

FILOSPORELLA EXILIS SP. NOV. ON SUBMERGED PLANT DEBRIS
FROM BELARUS

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Abstract: *Filosporella exilis* sp. nov. is described from decaying grass blades, *Equisetum fluviatile* stems and conifer wood debris submerged in a river in Belarus. Besides underwater macroconidia formed on annellated conidiogenous cells, it also has arthroconidia typically produced aerially. Two microconidial states may be present in some isolates: one with phialides with long, cylindrical collarettes and another whose conidiogenesis is unclear.

Key words: Taxonomy, aquatic hyphomycetes, streams.

A number of fungi with filiform conidia was isolated in autumn in the years 1996 and 1997 during a survey of watercourses of the Berezinsky Biosphere Reserve, Belarus. This was undertaken to obtain cultures of aquatic hyphomycetes for use in screening for antimicrobial activity (Gulis & Stephanovich 1996). Several of these isolates belong to an undescribed species of *Filosporella*. The new species fits well into the group of nonsporodochial members of this genus which produce aerial arthroconidia and have underwater conidia growing on annellate conidiogenous cells. It differs from all five known species and therefore it is described here as new.

Filosporella exilis Gulis & Marvanová sp. nov.

Figs. 1-4

Etym.: *exilis* (Lat.) = thin (refers to the conidial shape)

Fungi mitosporici, hyphomycetes. Teleomorphosis ignota.

Coloniae in agar maltoso modice crescentes, pallide griseae usque ad isabellinae, leviter colore aurantiaco tinctae. Mycelium aerium griseoalbum, mycelium in substrato atrum, partibus nonnullis rugosum. Pars reversa atra, pallescens et leniter aurantiaca ad marginem versus. Hyphae aerae cum parietibus hyalinis vel fuscis, hyphae in substrato brunneae.

Status macroconidialis: *Filosporella*: subaquaticus. Conidiophora semimacronematosa, simplicia vel parce ramosa, septata, usque ad 980 x 3-4 µm, rami 17-69 x 2.5-3 µm, saepe binatim vel ternatim acrogeni vel laterales, typice adpressi. Cellulae conidiogenae incorporatae vel raro discretae, apicales vel laterales, 19-42 x 2.5-3 µm, percurrenter vel raro sympodialiter proliferantes, in parte distali saepe densiter annellatae subinflataeque.

Conidia singularia, apicalia, recta vel leniter curvata, cylindrica vel subangustata ad apicem versus, (90-) 128-180 (-240) x 2.5-3.5 μm , apex obtusus vel subulatus, basis truncata vel cum extensione subulata, percurrente, usque ad 31 μm longa ornata. Secessio schizolytica.

Status arthroconidialis: typice acrius, sed etiam subaquaticus. Parietes cellularum fuscae usque ad hyalinae. Conidiophora micronematosae vel semimacronematosae. Hyphae conidiogenae simplices vel ramificatae, fragmentatio schizolytica. Arthroconidia cylindrica vel subfusioidea, recta vel paulo curvata, nonnumquam cum cellulis inflatis, pauciseptata, raro ellipsoidea et aseptata, usque ad 200 x 2-2.8 μm , extremitates truncatae cum cicatrice subincrassata vel rotundata.

Status microconidialis I: subaquaticus. Conidiophora semimacronematosae, simplicia vel pauca ramosa, apicalia vel lateralia, usque ad 570 x 2.5-3.5 μm , septata, cellulae parietibus glabris vel verrucosis, fuscis. Cellulae conidiogenae phialidicae, acropneurogenae, singillatim vel usque ad 4 per apicem conidiophori, ovaes vel subcylindricae, fuscae vel hyalinae, 10-17 x 2.8-4.2 μm , collum cylindricum, 5-20 x 2-2.5 μm . Conidia bacilliformia, 7.5-10 x 1.5-2 μm .

Status microconidialis II: typice ad fragmenta macroconidiorum vel ad arthroconidia, conidiophoris absentibus. Cellulae conidiogenae incorporatae, determinatae, saepe vacuae, apicales et subulatae vel intercalares cum extensione conidiifera laterali. Modus conidiogenesis ignotus. Conidia aggregata, bacilliformia, nonnumquam leniter inflata in media parte, 4.5-11 x 0.8-1.2 μm , cicatrix nonnumquam subincrassata.

Habitat: ad caulem putridum *Equiseti fluviatilis* L. in flumine Busianka submersum, prope viam publicam inter oppida Minsk et Lepel, in reservatione Berezinski dicta, regione Vitebsk, Belarussia, mense Septembro 1996 V.I. Gulis legit.

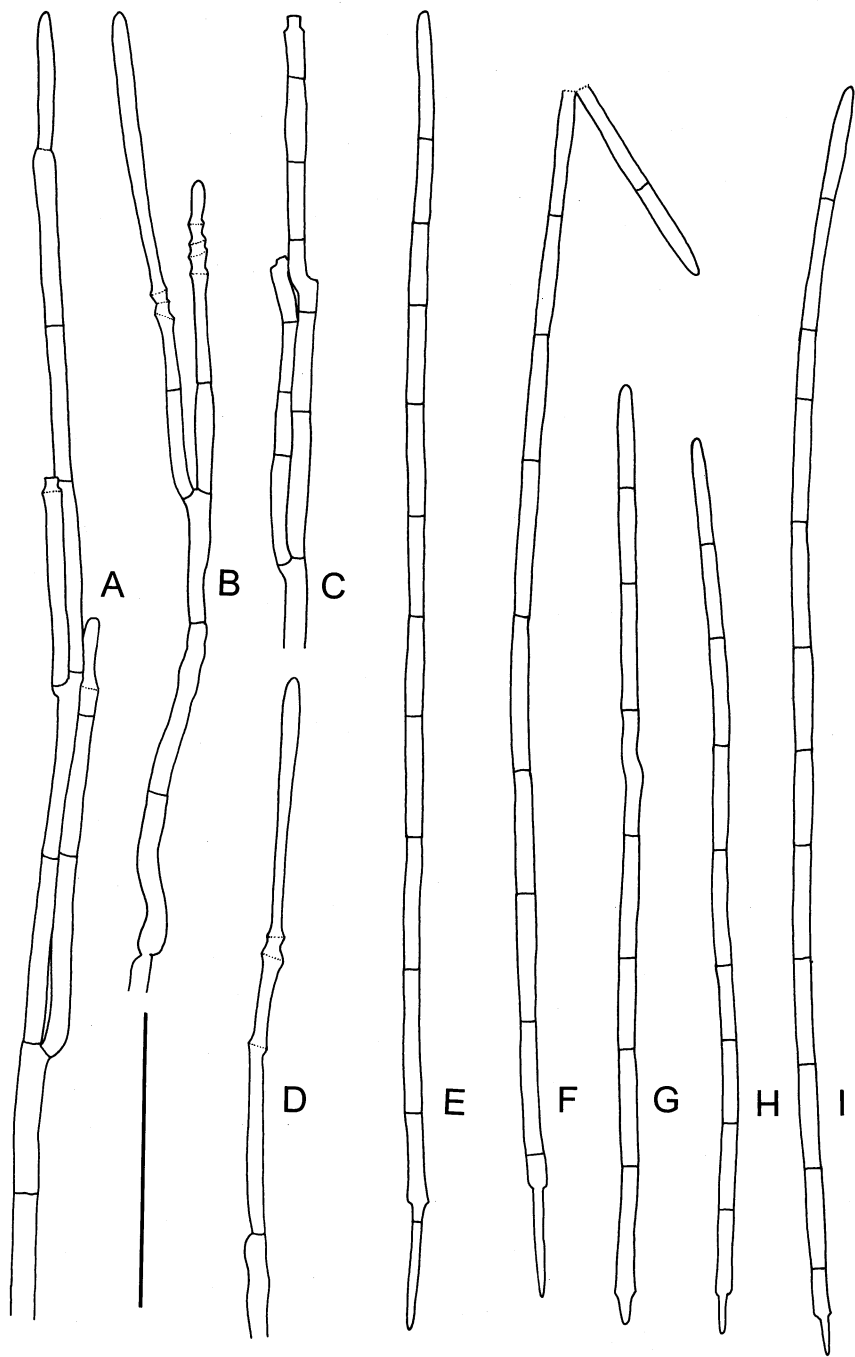
Holotypus: IMI 378148, ex VG-98a.

Mitosporic fungi, hyphomycetes. Teleomorph unknown.

Colonies (2% malt agar) growing moderately fast, reaching 15-25 mm diam. in 13 days at 15° C, pale grey to isabelline with an orange hue. Reverse blackish in the centre, paler or light orange at margin. Aerial mycelium cottony, dense and funiculose in the centre, sparse towards margin, whitish, becoming greyish with age; hyphae hyaline to fuscous, substrate mycelium fuscous to brown, hyphae up to 5 μm wide, walls slightly thickened, glabrous to verrucose. Reddish brown soluble pigment is produced in stationary liquid culture (potato maltose broth) in all isolates.

Macroconidial state: *Filospora* (Fig. 1 A-I, Fig. 3 A-D, Fig. 4 A,B,D,E, G-I), produced underwater, preferably at ca. 5° C. Conidiophores semimacronematous, simple or sparsely branched, septate, up to 980 x 3-4 μm , branches few, in one or two orders, 17-69 x 2.5-3 μm , acrogenous and then often binate or ternate, or lateral and then alternate, adpressed or slightly diverging. Conidiogenous cells integrated or discrete, 19-42 x 2.5-3 μm , apical or less often lateral, proliferations percurrent or rarely sympodial, distal part closely annellate, often slightly inflated. Conidia integrated with the conidiogenous cell at an early stage of development, single, straight, rarely slightly curved, cylindrical to slightly attenuate towards apex, (90-) 128-180 (-240) x 2.5-3.5 μm , up to 15 septate, apex rounded or subulate, base truncate or with a subulate percurrent extension up to 31 μm long, appearing soon after conidium release. Conidial secession schizolytic.

Fig. 1. *Filospora exilis*, VG-98a. A,B,D. Conidiophores with developing conidia of the *Filospora* state. Note the typical annellations on the conidiogenous cells. C. Spent conidiophore. E-I. Detached conidia. Scale bar = 50 μm .



Arthroconidial state (Fig. 2 D-H, Fig. 3 E-J, Fig. 4 C,F, J-M): typically produced aerially, but also underwater, cell walls fuscous to hyaline. Conidiophores micronematous or semimicronematous and then with sympodial elongations. Conidiogenous hyphae single or aggregated at the conidiophore apex, simple or branched, fragmentation schizolytic. Arthroconidia cylindrical or subfusoid, straight or gently curved, sometimes with inflated cells, few-septate, up to $200 \times 2-2.8 \mu\text{m}$, ends truncate with slightly thickened scars or somewhat swollen. Ellipsoid aseptate arthroconidia appear rarely.

Microconidial state I (Fig. 2 A-C, Fig. 3 K-O): produced underwater. Conidiophores semimicronematous, simple or sparsely branched, apical or lateral, up to $570 \times 2.5-3.5 \mu\text{m}$, septate, cell walls smooth or verrucose, fuscous. Conidiogenous cells phialidic, acro-pleurogenous, single or up to four per conidiophore apex, oval to almost cylindrical, fuscous or hyaline, $10-17 \times 2.8-4.2 \mu\text{m}$, collarete cylindrical, $5-20 \times 2-2.5 \mu\text{m}$, periclinal thickening distinct. Microconidia bacilliform, hyaline, $7.5-10 \times 1.5-2 \mu\text{m}$. Germination not seen.

Microconidial state II (Fig. 2 I-N): typically on fragmented macroconidia or on arthroconidia, in water, conidiophores lacking. Conidiogenous cells integrated, determinate, hyaline, often seen empty, apical and subulate or intercalary with a lateral conidiiferous peg. Method of conidiogenesis unknown. Conidia single or grouped, bacilliform, sometimes slightly inflated in the median part, hyaline, $4.5-11 \times 0.8-1.2 \mu\text{m}$, scar often somewhat thickened. Germination not seen.

Cultures examined: VG-86, VG-97, from unidentified submerged decaying grass blades, Sep. 1996, river Serguch nr. the village Kraitsy, 160 m alt., the Berezinsky Biosphere Reserve, Vitebsk Region, Belarus; VG-98, VG-98a, from decaying stems of *Equisetum fluviatile*, Sep. 1996, river Busianka nr. Minsk-Lepel highway, 170 m alt., the Berezinsky Biosphere Reserve, Vitebsk Region, Belarus; VG-130, VG-131, VG-132, from submerged decaying branch of *Picea abies* (L.) Karst. with needles, Oct. 1997, Serguch Canal nr. the village Kvetche, 160 m. alt., the Berezinsky Biosphere Reserve, Vitebsk Region, Belarus. All material collected and cultures isolated by V.I. Gulis.

The macroconidia are produced in a few days after submergence of culture slices, but at the same time there are also the arthroconidia, which to some extent overlap in size and shape with the former.

The isolates are slightly different from each other: the colony diam. under the same conditions varies from 15 mm in VG-98a to 25 mm in VG-98. The colony colour is pale grey in VG-98a, pale greyish-olive with an orange hue in the rest of isolates, producing copious honey-coloured exudate in VG-98. Along with this, the black reverse under the central part of the colony is surrounded by a pale zone in 98a, which is more distinct in VG-86 and VG-97 and vivid orange in VG-98. The *Filosporella* state is most pronounced in VG-98a, which reacts very positively (producing masses of conidia) when culture pieces in water are subjected to aeration. In the other isolates it declines from VG-86 through VG-97 to VG-86, but it is relatively abundant in the isolates VG-130, VG-131 and VG-132. There are also differences in the intensity of macroconidiophore branching: it is rather poor in VG-98a, where unbranched conidiophores are not exceptional. Macroconidia vary in size and shape: they are shorter ($125-175 \mu\text{m}$) and usually more pointed in VG-86, VG-98, VG-130, VG-131, VG-132.

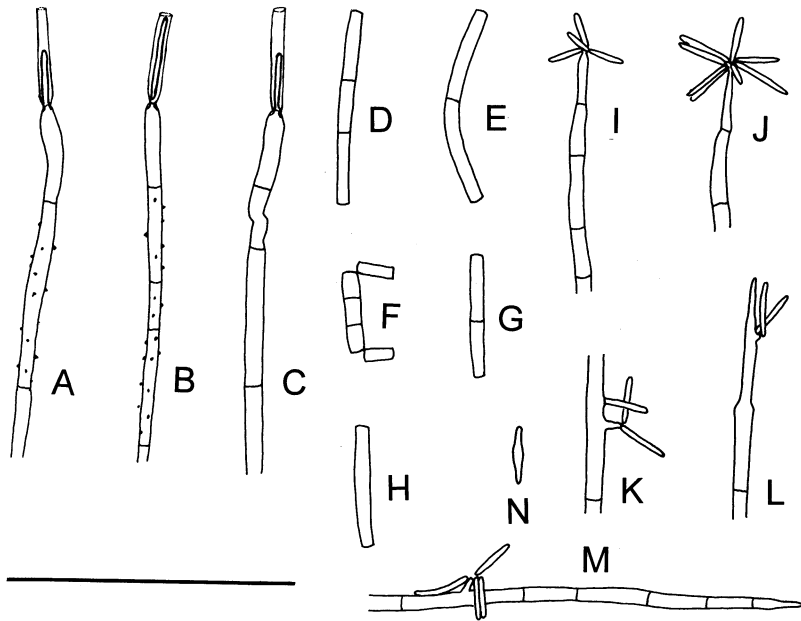


Fig. 2. *Filospora exilis*, VG-98a. A-C. Microconidial state I. Note the long collarettes on phialides. D-H. Arthroconidia. I-M. Microconidial state II. Conidiogenous cells with conidia. N. Medially inflated conidium. Scale bar = 50 μ m.

The arthroconidial state appears abundantly in all isolates on agar media without submergence and typically above water level, but sparsely also underwater in standing as well as aerated cultures. There are small differences in morphology: the arthroconidia are typically short-cylindrical in VG-97 and VG-86, often with inflated cells in the latter, sometimes fragmenting into short ellipsoid cells in the former isolate. They are very uniform in VG-98, VG-130, VG-131 and VG-132: cylindrical or slightly subfusoid, the majority ca. 20-30 μ m long. In VG-98a they are often long-cylindrical, sometimes indistinguishable from macroconidia.

Microconidial state I was observed in five isolates, i.e. VG-86, VG-97, VG-98a, VG-131 and VG-132. Microconidial state II was found in VG-97 and VG-98a. The exact method of conidiogenesis could not be interpreted with the aid of light microscope. Little fascicles of conidia are seen at the apices of conidiogenous cells, but it is unclear whether they are produced from a single or multiple conidiogenous locus.

It may be questioned whether it is wise to classify all the above isolates in the same species when there are such differences in conidium morphology, i.e. some isolates with short conidia and others with conidia nearly 2.5 times longer and arthroconidia either with inflated cells which do not appear as a result of ageing of the culture or still others with long-fusoid or cylindrical shape. However, we see a unifying feature in all these variations: (1) the conidiophores of the *Filospora* state are similar in the typically parallel arrangement of their branches, (2) there are always at least some cylindrical

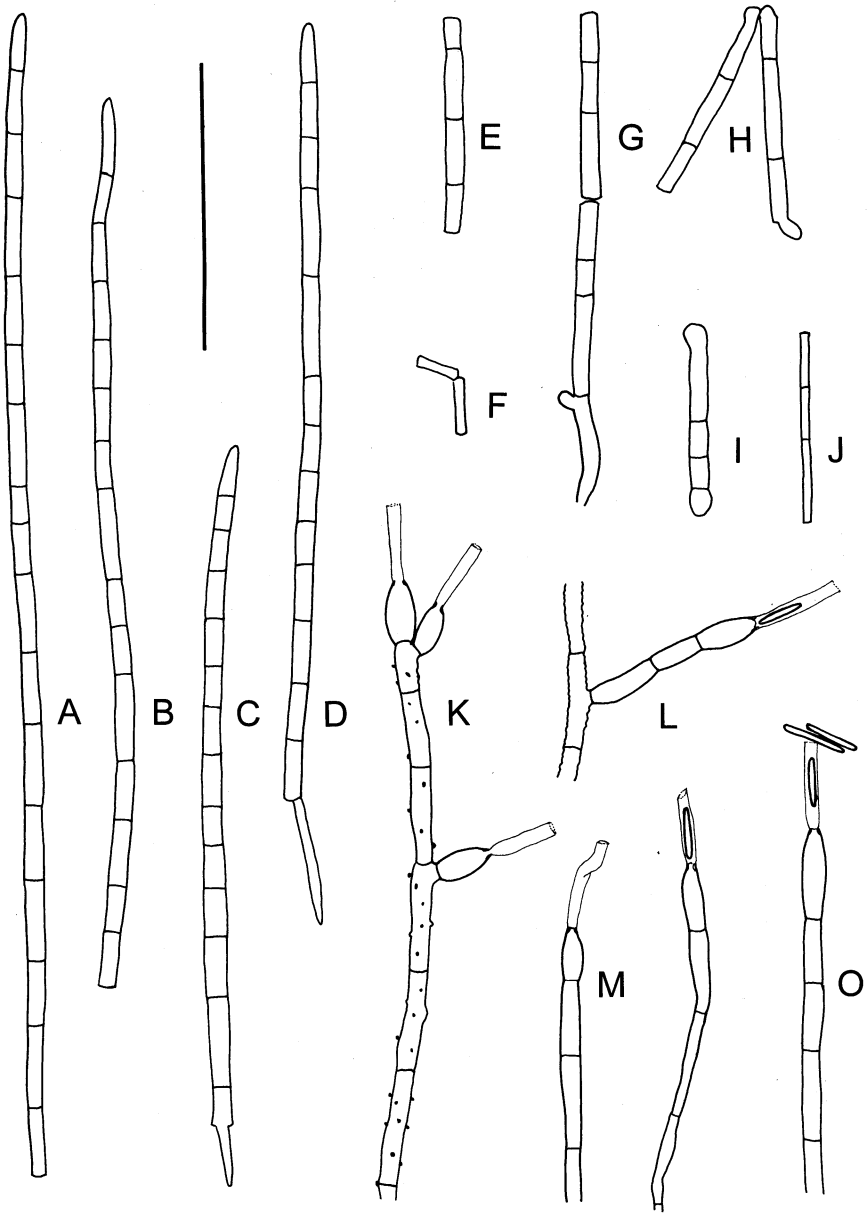


Fig. 3. *Filosporella exilis*, VG-97. A-D. Detached macroconidia. E-J. Arthroconidia. Note the somewhat inflated end-cells in I. K-O. Microconidial state I. Note the coarsely verrucose conidiophore in K and the rough-walled hypha in L. Scale bar = 50 μ m.

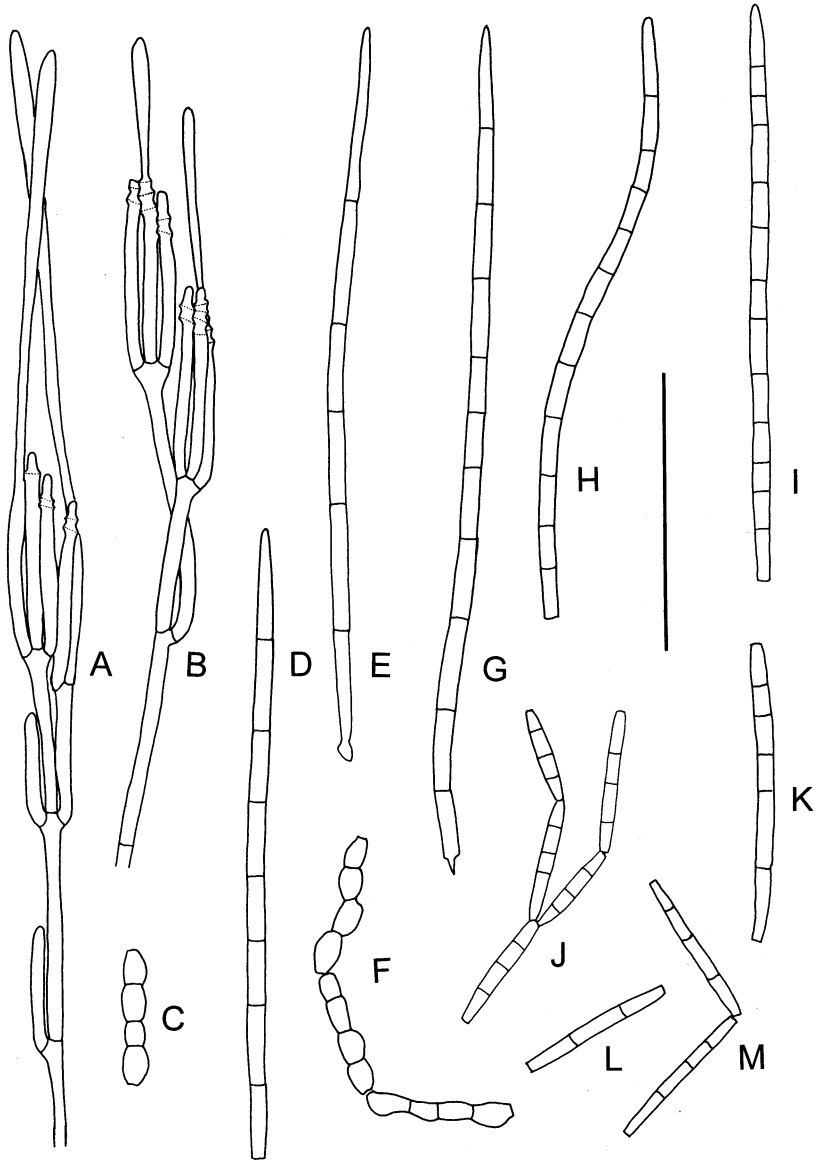


Fig. 4. *Filosporella exilis*, VG-86. A,B. Conidiophores with developing conidia. Note the profuse branching in contrast with VG-98a. D,E,G. Macroconidia. C,F. Arthroconidia with inflated cells. VG-98. H,I. Detached macroconidia. J-M. Arthroconidia. Scale bar = 50 μ m.

arthroconidia and (3) the absence of microconidial states in some isolates of the same species is known in other aquatic hyphomycetes also.

The disposition of this species in *Filosporella* is substantiated by branched conidiophores, and percurrent (rarely sympodial or intermediate) proliferation of conidiogenous cells resulting in close annellations. The conidia are most similar to those of *F. aquatica* Nawawi, but shorter and distinctly narrower. The series of new conidiogenous cells described by Nawawi (1976) as formed by percurrent proliferation after secession of each crop of conidia has not been seen in our species; in *F. exilis* the conidiogenous cells show only slight increase in length after secession of each conidium, this process leaving the characteristic "rings" on the conidiophore apices. Neither arthroconidia nor microconidial states have been observed in *F. aquatica*. *Filosporella annelidica* (Shearer & Crane) Crane & Shearer (1977) has conidia of similar size to our species, but these are typically sigmoid, with subulate ends. The conidiophores in *F. annelidica* are sporodochial, profusely branched at several levels. Phialidic microconidial state and/or aerial arthroconidia are present in three species, but in them the conidia of the *Filosporella* state are much shorter: sigmoid in *F. versimorpha* Marvanová *et al.* (1992) and straight in *F. fistucella* Marvanová & Fisher (1991) as well as in *F. pinguis* Marvanová & Bärlocher (1998). The latter, unlike any other described *Filosporella*, has globose microconidia.

The type species, *F. aquatica*, was described from the tropics and so far we are aware, it is known from the type locality only (Nawawi 1985). The other four species occur in temperate climates. Two of them were isolated as endophytes from symptomless living roots of *Alnus glutinosa* (L.) Gaertn. (Marvanová & Fisher 1991, Marvanová *et al.* 1992).

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References

- Crane J.L. & Shearer C.A. (1977). *Rogersia*, a later name for *Filosporella*. *Mycotaxon* 6: 27-28.
- Gulis V.I. & Stephanovich A.I. (1996). Search for antibiotic substances producers among aquatic hyphomycetes. In: Vimba E., Piterans A. & Avota I. (eds.). *Fungi and Lichens in the Baltic Region: Abstracts of the 13th International Conference on Mycology and Lichenology*, Riga, pp. 19-20.
- Marvanová L. & Bärlocher F. (1998). New species of *Filosporella*, *Pachycladina* and *Pleuropedium* from Canadian streams. *Mycol. Res.* 102: 750-754.
- Marvanová L. & Fisher P.J. (1991). A new endophytic hyphomycete from alder roots. *Nova Hedwigia* 52: 33-37.
- Marvanová L., Fisher P.J., Aimer R. & Segedin B.C. (1992). A new *Filosporella* from alder roots and from water. *Nova Hedwigia* 54: 151-158.
- Nawawi A. (1976). *Filosporella* gen. nov., an aquatic hyphomycete. *Trans. Br. Mycol. Soc.* 67:173-176.
- Nawawi A. (1985). Aquatic hyphomycetes and other water-borne fungi from Malaysia. *Malayan Nature J.* 39: 75-134.