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# Species diversity of freshwater hyphomycetes in some streams of Pakistan. Comparison of sampling techniques*

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**Freshwater hyphomycete communities in four streams were sampled by filtration of water samples, trapping conidia in artificial foam, and examination of alder leaf pack baits and random sampling of naturally occurring submerged leaves. No two communities of freshwater hyphomycetes detected by different sampling techniques used singly in a stream showed 100% similarity. Based on relative frequency values, the same 10 top-ranking species in a stream were detected by artificial foam trap and on the baited leaves. These species differed in ranking, and the community in artificial foam was richer than on baited leaves. Of the various communities detected by different sampling techniques used singly or in combination, those detected by artificial foam trap or a combination of this technique with leaf baiting or with leaf baiting and randomly sampled leaves or filtration showed the highest similarity to the total community based on data generated using four sampling techniques simultaneously.**

Key Words——freshwater hyphomycetes; species diversity; streams.

A number of studies have dealt with the ecology of freshwater hyphymycetes based on a single sampling technique. Seasonal periodicity of these fungi has been reported by examining the composition of conidia in river water (Iqbal, 1992; Iqbal and Webster, 1973b, 1977; Bärlocher and Rosset, 1981; Thomas et al., 1991) or in foam (Bärlocher, 1986; Chauvet, 1991; Gönczol, 1971, 1975, 1987; Iqbal and Bhatty, 1979). Several other studies have concentrated on freshwater hyphomycete communities occurring on submerged substrata such as naturally colonized submerged leaves picked up randomly (Iqbal et al., 1979; Chamier et al., 1984) and introduction of leaf packs to stream experimentally to study the colonizing freshwater hyphomycete communities (Bärlocher and Kendrick, 1974; Suberkropp, 1984; Suberkropp and Klug, 1976; Gönczol, 1989; Bärlocher, 1990; Chamier and Dixon, 1982).

Foam spora based on the examination of foam accumulated on the surface of water below rapids shows no correlation with the species composition of freshwater hyphomycetes in stream water (Iqbal and Webster, 1973a; Shearer and Webster, 1985c). The age of the foam and the origin of some of the conidia in it create difficulties in interpretation. Evidently the conidia may survive without germinating for weeks to months (Iqbal and Webster, 1973a), which tends to blur the changes occurring in an actively growing and reproducing fungal population (Bärlocher, 1992). An alternative to the use of natural foam is to trap conidia of these fungi in artificial foam. Artificial foam traps conidia efficiently and within a few minutes the foam is saturated with species present in the stream (Iqbal, 1993).

Examination of randomly sampled naturally colonized submerged leaves falls short of detecting the true community of freshwater hyphomycetes. Submerged leaf litter differs in age and quality at the time of sampling due to the continuous addition of deciduous leaves to temperate streams coupled with marked differences in breakdown rates among leaf species. Seasonal differences in species composition of freshwater hyphomycete communities are thus due to fungal succession as a result of changing substrate quality (Gessner et al., 1993). Leaf pack baiting provides a standard homogeneous known substratum which can be manipulated with respect to time and location (Shearer and Webster, 1985a). However, decay and successional speed vary with season (Suberkropp, 1984), so different successional stages can still be recorded even if identical exposure times are chosen (Gessner et al., 1993). Standardized leaf packs were thus not only exposed at different times of the year, but also retrieved at repeated intervals.

In the present study, water filtration (Iqbal and Webster, 1973b), examination of naturally colonized submerged leaves (Iqbal and Bhatty, 1979), the leaf pack baiting technique of Shearer and Webster (1985a, b, c), and trapping of conidia in artificial foam (Iqbal, 1993) were used simultaneously to work out the species diversity of freshwater hyphomycetes in four mountain streams. Freshwater hyphomycete communities based on data generated by these four techniques used simultaneously and communities detected by each sampling technique alone and in various combinations were compared to find

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the most reliable technique or combination of techniques to detect the "true" fungal community.

## Materials and Methods

**Description of Streams** Of the four streams studied, the sampling sites on the New Stream (grid reference SX 467978) and the Khanspur Stream (grid reference SX 464992) are situated in the Khanspur area (Murree Hills), and those on the Jabori Canal (grid reference SX 270681) and the Sanatorium Stream (grid reference SX 272627) are situated in the Dadar area, Siran Valley. For details of the profile, see Iqbal (1992, 1993).

The Jabori Canal is geographically very close to the Sanatorium Stream and has a similar water chemistry, except that more nitrates and phosphates find their way into the Jabori Canal when it passes through agricultural fields. This canal has a richer deciduous vegetation than the other streams. Some agricultural products such as wheat straw, rice straw and leaves of other crops find their way into the canal. The Khanspur Stream and the New Stream are also close to each other geographically and have similar riparian vegetation. Mostly they are sheltered with coniferous trees like *Pinus excelsa* Wall non Lamb., *Abies pindrow* Royle and a few trees of *Quercus dilatata* Lindl. ex Royle. Herbaceous and shrubby plants are very common on the banks.

Conductivity, pH, water temperature, nitrates and phosphates at the time of sampling are given in Table 1.

**Collection Techniques** Samples were collected once a month throughout the year (1986-1987) at intervals of 4 weeks (Jabori Canal, Sanatorium Stream) and 5 weeks (New Stream, Khanspur Stream). Four techniques were used to characterize the freshwater hyphomycete communities.

**1. Membrane filtration** Stream water was filtered to ascertain the relative contribution of each freshwater hyphomycete species to the conidial pool. Conidia filtered from 1 L of stream water were identified and counted according to the procedures of Iqbal and Webster (1973b).

**2. Leaf pack baiting** Leaf packs of *Alnus glutinosa* (L.) Gaertn., consisting of autumn-shed, air dried leaves with no visible fungal invasion, were placed in nylon nets with a mesh size of 1.5 mm. The leaves in packs were arranged in a single layer so as not to cover each other. All leaves used in this study were collected from a single alder tree (*A. glutinosa*) from Jhelum Valley (Kashmir). Branches were gently shaken and shed leaves were collected. Leaves were dried at room tepmerature for 7 days. Alder leaves were selected because they are readily colonized by a wide variety of freshwater hyphomycetes. Standardized packs of ten leaves were placed in streams every month and retrieved after exposure for four weeks (Jabori Canal and Sanatorium Stream) and about five weeks (New Stream and Khanspur Stream) throughout the year. Leaves were rinsed thoroughly in stream water. Each leaf was washed well with distilled water and cut up into ten discs of 1 $cm^2$. To remove conidia of their origin than the disc itself, these discs were forcibly aerated for 1/2 h. The discs were taken out, washed again and aerated singly at 20°C in MacCortney bottles each containing 15 ml distilled water. After 24 h, the resulting spore suspension was passed through an 8-$\mu$m pore size filter. Production of conidia has been as an estimate of species presence. To confirm the identities of conidia difficult to identify on the filter, disks incubated in distilled water in Petri dishes were also examined simultaneously with the filtering of the water/foam samples. Results are expressed either as percentage frequency, which is disc frequency expressed as a percentage, or as relative mean disc frequency (or relative frequency), which is the percentage of leaf discs per leaf pack on which a given species was recorded (Shearer and Webster, 1985a). Relative frenquencies were calculated by dividing the mean disc frequency of a species by

Table 1. Physico-chemical data of the four strems.

| Streams | The New Stream | The Jabori Canal | The Sanitorium Stream | The Khanspur Stream |
|---|---|---|---|---|
| Temperature (°C): | | | | |
| Range | 3-18.0 | 6-16 | 9-18 | 1-14 |
| Average | 9.9 | 11.2 | 11.2 | 8.0 |
| pH: | | | | |
| Range | 5.5-6.8 | 5.9-6.9 | 6.0-6.8 | 5.5-6.5 |
| Average | 6.3 | 6.3 | 6.4 | 6.1 |
| EC: | | | | |
| $\mu$S/20°C | 65-105 | 95-140 | 90-130 | 70-110 |
| Average | 95 | 120 | 105 | 90 |
| $NO_3$ mg/L: | | | | |
| Range | 3-7 | 15-25 | 5-11 | 2-8 |
| Average | 5.4 | 20.0 | 8.5 | 6.2 |
| $PO_4$ mg/L: | | | | |
| Range | 0.05-0.2 | 0.6-8.5 | 0.1-3.5 | 0.04-0.3 |
| Average | 0.1 | 6.6 | 1.85 | 0.15 |

the sum of the mean disc frequency values of all species foun in the sample.

**3. Random leaf sampling** Naturally occurring submerged leaves were collected randomly from the stream beds. These leaves were processed by the procedure described above for leaf packs.

**4. Trapping of conidia in artificial foam** An artificial foam trap was developed below rapids by adding detergent to the stream water. The amount of stream water passing through this foam trap in 5 min was about 500 L. Foam thus formed below the rapids was collected into a beaker. The foam was allowed to settle and later it was filtered through a membrane of 8-$\mu$m pore size. The filter was fixed with FAA, dried and stained with 0.1% cotton blue in lactic acid. The filters were processed according to the procedures of Iqbal and Webster (1973b). The conidia were identified and counted. Results were reported as relative frequencies or percentage frequencies as in the filtration method.

To compare the freshwater hyphomycete communities detected by the four sampling techniques, Sorensen's index of similarity was calculated (Mueller-Dombois and Ellenberg, 1974). This index is defined as the number of common species in the two communities divided by the average number of species in the two communities.

An index of diversity in freshwater hyphomycete communities was calculated by the Shannon-Weiner function $H = -\sum_{i=1}^{S} (P_i)(\mathrm{Log}_2 P_i)$, where H = index of species diversity, S = number of species and $P_i$ = proportion of total sample belonging to $i$th species (Krebs, 1978).

## Results

### Freshwater hyphomycete communities detected by differ-

Table 2. Colonization, number, relative frequencies (Rf) on leaves, mean number $L^{-1}$ and relative frequency of conidia in stream water and in artificial foam for the ten top-ranked species of freshwater hyphomycetes in the Jabori Canal detected by four sampling techniques used singly and simultaneously.

| 10 October, 1986<br>Randomly collected leaves (R)<br>No. of species:26 | | | 9 Novcember, 1986<br>Baited leaves (B)<br>25 | | | 9 November, 1986<br>Randomly collected leaves (R)<br>29 | | | 9 November, 1986<br>ABRF<br>39<br>H diversity: −2.8995421 | |
|---|---|---|---|---|---|---|---|---|---|---|
| Species | *f* (%) | *Rf* | Species | *f* (%) | *Rf* | Species | *f* (%) | *Rf* | Species | *Rf* |
| *Alatospora acuminata* | 54.44 | 0.121 | *F. curvula* | 61.10 | 0.122 | *A. acuminata* | 48.88 | 0.117 | *F. curvula* | 0.593 |
| *Flagellospora curvula* | 44.44 | 0.099 | *A. acuminata* | 55.55 | 0.111 | *F. curvula* | 47.77 | 0.114 | *A. acuminata* | 0.363 |
| *Tetracladium marchalianum* | 41.10 | 0.091 | *A. longissima* | 41.11 | 0.082 | *A. longissima* | 35.55 | 0.085 | *A. longissima* | 0.312 |
| *Lemonniera aquatica* | 38.88 | 0.086 | *C. aquatica* | 41.11 | 0.082 | *T. chaetocladium* | 34.44 | 0.083 | *A. tetracladia* | 0.267 |
| *Anguillospora longissima* | 37.77 | 0.184 | *A. tetracladia* | 37.77 | 0.076 | *C. aquatica* | 27.77 | 0.066 | *C. atuatica* | 0.263 |
| *Clavariopsis aquatica* | 33.33 | 0.074 | *T. marchalianum* | 31.11 | 0.062 | *G. inflata* | 26.66 | 0.064 | *T. marchalianum* | 0.254 |
| *Articulospora tetracladia* | 32.22 | 0.071 | *T. chaetocladium* | 28.88 | 0.058 | *L. aquatica* | 24.44 | 0.058 | *T. chaetocladium* | 0.251 |
| *Tricladium chaetocladium* | 21.11 | 0.047 | *L. aquatica* | 27.77 | 0.055 | *A. tetracladia* | 22.22 | 0.053 | *L. aquatica* | 0.225 |
| *Triscelophorus monosporus* | 21.11 | 0.037 | *C. longibrachiata* | 22.22 | 0.044 | *T. marchalianum* | 21.11 | 0.050 | *G. inflata* | 0.189 |
| *Lunulospora curvula* | 18.88 | 0.042 | *G. inflata* | 17.77 | 0.035 | *C. longibrachiata* | 17.77 | 0.042 | *C. longibrachiata* | 0.163 |

| October | | | | | November | | | | |
|---|---|---|---|---|---|---|---|---|---|
| Filtration (F)<br>No. of species: 26<br>Total No. of conidia $L^{-1}$: 2680<br>H diversity: −1.2212 | | | Artificial foam (A)<br>31<br><br>−1.308 | | Filtration (F)<br>27<br>3640<br>−0.992 | | | Artificial foam (A)<br>31<br><br>−1.46 | |
| Species | Mean No. of conidia | *Rf* (%) | Species | *Rf* | Species | Mean No. of conidia | *Rf* | Species | *Rf* |
| *F. curvula* | 512 | 0.191 | *T. marchalianum* | 0.105 | *F. curvula* | 1030 | 0.283 | *T. marchalianum* | 0.096 |
| *H. lugdunensis* | 254 | 0.095 | *L. aquatica* | 0.101 | *A. longissima* | 298 | 0.082 | *A. acuminata* | 0.094 |
| *L. curvula* | 230 | 0.086 | *A. acuminata* | 0.097 | *C. aquatica* | 178 | 0.049 | *A. tetracladia* | 0.092 |
| *A. longissima* | 222 | 0.083 | *T. chaetocladium* | 0.097 | *A. tetracladia* | 167 | 0.046 | *L. aquatica* | 0.080 |
| *T. chaetocladium* | 131 | 0.049 | *A. tetracladia* | 0.083 | *T. marchalianum* | 167 | 0.046 | *T. chaetocladium* | 0.075 |
| *L. aquatica* | 115 | 0.043 | *G. inflata* | 0.081 | *H. lugdunensisH.* | 127 | 0.035 | *F. curvula* | 0.074 |
| *T. marchalianum* | 115 | 0.043 | *A. longissima* | 0.069 | *T. chaetocladium* | 127 | 0.035 | *C. aquatica* | 0.066 |
| *C. aquatica* | 112 | 0.042 | *F. curvula* | 0.057 | *L. aquatica* | 116 | 0.032 | *A. longissima* | 0.063 |
| *A. acuminata* | 104 | 0.039 | *C. aquatica* | 0.046 | *M. aquatica* | 109 | 0.030 | *G. inflata* | 0.061 |
| *A. tetracladia* | 102 | 0.038 | *C. longibrachiata* | 0.033 | *G. inflata* | 106 | 0.029 | *C. longibrachiata* | 0.052 |

ABRF: Community based on data generated by four sampling techniques used simultaneously, A: trapping conidia in artificial foam, B: baited leaves, R: randomly sampled leaves, F: filtration method.

**ent sampling techniques used singly at the submersion (October) and retrieval (November) of baited leaves** Freshwater hyphomycete communities detected by different sampling techniques used singly at the submersion and retrieval of baited leaves for the months of October and November are described (Tables 2-5) to compare the effectiveness of individual sampling techniques. This period coincides with the peak of leaf litter deposition in the autumn, resulting in the build up of a concentration of conidia in temperate streams.

Communities detected on randomly sampled 'R' leaves in October differed from those in November in the rankings and species composition. Species on 'R' leaves were more numerous in November than in October except in the New Stream, which had 16 species in October and 15 species in November (Tables 2-5). *Triscelophorus monosporus* Ingold and *Lunulospora curvula* Ingold were present in the 10 top-ranking species in October and absent in November in the Jabori Canal (Table 2). *Clavatospora longibrachiata* (Ingold) Nilsson ex Marvanová & Nilsson and *Geniculospora inflata* (Ingold) Nilsson ex Marvanová & Nilsson were absent in October and present in November in the Jabori Canal (Table 2). *Lemonniera terrestris* Tubaki was present in October and absent in November. Similarly, *Heliscus lugdunensis* Sacc. & Therry was absent in October and present in November in the Khanspur Stream (Table 3). *Flagellospora curvula* Ingold was the dominant species in October and was replaced by *Alatospora acuminata* Ingold in November in the Khanspur Stream (Table 3). *Anguillospora longissima* (de Wild.) Ingold was not in the 10 top-ranking species in October, but it replaced *Clavariopsis aquatica* de Wild. in the top-ranking species in November in the Sanatorium Stream (Table 4). *Anguillospora longissima* was present in the 10 top-ranking species in October and was replaced by *Tricladium chaetocladium*

Table 3. Colonization, number, relative frequencies (Rf) on leaves, mean number $L^{-1}$ and relative frequency of conidia in stream water and in artificial foam for the ten top-ranked species of freshwater hyphomycetes in the Khanspur Stream detected by four sampling techniques used singly and simultaneously.

| 12 October, 1986 Randomly collected leaves (R) No. of species: 22 | | | 12 November, 1986 Baited leaves (B) 21 | | | 12 November, 1986 Randomly collected leaves (R) 25 | | | 12 November, 1986 ABRF 35 H diversity: −2.8841483 | |
|---|---|---|---|---|---|---|---|---|---|---|
| Species | *f*(%) | *Rf* | Species | *f*(%) | *Rf* | Species | *f*(%) | *Rf* | Species | *Rf* |
| *Flagellospora curvula* | 54.44 | 0.131 | *F. curvula* | 57.77 | 0.139 | *A. acuminata* | 43.33 | 0.130 | *F. curvula* | 0.523 |
| *Alatospora acuminata* | 52.22 | 0.125 | *A. acuminata* | 51.11 | 0.123 | *F. curvula* | 41.11 | 0.124 | *A. acuminata* | 0.363 |
| *Tetracladium marchalianum* | 42.22 | 0.101 | *T. marchalianum* | 41.11 | 0.101 | *C. aquatica* | 32.22 | 0.097 | *C. aquatica* | 0.309 |
| *Anguillospora longissima* | 41.11 | 0.099 | *C. aquatica* | 38.88 | 0.094 | *T. marchalianum* | 31.11 | 0.094 | *T. marchalianum* | 0.296 |
| *Clavatosppora longibrachiata* | 34.44 | 0.083 | *A. tetracladia* | 34.44 | 0.083 | *A. tetracladia* | 22.22 | 0.067 | *A. longissima* | 0.279 |
| *Articulospora tetracladia* | 31.11 | 0.075 | *A. longissima* | 32.22 | 0.077 | *L. aquatica* | 22.22 | 0.067 | *A. tetracladia* | 0.231 |
| *Clavariopsis aquatica* | 27.77 | 0.067 | *L. aquatica* | 25.55 | 0.062 | *C. longibrachiata* | 20.00 | 0.060 | *L. aquatica* | 0.215 |
| *Lamonniera terrestris* | 25.55 | 0.061 | *T. chaetocladium* | 21.11 | 0.050 | *A. longissima* | 18.88 | 0.057 | *T. chaetocladium* | 0.207 |
| *Lemonniera aquatica* | 18.88 | 0.045 | *C. longibrachiata* | 20.00 | 0.048 | *T. chaetocladium* | 14.44 | 0.043 | *H. lugdunensis* | 0.201 |
| *Tricladium chaetocladium* | 17.77 | 0.042 | *H. lugdunensis* | 14.44 | 0.029 | *H. lugdunensis* | 12.22 | 0.036 | *C. longibrachiata* | 0.195 |

| October | | | | | November | | | | |
|---|---|---|---|---|---|---|---|---|---|
| Filtration (F) No. of species: 22 Total No. of conidia $L^{-1}$: 1490 | | | Artificial foam (A) 27 | | Filtration (F) 24 1780 | | | Artificial foam (A) 27 | |
| Species | Mean No. of conidia | *Rf* | Species | *Rf* | Species | Mean No. of conidia | *Rf* | Species | *Rf* |
| *L. curvula* | 372.5 | 0.250 | *A. longissima* | 0.116 | *F. curvula* | 341.8 | 0.192 | *T. chaetocladium* | 0.083 |
| *H. lugdunensis* | 236.9 | 0.159 | *T. chaetocladium* | 0.096 | *L. curvula* | 257.5 | 0.144 | *A. acuminata* | 0.074 |
| *F. curvula* | 205.6 | 0.138 | *T. marchalianum* | 0.090 | *H. lugdunensis* | 190.5 | 0.106 | *T. marchalianum* | 0.072 |
| *A. longissima* | 165.4 | 0.111 | *A. tetracladia* | 0.086 | *A. longissima* | 137.1 | 0.077 | *C. aquatica* | 0.071 |
| *L. aquatica* | 53.2 | 0.035 | *L. aquatica* | 0.082 | *C. longibrachiata* | 97.5 | 0.054 | *A. longissima* | 0.068 |
| *A. acuminata* | 47.7 | 0.032 | *A. acuminata* | 0.078 | *C. aquatica* | 85.4 | 0.048 | *F. curvula* | 0.063 |
| *T. marchalianum* | 46.2 | 0.031 | *C. longibrachiata* | 0.057 | *L. aquatica* | 70.6 | 0.040 | *G. inflata* | 0.060 |
| *A. tetracladia* | 44.7 | 0.030 | *C. aquatica* | 0.052 | *A. acuminata* | 64.1 | 0.036 | *A. tetracladia* | 0.052 |
| *C. aquatica* | 43.2 | 0.029 | *F. curvula* | 0.049 | *T. chaetocladium* | 61.0 | 0.034 | *C. longibrachiata* | 0.052 |
| *L. terrestris* | 34.0 | 0.022 | *H. lugdunensis* | 0.033 | *T. marchalianum* | 59.6 | 0.029 | *L. aquatica* | 0.048 |
| | | | | | *A. tetracladia* | 59.6 | 0.029 | | |

ABRF: Community based on data generated by four sampling techniques used simultaneously, A: trapping conidia in artificial foam, B: baited leaves, R: randomly sampled leaves, F: filtration method.

Table 4. Colonization, number, relative frequencies (Rf) on leaves, mean number $L^{-1}$ and relative frequency of conidia in stream water and in artificial foam for the ten top-ranked species of freshwater hyphomycetes in the Sanatorium Stream detected by four sampling techniques used singly.

| 10 October, 1986<br>Randomly collected leaves (R)<br>No. of species 18 | | | 9 November, 1986<br>Baited leaves (B)<br>16 | | | 9 November, 1986<br>Randomly collected leaves (R)<br>19 | | | 9 November, 1986<br>ABRF<br>30<br>H diversity: −2.2847881 | |
|---|---|---|---|---|---|---|---|---|---|---|
| Species | *f* (%) | *Rf* | Species | *f* (%) | *Rf* | Species | *f* (%) | *Rf* | Species | *Rf* |
| *Flagellospora curvula* | 46.66 | 0.1016 | *F. curvula* | 54.44 | 0.1060 | *F. curvula* | 50.00 | 0.110 | *F. curvula* | 0.620 |
| *Clavatospora longibrachiata* | 44.44 | 0.0968 | *G. inflata* | 50.00 | 0.0974 | *G. inflata* | 46.66 | 0.1026 | *C. longibrachiata* | 0.336 |
| *Alatospora acuminata* | 43.33 | 0.0944 | *A. acuminata* | 48.88 | 0.0952 | *A. tetracladia* | 40.00 | 0.0880 | *A. longissima* | 0.314 |
| *Lemonniera aquatica* | 40.00 | 0.0871 | *C. acuminata* | 46.66 | 0.0909 | *C. longibrachiata* | 38.88 | 0.0855 | *G. inflata* | 0.309 |
| *Articulospora tetracladia* | 38.88 | 0.0847 | *A. tetracladia* | 44.44 | 0.0865 | *L. aquatica* | 37.77 | 0.0831 | *T. chaetocladium* | 0.305 |
| *Geniculospora inflata* | 34.44 | 0.0750 | *L. aquatica* | 40.00 | 0.0779 | *T. chaetocladium* | 36.66 | 0.0806 | *A. tetracladia* | 0.302 |
| *Triscelophorus monosporus* | 31.10 | 0.0678 | *T. chaetocladium* | 37.77 | 0.0735 | *T. marchalianum* | 28.88 | 0.0635 | *T. marchalianum* | 0.260 |
| *Tetracladium marchalianum* | 30.00 | 0.0653 | *T. marchalianum* | 34.44 | 0.0671 | *A. acuminata* | 28.88 | 0.0635 | *L. aquatica* | 0.257 |
| *Tricladium chaetocladium* | 28.88 | 0.0629 | *A. longissima* | 31.10 | 0.0606 | *T. monosporus* | 27.77 | 0.0611 | *A. acuminata* | 0.241 |
| *Clavariopsis aquatica* | 22.22 | 0.0484 | *T. monosporus* | 30.00 | 0.0584 | *A. longissima* | 26.66 | 0.0586 | *T. monosporus* | 0.201 |

| October | | | | | November | | | | |
|---|---|---|---|---|---|---|---|---|---|
| Filtration (F)<br>No. of species: 15<br>Total No. of conidia $L^{-1}$: 1130 | | | Artificial foam (A)<br>21 | | Filtration (F)<br>16<br>1400 | | | Artificial foam (A)<br>24 | |
| Species | Mean No. of conidia | *Rf* | Species | *Rf* | Species | Mean No. of conidia | *Rf* | Species | *Rf* |
| *F. curvula* | 308.49 | 0.273 | *A. tetracladia* | 0.119 | *F. curvula* | 453.6 | 0.324 | *C. longibrachiata* | 0.098 |
| *H. lugdunensis* | 144.64 | 0.128 | *A. acuminata* | 0.114 | *H. lugdunensis* | 175.0 | 0.125 | *T. chaetocladium* | 0.096 |
| *A. longissima* | 129.95 | 0.115 | *C. longibrachata* | 0.096 | *A. longissima* | 159.6 | 0.114 | *A. tetracladia* | 0.090 |
| *L. curvula* | 103.96 | 0.092 | *A. longissima* | 0.082 | *C. longibrachiata* | 86.8 | 0.062 | *T. marchalianum* | 0.088 |
| *C. longibrachiata* | 63.28 | 0.056 | *G. inflata* | 0.076 | *T. chaetocladium* | 77.0 | 0.055 | *A. longissima* | 0.081 |
| *A. acuminata* | 53.11 | 0.047 | *F. curvula* | 0.075 | *T. marchalianum* | 58.8 | 0.042 | *F. curvula* | 0.080 |
| *A. tetracladia* | 50.85 | 0.045 | *T. marchalianum* | 0.072 | *A. tetracladia* | 53.2 | 0.038 | *G. inflata* | 0.078 |
| *T. monosporus* | 47.46 | 0.042 | *T. monosporus* | 0.066 | *L. aquatica* | 47.6 | 0.034 | *L. aquatica* | 0.062 |
| *T. marchalianum* | 42.94 | 0.038 | *L. aquatica* | 0.064 | *G. inflata* | 43.4 | 0.031 | *C. aquatica* | 0.050 |
| *G. inflata* | 35.03 | 0.031 | *T. chaetocladium* | 0.044 | *C. aquatica* | 42.0 | 0.030 | *T. monosporus* | 0.044 |

ABRF: Community based on data generated by four sampling techniques used simultaneously, A: trapping conidia in artificial foam, B: baited leaves, R: randomly sampled leaves, F: filtration method.

Ingold in November in the New Stream (Table 5).

Freshwater hyphomycete communities detected by filtration in stream water (F) had greater numbers of species accompanied with greater numbers of conidia in November than in October in all the streams studied (Tables 2–5). *Flagellospora curvula* dominated these communities except the one in October in the Khanspur Stream, which was dominated by *Lunulospora curvula* (Table 3). *Heliscus lugdunensis* was the second most common species in most of these communities (Tables 2–5). *Lunulospora curvula* occurred more frequently in the 10 top-ranking species in communities in October than in November in these streams. *Lemonniera terrestris* and *T. monosporus* were present in the 10 top-ranking species in October in the Khanspur Stream (Table 3) and the Sanatorium Stream (Table 4). *Geniculospora inflata* occurred in the top-ranking species less frequently. *Clavatospora longibrachiata* was present in the 10 top-ranking species more frequently in November than in October. *Tricladium chaetocladium* was absent from the 10 top-ranking species in October but present in the 10 top-ranking species in Nobember in these streams except in the New Stream (Table 5). *Flagellospora curvula, H. lugdunensis, L. curvula* and *A. longissima* were present abundantly in communities detected in stream water.

Freshwater hyphomycete communities detected in the artificial foam (A) at the time of submersion (October) and retrieval (November) of baited leaves in four streams showed minor differences in species composition and in the ranking of species. *Heliscus lugdunensis* was present in October and absent in November in the 10 top-ranking species in the Khanspur Stream (Table 3). *Geniculospora inflata* replaced this species in November (Table 3). *Tricladium chaetocladium* was absent in October and present in November in the 10 top-ranking species in the New Stream (Table 5). *Tetracladium marchalianum, Articulospora tetracladia, T. chaetocladium, Clavatospora longibrachiata, Clavariopsis aquatica* and

Table 5. Colonization, number, relative frequencies (Rf) on leaves, mean number $L^{-1}$ and relative frequency of conidia in stream water and in artificial foam for the ten top-ranked species of freshwater hyphomycetes in the New Stream detected by four sampling techniques used singly.

| 12 October, 1986 Randomly collected leaves (R) No. of species 16 | | | 12 November, 1986 Baited leaves (B) 13 | | | 12 November, 1986 Randomly collected leaves (R) 15 | | | 12 November, 1986 ABRF 23 H diversity: −2.3299992 | |
|---|---|---|---|---|---|---|---|---|---|---|
| Species | *f* (%) | *Rf* | Species | *f* (%) | *Rf* | Species | *f* (%) | *Rf* | Species | *Rf* |
| *Flagellospora curvula* | 54.44 | 0.1428 | *F. curvula* | 52.22 | 0.1278 | *F. curvula* | 40.00 | 0.1124 | *F. curvula* | 0.5922 |
| *Clavatospora longibrachiata* | 43.33 | 0.1137 | *A. tetracladia* | 47.77 | 0.1169 | *A. acuminata* | 38.88 | 0.1092 | *H. lugdunensis* | 0.4081 |
| *Alatospora acuminata* | 41.10 | 0.1078 | *A. acuminata* | 43.33 | 0.1060 | *A. tetracladia* | 36.66 | 0.1030 | *A. tetracladia* | 0.3509 |
| *Lemonniera aquatica* | 38.88 | 0.1020 | *C. longiburachiata* | 41.10 | 0.1006 | *C. longibrachiata* | 30.00 | 0.0843 | *C. longibrachiata* | 0.3459 |
| *Articulospora tetracladia* | 32.22 | 0.0845 | *C. aquatica* | 37.77 | 0.0924 | *C. aquatica* | 29.26 | 0.0822 | *A. acuminata* | 0.3322 |
| *Clavariopsis aquatica* | 31.10 | 0.0816 | *L. aquatica* | 36.66 | 0.0897 | *H. lugdunensis* | 28.88 | 0.0811 | *C. aquatica* | 0.3306 |
| *Tetracladium marchalianum* | 23.33 | 0.0612 | *H. lugdunensis* | 31.10 | 0.0761 | *L. aquatica* | 26.66 | 0.0749 | *A. longissima* | 0.3203 |
| *Heliscus lugdunensis* | 20.00 | 0.0524 | *G. inflata* | 29.26 | 0.0716 | *T. marchalianum* | 26.66 | 0.0749 | *L. aquatica* | 0.2866 |
| *Anguillospora longissima* | 18.88 | 0.0495 | *T. marchalianum* | 28.88 | 0.0688 | *T. chaetocladium* | 22.22 | 0.0624 | *T. marchalianum* | 0.2717 |
| *Geniculospora inflata* | 17.77 | 0.0466 | *T. chaetocladium* | 26.66 | 0.0652 | *C. inflata* | 22.22 | 0.0624 | *T. chaetocladium* | 0.2216 |

| October | | | | | November | | | | |
|---|---|---|---|---|---|---|---|---|---|
| Filtration (F) No. of species: 14 Total No. of conidia $L^{-1}$: 920 | | | Artificial foam (A) 21 | | Filtration (F) 15 1200 | | | Artificial foam (A) 20 | |
| Species | Mean No. of conidia | *Rf* | Species | *Rf* | Species | Mean No. of conidia | *Rf* | Species | *Rf* |
| *F. curvula* | 239.2 | 0.26 | *A. tetracladia* | 0.106 | *F. curvula* | 303.6 | 0.253 | *C. aquatica* | 0.101 |
| *H. lugdunensis* | 166.5 | 0.181 | *C. longibrachata* | 0.104 | *H. lugdunensis* | 240.0 | 0.200 | *A. tetracladia* | 0.100 |
| *A. longissima* | 124.2 | 0.135 | *L. aquatica* | 0.101 | *A. longissima* | 153.6 | 0.128 | *F. curvula* | 0.099 |
| *L. curvula* | 73.6 | 0.080 | *F. curvula* | 0.096 | *C. longibrachiata* | 84.0 | 0.070 | *C. longibrachiata* | 0.091 |
| *C. longibrachiata* | 69.0 | 0.075 | *C. aquatica* | 0.096 | *C. aquatica* | 66.0 | 0.055 | *T. marchalianum* | 0.087 |
| *C. aquatica* | 61.6 | 0.067 | *T. marchalianum* | 0.094 | *T. giganteum* | 52.8 | 0.044 | *L. aquatica* | 0.086 |
| *L. aquatica* | 40.5 | 0.044 | *A. longissima* | 0.087 | *T. marchalianum* | 49.2 | 0.041 | *A. acuminata* | 0.081 |
| *A. tetracladia* | 38.6 | 0.042 | *A. acuminata* | 0.081 | *L. aquatica* | 43.2 | 0.036 | *A. longissima* | 0.079 |
| *T. marchalianum* | 37.7 | 0.041 | *H. lugdunensis* | 0.065 | *A. acuminata* | 43.2 | 0.036 | *T. chaetocladium* | 0.055 |
| *A. acuminata* | 23.0 | 0.025 | *G. inflata* | 0.033 | *A. tetracladia* | 37.2 | 0.031 | *H. lugdunensis* | 0.051 |

ABRF: Community based on data generated by four sampling techniques used simultaneously, A: trapping conidia in artificial foam, B: baited leaves, R: randomly sampled leaves, F: filtration method.

*A. longissima* occurred abundantly in communities detected in artificial foam. More species were detected in the artificial foam trap than on filters.

A greater number of species colonized baited leaves (B) in the Jabori Canal (Table 2) than in other streams. However, slightly more species occurred on the randomly sampled 'R' leaves than on baited 'B' leaves in each stream (Tables 2–5). *Flagellospora curvula* dominated freshwater hyphomycete communities occurring on baited leaves in all the streams studied. *Alatospora acuminata* was the second most common species in the Jabori Canal (Table 2) and the Khanspur Stream (Table 3). *Geniculospora inflata* was the second most common species in the Sanatorium Stream (Table 4). *Articulospora tetracladia* was the second most common species on the baited 'B' leaves submerged in the New Stream (Table 5). These communities detected on baited 'B' leaves in four streams differed in species composition and in the rankings of species. *Heliscus lugdunensis* was present in the 10 top-ranking species in the Khanspur Stream (Table 3) and the New Stream (Table 5). *Triscelophorus monosporus* was present in the 10 top-ranking species in the Sanatorium Stream (Table 4). *Geniculospora inflata* was present in the 10 top-ranking species in the Jabori Canal (Table 2) and Sanatorium Stream (Table 4). *Flagellospora curvula, Alatospora acuminata, Clavatospora longibrachiata, Tetracladium marchalianum, Tricladium chaetocladium, Articulospora tetracladia, Anguillospora longissima* and *Lemonniera aquatica* de Wild. were common species in these communities detected on the baited 'B' leaves submerged in these four streams.

Freshwater hyphomycete communities based on data generated by using different sampling techniques simultaneously (ABRF) in November were richer than those detected by different techniques used singly in each of the four streams studied (Tables, 2–5). *Flagellospora curvula* was the dominant species and *Alatospora acuminata* was the second most common spe-

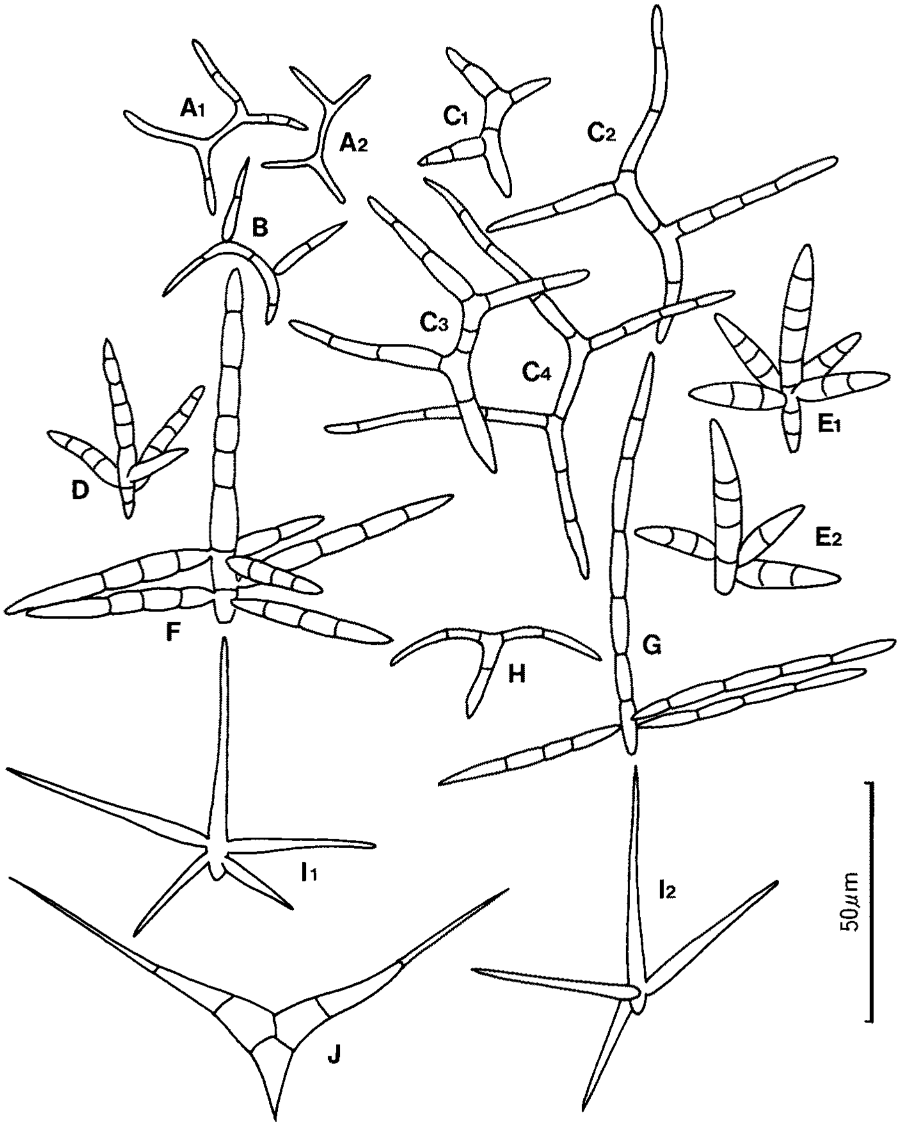


Fig. 1. A: *Tricladium* sp. 1 from foam. B: *Tricladium* sp. 2 from foam. C: *Tricladium* sp. 3 from foam. D: *Triscelophorus* sp. 1 from foam. E: *Triscelophorus* sp. 2 from foam. F: *Dendrospora* sp. 1 G: *Dendrospora* sp. 2 (*D. fustosa*) H: Unknown I: *Triscelophorus* sp. 3 from foam. J: Unknown sp. 3.

cies in the Jabori Canal (Table 2) and the Khanspur Stream (Table 3). *Clavatospora longibrachiata* and *Heliscus lugdunensis* were the second dominant species in the Sanatorium Stream and in the New Stream, respectively (Tables 4, 5). *Flagellospora curvula, Clavatospora longibrachiata, Auguillospora longissima, Articulospora tetracladia, Lemonniera aquatica, Tricladium chaetocladium, Tetracladium marchalianum* and *Alatospora acuminata* formed the nucleus of the 10 top-ranking species in the four communities detected in these streams. In addition, *Clavariopsis aquatica* and *Geniculospora inflata* in the Jabori Canal (Table 2), *Clavariopsis aquatica* and *Heliscus lugdunensis* in the Khanspur Stream, the New Stream (Tables 3, 5) and *Geniculospora inflata* and *Triscelophorus monosporus* in the Sanatorium Stream (Table 4) formed the 10 top-ranked species. These four communities differed from each other in their rankings and species composition. The freshwater hyphomycete community in the Jabori Canal showed higher species diversity (H index$=-2.899542$) than in the other streams (Tables 3, 4, 5).

**Comparison of techniques** Freshwaer hyphomycete species collected from each stream once a month for a year (1986-87) with each sampling method are given in Table 6. Conidia of unidentifiable species of freshwater hyphomycetes are illustrated in Figs. 1 and 2. Freshwater hyphomycete communities based on the data generated (from samples taken once a month for a year) by the four sampling techniques used simultaneously (ABRF) had 50 species in the New Stream, 52 in the Sanatorium Stream, 55 in the Khanspur Stream and 74 in the Jabori Canal (Table 6). Several species, e.g., *Bacillispora aquatica* Nilsson, *Bacillispora inflata* Iqbal & Bhatty, *Dimorphospora foliicola* Tubaki, *Flagellospora minuta*

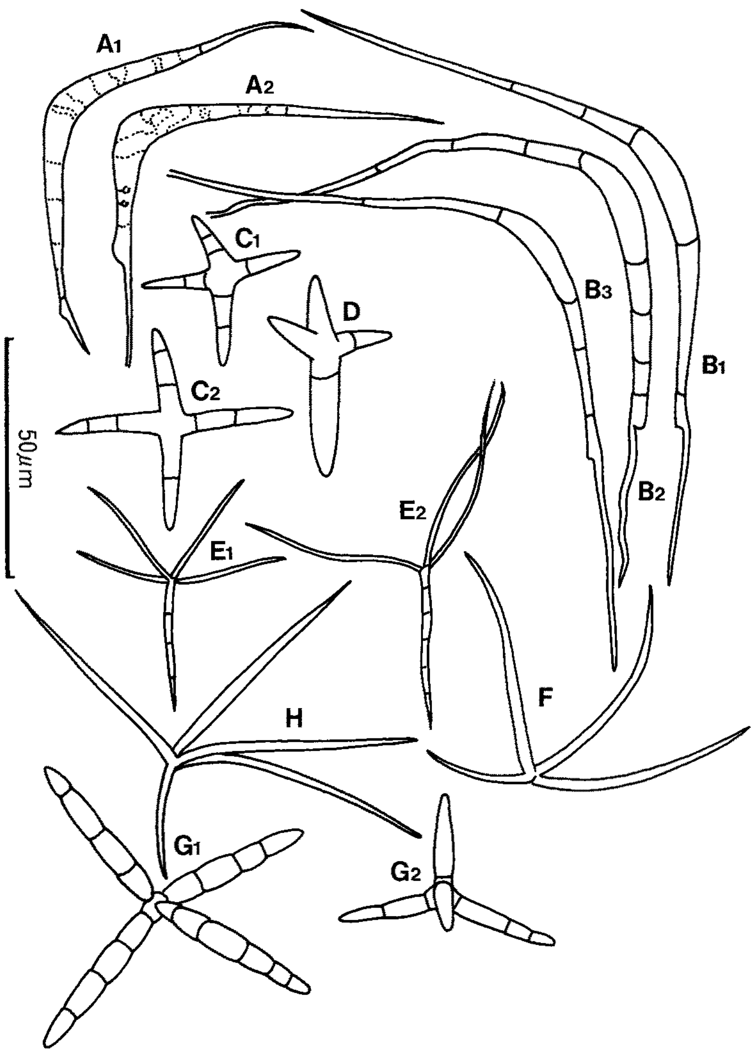


Fig. 2. A: *Mycocentrospora* sp. 1 from foam. B: *Mycocentrospora* sp. 2 from foam. C: Tetraradiate conidia possibly of *Lemonniera* sp. from foam. D: An unknown species of *Heliscus* from filters. E: *Clavariopsis* sp. from foam. F: Unknown tetraradiate conidia from foam. G: Unknown tetraradiate conidia (possibly unknown species of *Lemonniera*).

Iqbal & Bhatty, *Flegellospora stricta* Nilsson and *Margaritispora aquatica* Ingold were never detected by the filtration method. *Actinospora megalospora* Ingold, *Articulospora angulata* Tubaki, *Campylospora chaetocladia* Ranzoni, *Culicidospora aquatica* Petersen, *Dendrospora fusca* Descals & Webster, *Dwayaangam cornuta* Descals, *Flabellospora acuminata* Descals, *F. verticillata* Alasoadura, *Gyoerffyella speciosa* (Miura) Dudka, *Heliscella stellata* (Ingold & Cox) Marvanová, *Tetracladium furcatum* Descals and a few unknown species, were detected by filtration and/or were trapped in artificial foam (Table 6). Several species, e.g., *Alatospora pulchella, Anguillospora gigantea, Clavariana aquatica* Nawawi, *Pleuropedium tricladioides* Marvanová & Iqbal and *Tricladium eccentricum* Petersen, were found on naturally colonized submerged leaves and not on the baited leaves.

Freshwater hyphomycete communities detected by different sampling techniques used sighly and in various combinations were compared with the "true" community based on data generated using the four sampling techniques simultaneously (ABRF) (Table 7). The index of similarity in species composition between randomly (R) sampled leaves and the "true" community varied from 64.86 to 83.14%. Similarity index between communities detected on baited (B) leaves and the "true" community in these streams varied from 59.15 to 81.72%. communities detected on filters and the "true" community showed indices of similarity varying from 70.12 to 76.68%. The index of similarity between the "true" community and that detected in the artificial (A) foam concentrate was highest in each stream. Indices of similarity in these communities varied from 88.88% in the New Stream to 94.28% in the Jabori Canal (Table 7).

Communities based on data generated by a combination of artificial foam trap and baited leaves (AB) showed similarity index of 95.83% with the "true" community in the New Stream, 98.03% in the Sanatorium Stream and

Table 6. Species of freshwater hyphomycetes in four mountain streams detected using four different sampling techniques.

| Species | The New Stream | | | | The Jabori Canal | | | | The Sanatorium Stream | | | | The Khanspur Stream | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| *Actinospora megalospora* Ingold | – | – | – | – | A | – | – | F | – | – | – | – | – | – | – | – |
| *Alatospora acuminata* Ingold | A | B | R | F | A | B | R | F | A | B | R | F | A | B | R | F |
| *A. constricta* Dyko | A | B | R | – | A | B | – | F | A | B | R | F | A | B | R | F |
| *A. pulchella* Marvanová | A | – | – | – | A | – | R | F | – | – | – | – | A | – | R | F |
| *Anguillospora crassa* Ingold | A | – | – | F | A | B | R | F | A | – | R | F | A | B | R | F |
| *A. curvula* Iqbal | – | B | – | – | – | – | – | – | – | – | – | – | A | – | – | F |
| *A. gigantea* Ranzoni | – | – | R | – | A | – | – | F | A | – | R | F | – | – | – | – |
| *A. longissima* (Sacc. & Syd.) Ingold | A | B | R | F | A | B | R | F | A | B | R | F | A | B | R | F |
| *Articulospora angulata* Tubaki | – | – | – | – | A | – | – | – | – | – | – | – | – | – | – | – |
| *A. proliferata* Jooste, Roldan & Merwe | – | – | – | – | A | B | R | F | – | – | – | – | – | – | – | – |
| *A. tetracladia* Ingold | A | B | R | F | A | B | R | F | A | B | R | F | A | B | R | F |
| *Bacillispora aquatica* Nilson | – | B | R | – | – | B | R | – | – | B | R | – | – | B | R | – |
| *B. inflata* Iqbal & Bhatty | – | B | R | – | – | B | R | – | – | B | R | – | – | B | R | – |
| *Campylospora chaetocladia* Ranzoni | A | – | – | F | A | – | – | F | A | – | – | F | A | – | – | F |
| *Clavariana aquatica* Nawawi | A | – | – | F | A | – | R | F | A | – | R | F | – | – | – | – |
| *Clavariopsis aquatica* de Wildeman | A | B | R | F | A | B | R | F | A | B | R | F | A | B | R | F |
| *C. azlanii* Nawawi | – | – | – | – | A | B | R | F | – | – | – | – | – | – | – | – |
| *Clavariopsis* sp. (2E) | – | – | – | – | A | – | – | – | – | – | – | – | – | – | – | – |
| *Clavatospora longibrachiata* (Ingold) Nilsson ex Marvanová & Nilsson | A | B | R | F | A | B | R | F | A | B | R | F | A | B | R | F |
| *Culicidospora aquatica* Petersen | A | – | – | F | A | – | – | F | A | – | – | F | A | – | – | – |
| *C. gravida* Petersen | – | – | – | – | A | – | – | – | – | – | – | – | – | – | – | – |
| *Dactylella aquatica* (Ingold) Ranzoni | A | B | R | F | A | B | R | F | A | B | R | F | A | B | R | F |
| *Dendrospora fusca*? (IG) | – | – | – | – | A | – | – | – | – | – | – | – | – | – | – | – |
| *Dendrospora* sp. (IF) | – | – | – | – | A | – | – | – | A | – | – | – | – | – | – | – |
| *Dimorphospora filiicola* Tubaki | – | B | – | – | – | B | R | – | – | B | R | – | – | B | R | – |
| *Dwayaangam cornuta* (Descals) Descals & Webster | – | – | – | – | A | – | – | F | – | – | – | – | – | – | – | – |
| *Flabellospora acuminata* (Descals) Descals & Webster | – | – | – | – | – | – | – | – | A | – | – | – | – | – | – | – |
| *F. verticillata* Alasoadura | – | – | – | – | A | – | – | F | A | – | – | – | – | – | – | – |
| *Flagellospora curvula* Ingold | A | B | R | F | A | B | R | F | A | B | R | F | A | B | R | F |
| *F. fusarioides* Iqbal | A | B | R | F | A | B | R | F | – | B | R | – | A | B | R | F |
| *F. minuta* Iqbal & Bhatty | – | B | R | – | – | – | – | – | – | – | – | – | – | B | – | – |
| *F. penicillioides* Ingold | – | – | R | – | – | B | R | – | – | – | R | – | A | B | R | F |
| *F. stricta* Nilson | – | – | – | – | – | B | R | – | – | – | – | – | – | – | – | – |
| *Geniculospora inflata* (Nilsen) Nilsen ex Marvanová & Nilsen | A | – | – | F | A | B | R | F | A | – | – | F | A | – | R | F |
| *Gyoerffyella speciosa* (Miura) Dudka | – | – | – | F | – | – | – | – | – | – | – | – | A | – | – | – |
| *Heliscella stellata* (Ingold & Cod) Marvanová | – | – | – | – | A | – | – | F | – | – | – | F | A | – | – | – |
| *Heliscus lugdunensis* Sacc. & Therry | A | B | R | F | A | B | R | F | A | B | R | F | A | B | R | F |
| *Heliscus* sp. (2D) | – | – | – | F | – | – | – | – | – | – | – | – | – | – | – | – |
| *Latrimulosa uniinflata* Matsushima | – | – | – | – | A | – | – | F | – | – | – | – | A | – | – | – |
| *Lemonniera aquatica* de Wildeman | A | B | R | F | A | B | R | F | A | B | R | F | A | B | R | F |
| *L. centrosphaera* Marvanová | A | – | – | F | A | B | – | F | A | B | R | F | A | B | R | – |
| *L. cornuta* Ranzoni | A | – | R | – | A | – | R | F | A | B | R | F | A | B | R | F |
| *L. filiformis* Petersen ex Dyko | A | – | – | F | A | B | – | F | A | B | R | F | A | B | R | – |
| *L. terrestris* Tubaki | A | – | R | F | A | B | R | F | A | B | R | F | A | B | R | F |
| *Lemonniera* sp. I (2C) | – | – | – | – | A | – | – | – | – | – | – | – | – | – | – | – |
| *Lemonniera* sp. II (2G) | A | – | – | – | A | – | – | – | – | – | – | – | – | – | – | – |
| *Lunulospora curvula* Ingold | A | B | R | F | A | B | R | F | A | B | R | F | A | B | R | F |
| *Margaritispora aquatica* Ingold | – | B | R | – | – | B | – | – | – | B | R | – | – | B | R | – |

| Species | The New Stream | | | | The Jabori Canal | | | | The Sanatorium Stream | | | | The Khanspur Stream | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| *Mycocentrospora acerina* Deighton | A | B | R | F | A | – | R | F | A | B | R | F | A | – | R | F |
| *M. clavata* Iqbal | A | – | – | – | A | B | R | F | A | B | R | F | A | B | R | F |
| *Mycocentrospora* sp. 1 (2A) | – | – | – | – | A | – | – | – | A | – | – | – | – | – | – | – |
| *Mycocentrospora* sp. 2 (2B) | – | – | – | – | A | – | – | – | A | – | – | – | – | – | – | – |
| *Pleuropedium tricladioides* Marvanová & Iqbal | A | – | – | – | A | – | R | F | A | – | R | – | A | – | R | – |
| *Scorpiosporium angulatum* (Ingold) Iqbal | A | – | – | F | A | – | – | F | – | – | – | – | A | B | – | – |
| *S. minutum* Iqbal | – | – | – | – | A | – | R | F | – | – | – | – | – | B | R | – |
| *Tetrachaetum elegans* Ingold | A | – | – | F | A | – | R | F | A | – | R | F | A | B | R | – |
| *Tetracladium furcatum* (Descals) Descals & Webster | – | – | – | – | A | – | – | – | – | – | – | – | – | – | – | – |
| *T. marchalianum* de Wildeman | A | B | R | F | A | B | R | F | A | B | R | F | A | B | R | F |
| *T. maxilliforme* (Rostrup) Ingold | – | – | – | – | A | – | R | F | A | B | R | – | A | – | – | – |
| *T. setigerum* (Grove) Ingold | A | – | – | F | A | – | R | F | A | B | R | – | A | – | R | F |
| *Tricellula aquatica* Webster | A | – | – | – | A | – | R | – | A | – | R | – | A | B | – | F |
| *Tricladium attenuatum* Iqbal | A | – | – | – | A | B | R | F | – | B | R | – | A | B | R | F |
| *T. chaetocladium* Ingold | A | – | – | F | A | B | R | F | A | B | R | F | A | B | R | F |
| *T. eccentricum* Petersen | A | – | – | – | A | – | R | F | A | – | – | – | A | – | – | F |
| *T. giganteum* Iqbal | A | – | – | F | A | – | R | F | A | – | – | F | A | B | R | F |
| *T. splendens* Ingold | A | – | – | – | A | B | R | F | A | – | R | – | A | B | R | F |
| *Tricladium* sp. 1 (IA) | A | – | – | – | A | – | – | – | – | – | – | – | – | – | – | – |
| *Tricladium* sp. 2 (IB) | – | – | – | – | A | – | – | – | – | – | – | – | A | – | – | – |
| *Tricladium* sp. 3 (IC) | – | – | – | – | A | – | – | – | A | – | – | – | – | – | – | – |
| *Tripospermum mytri* (Lind.) Hughes | – | – | – | – | A | – | R | – | – | – | – | – | – | B | – | – |
| *Triscelophorus monosporus* Ingold | A | B | R | – | A | B | R | F | A | B | R | F | A | B | R | F |
| *Triscelophorus* sp. 1 (ID) | – | – | – | – | A | – | – | – | – | – | – | – | A | – | – | – |
| *Triscelophorus* sp. 2 (IE) | – | – | – | – | A | – | – | – | A | – | – | – | A | – | – | – |
| *Triscelophorus* sp. 3 (II) | – | – | – | – | A | – | – | – | – | – | – | – | A | – | – | – |
| Unknown sp. 1 (IH) | – | – | – | – | A | – | – | – | – | – | – | – | – | – | – | – |
| Unknown sp. 2 (IJ) | – | – | – | – | – | – | – | – | A | – | – | – | – | – | – | – |
| Unknown sp. (2F) | – | – | – | – | A | – | – | – | A | – | – | – | – | – | – | – |
| Unknown sp. (2H) | – | – | – | – | A | – | – | – | – | – | – | – | – | – | – | – |
| *Varicosporium delicatum* Iqbal | – | – | – | – | – | – | – | – | – | – | – | – | – | B | – | – |
| *V. elodeae* Kegel | A | – | – | – | A | B | – | F | A | – | R | F | A | B | R | – |
| *Volucrispora graminea* (Ingold) McDougal & Denn. | A | – | – | – | A | – | R | F | – | – | – | – | A | B | R | – |
| Total: | 40 | 21 | 24 | 27 | 66 | 32 | 41 | 47 | 43 | 28 | 37 | 30 | 47 | 38 | 38 | 31 |

Presence or absence of species detected by:
A=trapped in artificial foam; B=baited leaves; R=randomly collected naturally colonized submerged leaves; F=filtration of stream water.

100% in the Khanspur Stream and the Jabori Canal. Communities detected by the combination of baited leaves and randomly sampled leaves (BR) showed the lowest similarity index with the "true" community of 64.86% in the New Stream, 70.76% in the Jabori Canal, 83.14% in the Sanatorium Stream and 87.75% in the Khanspur Stream (Table 7). Communities based on data generated by trapping conidia in artificial foam, baited leaves and filtration of stream water (ABF) or randomly sampled leaves (ABR) showed a similarity index of 97.95% with the "true" community in the New Stream, 99.02% in the Sanatorium Stream, and 100% in the Jabori Canal and the Khanspur Stream (Table 7).

## Discussion

There was no correlation between number of conidia in stream water and the degree of colonization of these fungi on submerged leaves. *Heliscus lugdunensis, Lunulospora curvula, Flagellospora curvula* and, at times, *Anguillospora longissima* dominated the freshwater hyphomycete communities detected by filtration techniques (F). On the other hand, *A. acuminata, T. marchalianum, C. aquatica, L. aquatica* and *C. longibrachiata* were dominant species on the baited leaves (B) and the randomly collected leaves (R). Chamier and Dixon (1982), Sanders and Anderson (1979), Shearer and Webster (1985c) also found large discrepancies for individual

Table 7. Similarity indices between freshwater hyphomycete communities detected by each sampling technique used alone and in various combinations and the total number of species (T) in a stream based on data generated by four techniques used simultaneously.

| Streams | T* | Freshwater hyphomycete communities based in the data generated using sampling techniques singly and in different combinations | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | A | B | R | F | AB | AF | AR | BF | BR | FR | ABF | ABR | ARF | BRF | ABRF |
| The New Stream | 50 | 88.88 | 59.15 | 64.86 | 70.12 | 95.83 | 91.3 | 95.83 | 83.72 | 64.86 | 82.35 | 97.95 | 97.95 | 97.95 | 88.37 | 100.00 |
| The Jabori Canal | 74 | 94.28 | 60.37 | 71.30 | 76.68 | 100.00 | 95.77 | 99.31 | 83.46 | 70.76 | 83.07 | 100.00 | 100.00 | 99.32 | 84.37 | 100.00 |
| The Sanatorium Stream | 52 | 90.52 | 70.00 | 83.14 | 73.17 | 98.03 | 92.78 | 99.02 | 84.44 | 83.14 | 89.36 | 99.02 | 99.02 | 100.00 | 89.36 | 100.00 |
| The Khanspur Stream | 55 | 92.15 | 81.72 | 81.72 | 72.09 | 100.00 | 92.15 | 95.32 | 90.00 | 87.75 | 86.59 | 100.00 | 100.00 | 96.22 | 91.08 | 100.00 |

* Total number of species (T) in a freshwater hyphomycete community detected by sampling techniques used simultaneously. Communities of freshwater hyphomycetes detected by sampling techniques used singly; A=community detected in Artificial foam trap; B=baited leaves; R=on naturally colonized randomly sampled submerged leaves; F=in stream water; and in different combinations, e.g., AB=community based on data generated through artificial foam and baited leaves.

freshwater hyphomycete species between the number of conidia/L available for colonization and the actual fruiting pattern on baited leaves. However, some measure of the success of freshwater hyphomycete species in colonizing submerged baited leaves can be ascertained by comparing the composition of the conidial pool available for colonization with the species actually colonizing and sporulating on leaves exposed to the conidial pool.

The composition of the conidial pool of different species and the concentration of conidia of each species can accurately be worked out by the filtration method. Twenty-six species of freshwater hyphomycete detected on filters formed the conidial pool to the time of submersion of leaf baits in the Jabori Canal on October. This conidial pool was dominated by *F. curvula*. *Heliscus lugdunensis* and *L. curvula* (Table 2) were the second and third most common species. Twenty-two species formed the conidial pool in the Khanspur Stream in October. *Lunulospora curvula, H. lugdunensis* and *F. curvula* dominated this community (Table 3). The conidial pool had 15 species in the Sanatorium Stream (Table 4) and 14 species in the New Stream (Table 5). *Flagellospora curvula, H. lugdunensis* and *A. longissima* occurred dominantly in these conidial pools. All of the conidia present in the pool may not anchor on the submerged substrata. Different conidia of freshwater hyphomycetes are trapped on substrates (Webster, 1959) and by air bubbles selectively (Iqbal and Webster, 1973a). Incidentally conidia which anchor more efficiently on submerged substrates are caught up in foam concentrate with the same efficiency.

The trapping efficiency of air bubbles accumulating in artificial foam concentrate and the anchoring efficiency of submerged substrates can be assumed to be relatively constant for conidia of any species in different streams. Conidia trapped in foam concentrate thus represent proportionately the numerical strength of the inoculum potential to anchor on the submerged substrates. Not all conidia anchoring on the submerged material are viable. Temperature of stream water and residence period of a conidium in stream water will influence the viability of conidia (Iqbal and Webster, 1973a; Sridhar and Bärlocher, unpublished data), and the presence of inhibitors in submerged material may also affect the colonization (Gunasekara et al., 1983). Freshwater hyphomycete communities detected on the baited leaves are thus the outcome of successful colonization by viable conidia. The communities on baited leaves (B) were

Table 8. Floristic details of mountain streams detected by different sampling techniques.

| | Techniques | The New Stream | The Sanitorium Stream | The Jabori Canal | The Khanspur Stream |
|---|---|---|---|---|---|
| A. | Range in No. of species | 14–23 | 12–28 | 16–34 | 12–33 |
| | Average No. of species | 19.4 | 19.7 | 25 | 22.6 |
| B. | Range in No. of species | 7–14 | 7–16 | 14–26 | 13–20 |
| | Average No. of species | 10.3 | 11.5 | 18.3 | 16.8 |
| F(a) | Range in No. of species | 4–18 | 5–20 | 7–29 | 6–25 |
| | Average No. of species | 11.4 | 12.2 | 17.1 | 16.7 |
| F(b) | Range in No. of conidia/L | 220–1510 | 360–1800 | 310–6510 | 330–202 |
| | Average No. of donicia | 720.8 | 985.8 | 2500 | 1067.5 |
| R | Range in No. of species | 8–18 | 10–19 | 14–30 | 12–26 |
| | Average No. of species | 11.8 | 12.8 | 21.6 | 18.8 |

Communities of freshwater hyphomycetes detected by different sampling techniques. A=trapping of conidia in artificial foam, B=leaf baits, F=filtration of stream water, R=random sampling of naturally colonized decaying submerged leaves.

dominated by *F. curvula. Alatospora acuminata* was the second most common species in the Jabori Canal (Table 2) and the Khanspur Stream (Table 3). *Geniculospora inflata* and *A. tetracladia* were the second most common species in the Sanatorium Stream (Table 4) and the New Stream (Table 5) respectively. Other species among the 10 top-ranking species in all streams were *T. marchalianum, Articulospora tetracladia, Anguillospora longissima, Lemonniera aquatica, T. chaetocladium* and *C. longibranchiata. Geniculospora inflata* in the Jabori Canal, *H. lugdunensis* in the Khanspur Stream, *G. inflata* and *T. monosporus* in the Sanatorium Stream, and *G. inflata* and *H. lugdunensis* in the New Stream were also among the 10 top-ranking species. The communities in the artificial foam (A) were richer than those on the baited leaves (B), but the 10 top-ranking species were common to these two communities, although the orders of ranking were different (Tables 2-5). There were always fewer species of freshwater hyphomycetes on the baited leaves (B) than on the randomly collected leaves (R) (Tables 2-5). The communities on the randomly sampled leaves represent species actually sporulating inside streams (Shearer and Webster, 1985c). These communities represent a successional stage on the randomly sampled leaves (Gessner et al., 1993) and thus fall short of the complete community occurring inside the stream. The actual community of freshwater hyphomycete species will, therefore, be a community based on the data generated by these four sampling techniques used simultaneously. Communities of freshwater hyphomycetes in November so synthesized had 39 species in the Jabori Canal (Table 2), 35 species in the Khanspur Stream (Table 3), 30 species in the Sanatorium Stream (Table 4) and 23 species in the New Stream (Table 5). These communities were richer than those detected by the different sampling techniques used singly. All of the these communities were dominated by *F. curvula. Flagellospora curvula, Alatospora acuminata, Anguillospora longissima, Articuloapora tetracladia, T. marchalianum, L. aquatica, T. chaetocladium* and *C. longibrachiata* were common species to all of these communities. *Alatospora acuminata* was the second most common species in the Jabori Canal (Table 2) and the Khanspur Stream (Table 3), *C. longibrachiata* and *H. lugdunensis* were the second most common species in the Sanatorium Stream (Table 4) and the New Stream (Table 5). These communities, however, differed in the species composition and in rankings of the 10 top-ranking species. *Anguillospora longissima* was the third-ranking species in the Jabori Canal and the Khanspur Stream. The occurrence of *A. longissima* on wood substrata (Shearer, 1992) may be a factor in its abundance in the canal and the Sanatorium Stream. *Heliscus lugdunensis* was absent from the 10 top-ranking species in the Jabori Canal and it was ninth-ranking species in the Khanspur Stream and second in the New Stream. The occurrence of this species on coniferous needles (Iqbal et al., 1980) and woody substrata (Webster, 1992; Shearer, 1992) may contribute to its abundance in the Khanspur Stream and the New Stream.

In all comparisons between communities detected by the four sampling techniques used singly to investigate freshwater hyphomycete species from each stream once a month for a year, discrepancies were found in the rankings of species in the communities (Table 6). Similar discrepancies have been reported by Shearer and Lane (1983), Shearer and Webster (1985c).

Differences in species composition detected by different sampling techniques used singly in the same stream are indicative of the ineffectiveness of the sampling technique to detect a complete freshwater hyphomycete community. The freshwater hyphomycete community based on data generated from samples taken once a month during 1986-87 by four sampling techniques used simultaneously was richer (74 species) (Table 6) and accompanied by greater number of conidia in the Jabori Canal than other streams (Table 8). The freshwater hyphomycete community in the Jabori Canal also showed a higher species diversity (−2.8995421) than in the other streams.

The Jabori Canal has a rich riparian vegetation (Iqbal, 1992). Substrata in the form of deciduous leaves contributed by deciduous trees, and agricultural products such as wheat straw and rice straw affected the number of conidia in the Jabori Canal water, and this is well illustrated by the seasonal increase in conidia at the time of leaf fall (Iqbal and Webster, 1973b; Bärlocher and Rosset, 1982; Eggenschwiler and Bärlocher, 1983; Shearer and Webster, 1985a). Higher amounts of $NO_3$-N and $PO_4$-P in the canal water (Table 1) are another factor in the rich freshwater hyphomycete flora. Gunasekera et al. (1983) found that enriching river water with ecologically probable amounts of $NO_3$-N and $PO_4$-P enhanced the freshwater hyphomycete activity. Faster rates of degradation of baited leaves in the Jabori Canal and Sanatorium Stream than in the New Stream and the Khanspur Stream (Iqbal, unpublished data) and the higher number of conidia in the Jabori Canal (Table 8) in the presence of higher amounts of $NO_3$-N and $PO_4$-P than in the other three streams are in line with the findings of Suberkropp (1991).

The coniferous needles contributed by the coniferous trees sheltering the other three streams mostly resist the fungal attack due to the presence of inhibitors (Bärlocher and Oertli, 1978) and are a poor source of energy for these fungi (Rosset et al., 1982). Thus these needles have a poor freshwater hyphomycete flora (Iqbal et al., 1990a) as compared to deciduous leaves (Iqbal et al., 1990b).

Comparison of the communities detected by the different sampling techniques used singly and in various combinations with the "true" community based on data generated using four sampling techniques simultaneously showed that the technique of trapping conidia in artificial foam (A) used singly or in combination with leaf baiting technique (AB), or with leaf baiting technique and random sampling of leaves (ABR) or filtration (ABF), proved more reliable than other methods or combinations in all the streams to detect a community of freshwater hyphomycete closer to the "true" community (Table 7).

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