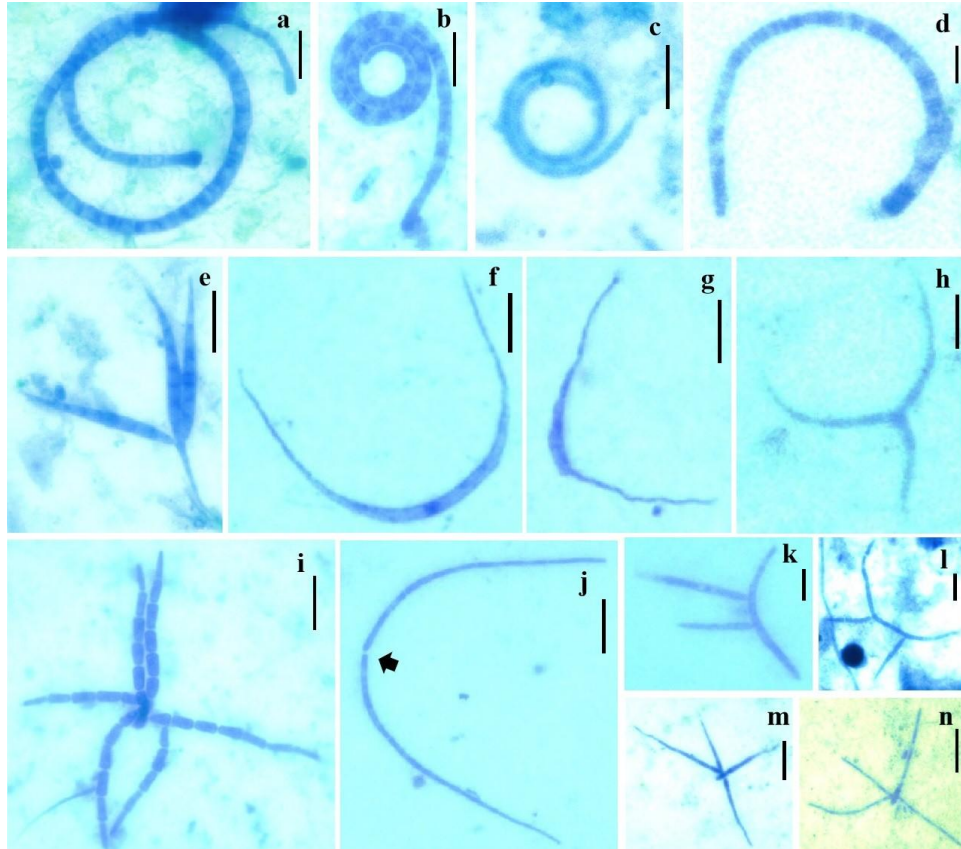


Fig. 6 - Conidia of *Helicomyces colligatus* (a), *H. hyderabadensis* (b), *Helicosporium aureum* (c), *Helicosporium* sp. (d), *Isthmotricladia gombakiensis* (e), *Lunulospora curvula* (f), *L. cymbiformis* (g), *Retiarius* sp. (h), *Speiopsis pedatospora* (i), *Synnematophora constricta* (arrow, deep constriction) (j), *Tricladium splendens* (k), *Tricladium* sp. (l), *Triscelophorus acuminatus* (m), and *T. monosporus* (n) (scale bar: 20 μ m)

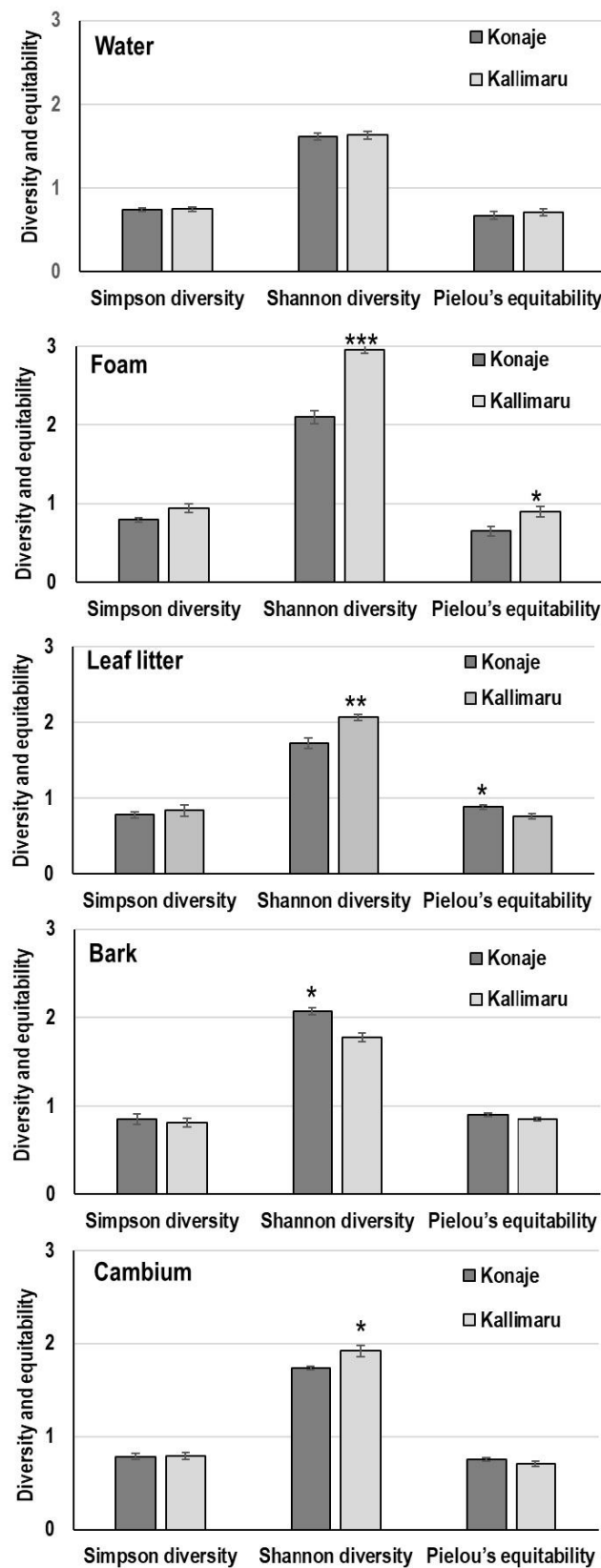


Fig. 7 - Diversity and equitability of aquatic hyphomycetes in water, foam, leaf litter, bark, and cambium samples in two coastal streams (n=3, mean±SD: *, p<0.05; **, p<0.01; ***, p<0.001)



4. Conclusions

Among the aquatic hyphomycetes reported in this study, nine species are new records to the Indian subcontinent (*Campylospora leptosoma*, *Clavariana aquatica*, *Condylospora vietnamensis*, *Helicoma atroseptatum*, *Helicomycetes colligatus*, *H. hyderabadensis*, *Helicosporium aureum*, *Hydrometraspora symmetrica*, and *Setosynnema isthmosporem*). Usually, *Clavatospora tentacula*, *Condylospora spumigena*, *Flabellospora crassa*, *F. multiradiata*, *F. verticillata*, *Isthmotricladia gombakiensis*, *Lunulospora cymbiformis*, *Synnematophora constricta*, and *Varicosporium elodeae* occur in the cold and high altitudinal aquatic niches of the Western Ghats streams. Surprisingly, these species were also found in the coastal streams studied, possibly due to the physicochemical characteristics of the streams.

Water filtration and foam assessment will provide a quick and authentic assessment of species richness and diversity in a specific stream, which could be compared with other freshwater bodies for assessment, representing the landscape of the catchment area. Conidia of aquatic hyphomycetes are transported from the canopy to the tree floors (soil, leaf, wood, bark, and others), and they seem to have several functions, especially decomposition and disseminating their propagules to other habitats (forest floors and streamlets).

Bubble chamber incubation of different substrates represents the colonization ability of aquatic hyphomycetes. Species richness and conidial output by bark were limited compared to leaf litter and cambium, while conidial output in leaf litter was the highest. A comparison of colonized aquatic hyphomycetes in leaf and woody litter is necessary to understand the dynamics of aquatic hyphomycetes in aquatic and semi-aquatic ecosystems. As aquatic hyphomycetes occur in wide geographic ranges and ecological niches, their contributions to organic matter decomposition, biogeochemical cycles, and animal nutrition need further investigation.

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Conflict of Interest

There is no conflict of interest.

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