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New *Blennothrix*-species (Cyanophyceae/Cyanobacteria) from Nepal

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Abstract A new species of the cyanophyte/cyanobacterial genus *Blennothrix* (*B. ganeshii*) from running water in the Kathmandu Valley, Nepal, is described. The morphology of trichomes and filaments, type of filament branching, life cycle and the fine structure of cells are described, and the existence of the genus *Blennothrix* KÜTZ. ex ANAGN. et KOM. and its taxonomic classification into the family Oscillatoriaceae *sensu stricto* are proved. The genus *Blennothrix* now comprises 13 species; the comparison of diacritical features of *B. ganeshii* with the nearest *B. fontana* JAO is discussed.

Introduction

The genus *Blennothrix* with the type species *B. vermicularis* was established by KÜTZING (Phycol. gener., p. 226) in 1843. However, this genus and species were joined to the genus *Hydrocoleum* by GOMONT in a monograph (1892), which was proclaimed as the starting-point of oscillatorialean cyanophytes. This concept was commonly accepted by all scientists, and the generic name "*Blennothrix*" did not occur in any later studies and monographs. GOMONT's concept was also accepted in the well-known monograph of GEITLER (1932) which has served up to now as the most important determination key for cyanophytes.

In the last few decades, several taxonomic features in cyanophyte taxonomy were re-evaluated after the application of modern methods. The importance of new cytomorphological characters was recognized (type of cell division, position of thylakoids, structure of cell walls and sheaths, types of false branching, type of hormogonia formation, etc.), and the instability and variation of some traditional differentiating characters (presence or absence of sheaths, number of trichomes in a sheath, dimensions) were studied.

The traditional genus *Hydrocoleum* was also found to be heterogeneous during the reconstruction of the cyanophyte/cyanobacterial system based on the new recognized facts. The typical part of this genus, based on the type-species *H. homoeo-*

trichum KÜTZ. ex GOM. 1892, is related by trichome structure to the genus *Phormidium* and must be classified as a member of the family Phormidiaceae. On the other hand, the group of species with more or less wide trichomes (usually over 10 μm wide) and short discoid cells (to which *Blennothrix vermicularis* also belongs) is related by cell structure, type of cell division and trichome structure to the typical genera of the Oscillatoriaceae *sensu stricto* (*Oscillatoria* and *Lyngbya*) and must be classified into this family (within the order Oscillatoriales). Therefore, KÜTZING's "*Blennothrix*" was validated by ANAGNOSTIDIS & KOMÁREK (1988) for this genus, which contains 12 species. The main diacritical features of this genus are (Fig. 1a–f):

— Filamentous cyanophytes without heterocytes, akinetes and true branching of trichomes.
— Obligatory presence of sheaths, which contain one or several trichomes (a).
— Sheaths sometimes branched (b), opened at the apex (c).
— Trichomes composed from very short ("discoid") cells (d), dividing by the crosswise centripetal cleavage in a rapid sequence (oscillatoriacean type; comp. our Fig. 6a).
— Trichome disintegration into hormogonia by help of necridic cells (e).
— Growth and development of hormogonia is connected with a special type of sheath branching (f), but the "plectonematoid" type of trichome branching is missing.

The genus *Blennothrix* differs from the most related genera within the Oscillatoriaceae *sensu stricto* by the obligatory presence of sheaths from *Oscillatoria*, by the type of branching and by the obligatory presence of more trichomes in a sheath from *Lyngbya* and *Plectonema*, and by the absence of "plectonematoid" branching from *Plectonema*.

During a bilogical expedition by the Hokkaido University, Sapporo, from April to June 1968 to the Himalayas in central Nepal, and a later botanical expedition organized by the National Science Museum, Tokyo, and the Department of Medicinal Plants/HMG, Nepal, from July to October 1986 to the same region, a new interesting species of the genus *Blennothrix* (*B. ganeshii* spec. nova) was found.

Methods, description of locality

The new cyanophyte species was collected from stony substrate in streams and from an artificial furrow (aquaduct) with streaming, slightly eutrophised water between fishponds at Godawari, SE from Kathmandu, Nepal. The filaments were attached to the substrate, mixed with *Cladophora* sp., and appeared macroscopically also like fine *Cladophora*-species (Fig. 2). The type material was collected by one author (M.W.) on September 11th 1986, and is deponed in the National Science Museum (TNS), Tsukuba, under the no. 53354. The previous material was collected by the same author on April 18th 1968 in a natural stream, also near Godawari in the same area (temp. of water=17°C, pH=about 8).

The fixative used was about 3% glutaraldehyde. These cells were postfixed with

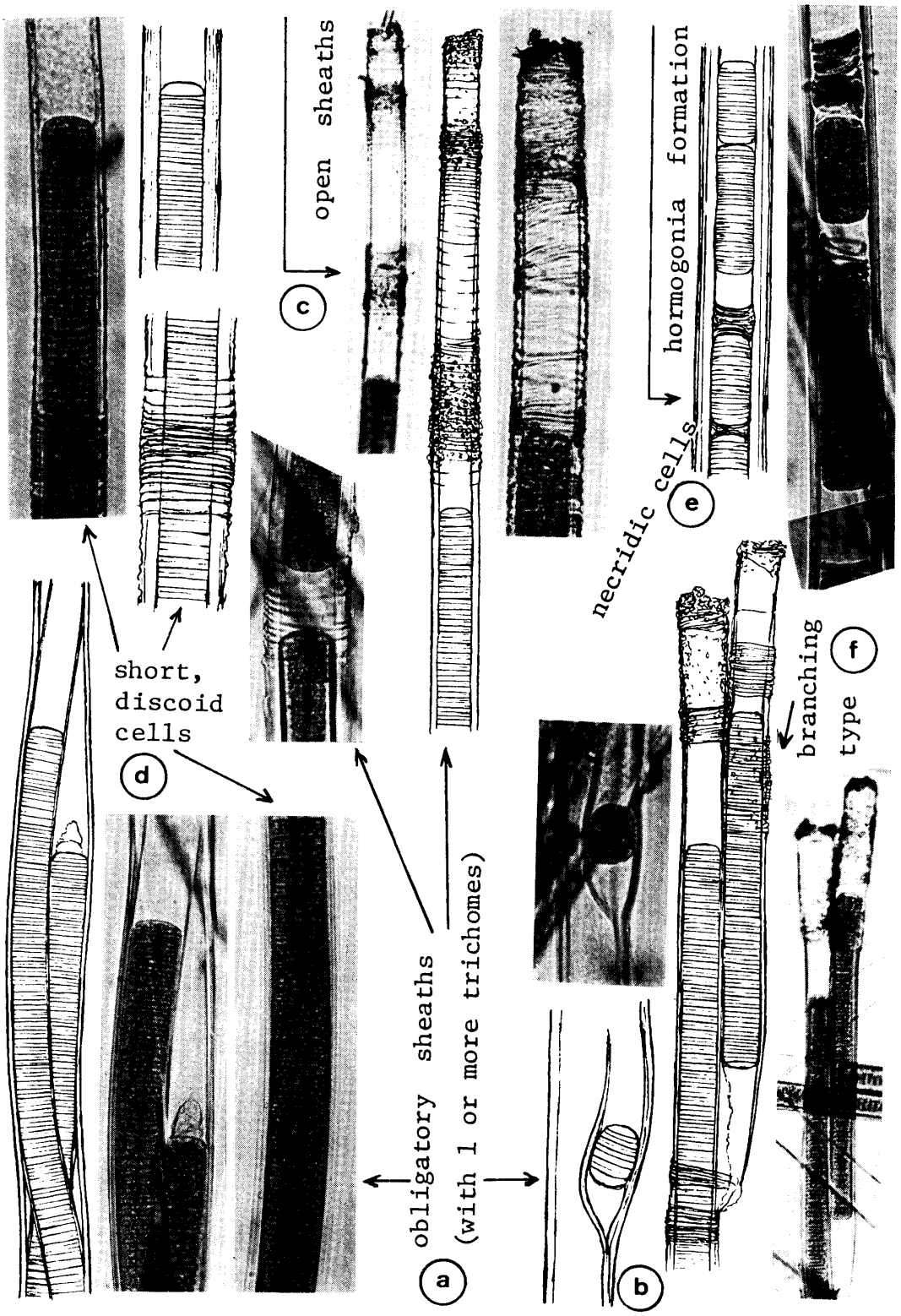


Fig. 1. Generic features of the genus *Blennothrix* demonstrated on the species *B. ganeshii* (see text).

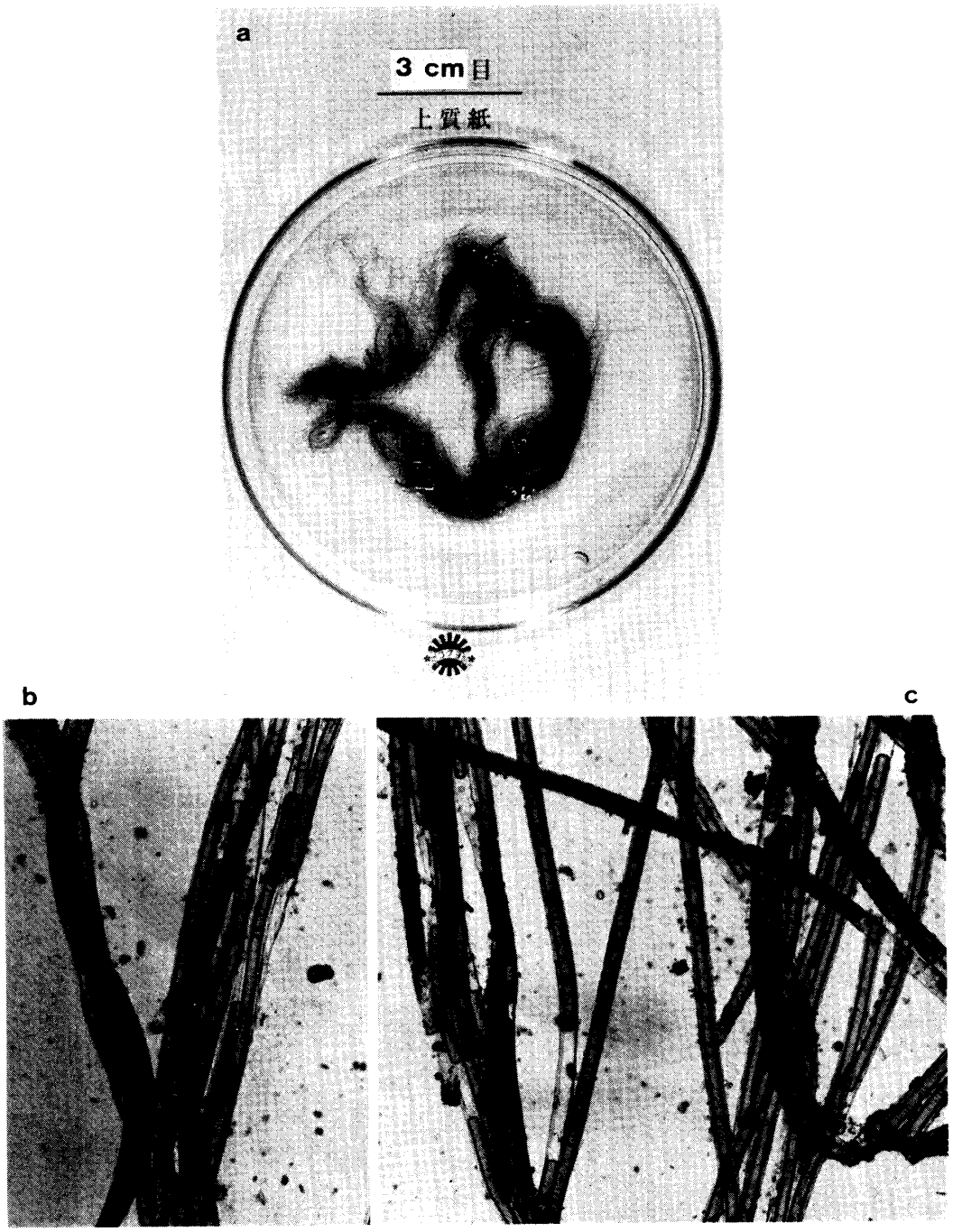
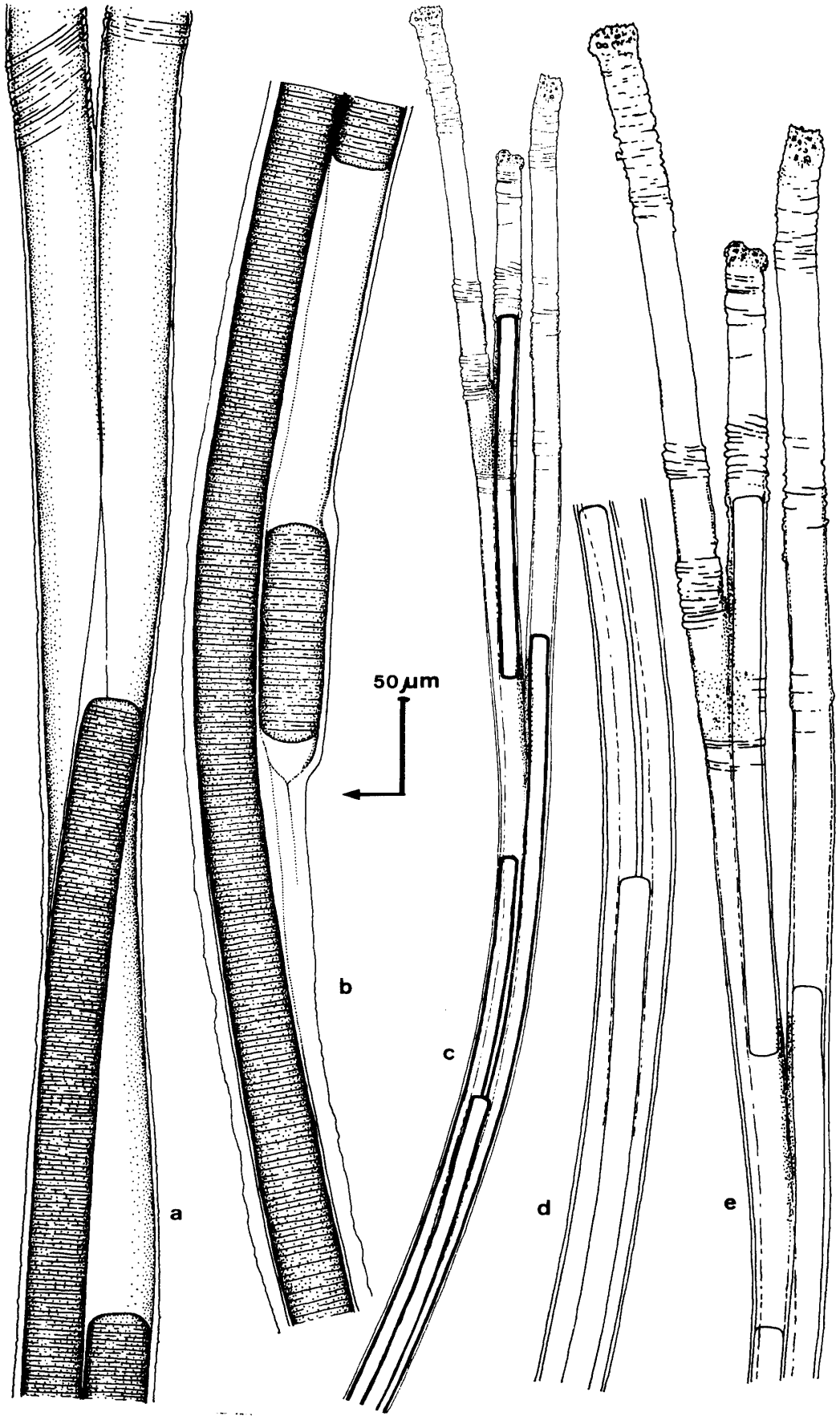


Fig. 2. *Blennothrix ganeshii*; a–macroscopic cluster of filaments, b–c–fasciculated filaments with more trichomes in a sheath.

Fig. 3. *Blennothrix ganeshii* (iconotype); a–detail of filament with two trichomes in a sheath, b–detail of filament with germinating hormogonium within the mother sheath, c – end of branched filament, d–e – details of end of branched filament. (Orig.)



2% Osmium tetraoxide, dehydrated with ethanol, and embedded in SPURR's epoxy resin. Thin sections were stained with uranyl acetate and lead citrate, then examined with a JEM 1200 EX electron microscope.

Results and discussion

Trichomes of the specimens from Nepal are thick, cylindrical, not attenuated at the ends, not constricted or slightly constricted at the cross walls. Cells are very short, 1.3 μm (after division) up to 4 μm long, with yellowish green or olive-green, finely homogeneously granular content, of the same type as in other oscillatoriacean genera. They divide by centripetal cleavage, sometimes in a rapid sequence (indistinct meristematic zones at the trichomes may occur). Width of filaments=46–100 μm, of trichomes=in average 35 μm. End cells are flattened, slightly rounded, sometimes with slightly thickened outer cell wall, without calyptra, but occasionally with remnants of cell walls of necridic cells (Figs. 3–5).

From EM-cross sections (Fig. 6) the basic structure of the cells (tyhlakoid arrangement, cell walls, etc.) are characteristic of all oscillatoriacean species (comp., e.g., HAGEDORN 1961, FJERDINGSTAD *et al.* 1976, etc; Oscillatoriaceae *sensu stricto*, see in ANAGNOSTIDIS & KOMÁREK 1988). Thylakoids are flexuous and situated more-or-less irregularly over the whole cell volume, sometimes lacunous. Polyphosphate granules and carboxysomes are also irregularly distributed. Cell division is characteristic for Oscillatoriaceae *sensu stricto* (Fig. 6a), and the rapid cell division of discoid cells (formation of new cell walls before the separation of the preceding divinding cells) was proved. The cell content seems to be vacuolized, but long-time, formol preserved material was used for EM, and fine structure was destroyed by this preservation. Sheaths are more-or-less homogeneous and their lamellation is evidently caused by the formation of new sheaths of growing hormogenoia and trichomes within mother sheaths, after disintegration and branching of the original trichome.

Sheaths are colorless, firm up to 10(–12) μm wide, usually lamellted more-or-less parallel to the length. Zones often occur with crosswise lamellation, particularly at the ends of sheaths. This lamellation is usually connected with the gelatinization of the outer sheath surface and with accumulation of small detritus particles on the surface (Fig. 4C). Sheath walls are more-or-less equally thick along the whole filament length. Each trichome also forms its own sheath and also when situated in parallel to and within the mother sheath.

Trichomes disintegrate into hormogonia of different cell number and length by help of one or several necridic cells. Hormogonia liberate from sheaths, or grow parallel within the mother sheath. After formation of hormogonia there occurs a special type of false branching (Fig. 7). Each hormogonium forms its own sheath within the mother sheath and grows firstly parallel to the original trichome. However, sometimes it is shifted aside from the original sheath, after a partial gelatinization of the mother sheath. Morphology of the whole thallus results from this branching

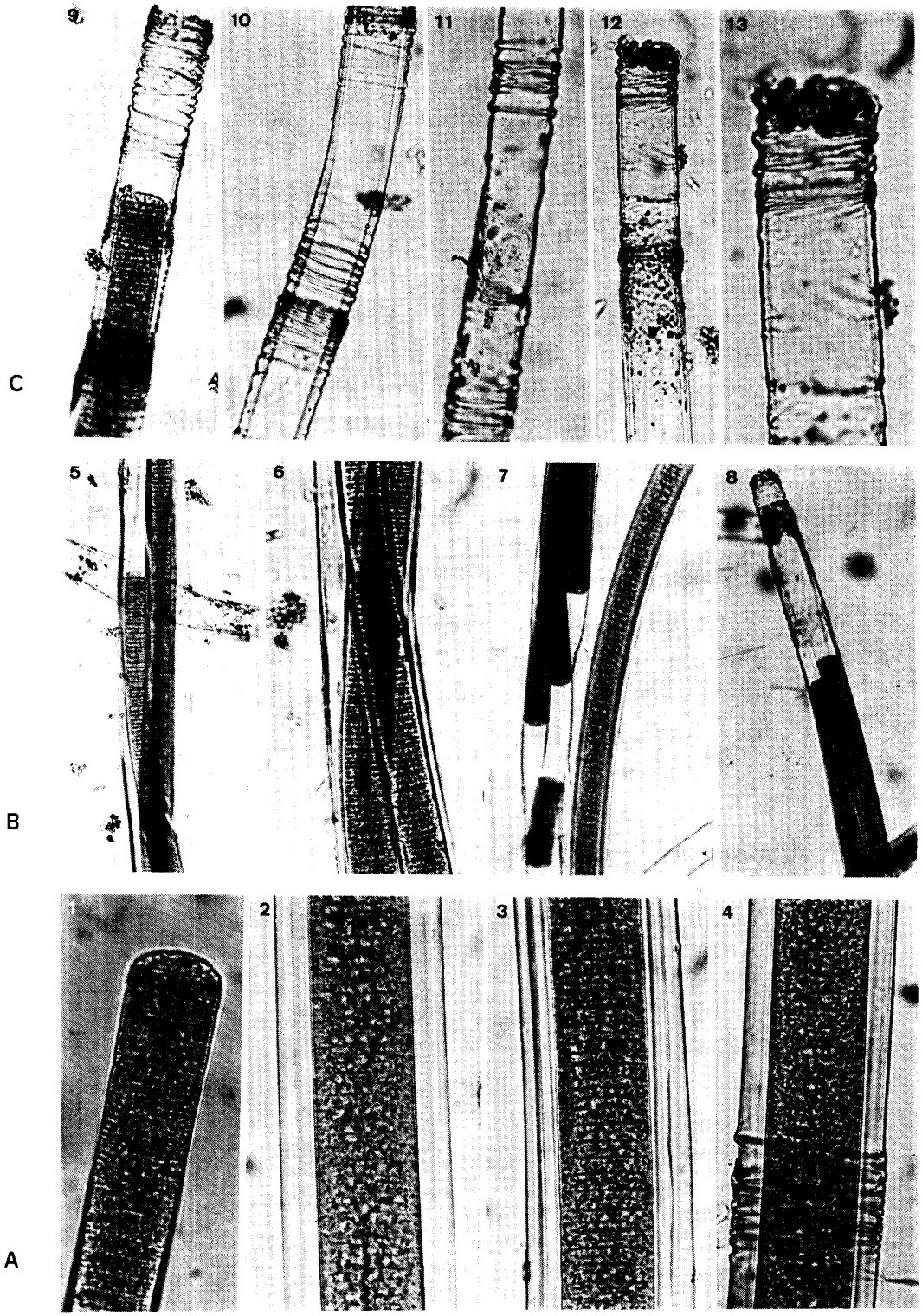


Fig. 4. *Blennothrix ganeshii*; A – parts of filaments with one trichome in a sheath (1 = end of free trichome withouth sheath), B – parts of filaments with more trichomes in a sheath, C – sheaths (13 = detail of empty end of a sheath).

process. Filaments are up to 6 cm long, more-or-less oriented in parallel, fasciclated branches (Fig. 2b–c). During this process, false branching of the "plectonematoid" or "scytonematoid" types never occurs, i.e., with the growth of disrupted trichome ends outside of the mother sheath. In *Blennothrix*, branching always follows after the formation of hormogonia. The initial growth is within the mother sheath and only after formation of its own sheath, does the apical end pass through the mother sheath. The resulting thallus has a typical polarized structure, differentiated in basal (joined to the substrate) and apical ends. However, this polarity is not manifest in trichome and filament morphology.

Our results (morphology of trichomes and filaments, type of branching, structure of cells, cell division, life cycle) support the existence of the special genus *Blennothrix* and its classification into the family Oscillatoriaceae [S. F. GRAY] HARV. ex KIRCHN. 1989 *sensu stricto* (comp. ANAGNOSTIDIS & KOMÁREK 1988). Within the genus *Blennothrix* are known species, from which several occur in marine habitats. The freshwater species grow mostly in clear, unpolluted waters, mainly in mountain lakes and springs. Our *Blennothrix ganeshii* belongs ecologically into this group. The most related species, morphologically and ecologically, is *Blennothrix fontana* (JAO) ANAGN. et KOM. 1988, described by JAO (1944) as "*Hydrocoleus fontanus*" from the periphyton on stones in the rapid stream in the district Yangso, South China. This species differs from *B. ganeshii* by somewhat smaller dimensions, and by the morphology of sheaths and thallus (Fig. 8). All other freshwater species have substantially smaller dimensions (see GEITLER 1932, p. 1146–1158, under *Hydrocoleus* KÜTZING).

Diagnosis

Blennothrix ganeshii, spec. nova: Thallus fasciculatus, luteo-viridis vel brunescens, ad 6 cm longus, ad lapides in rivulis adhaerens. Fila 46–100 μm crassa, paralleliter in thallo disposita, flexuosa et paucim intricata, superne pseudoramosa; intra vagina 1–4 trichomae; vaginae achroae, firmae, ad 10(–12) μm crassae, leviter lamellosae, ad partibus apicalibus partim perpendiculariter lamellosae, non incrassatae, apertae. Trichomata plus minusve 35 μm crassa, ad dissepimenta non constricta, apice non attenuata, obtuse rotundata, lutescente virides, sparsim granulata. Cellulae diametro trichomatibus, 1.3–4 μm longae, ad dissepimenta non granulatae.—Habitatio: Ad lapides in rivulis rapide fluentibus prope Godawari, Nepal centralis (1550 m supra mare).—Iconotypus: figura nostra 3a–e. Typus: TNS no. 53354 (in Museo Scienti Nationis, Tsukuba, depositus).

Etymology: This new species is the largest one from 13 members of this genus and characterized by the special morphology of prominent tube-like, trunk-shaped sheaths. It is named in honour of Genesh, the elephant-headed God, one of the most popular divinities in Hinduism, and the most popular in the Kathmandu Valley. Because he is regarded as a God of good luck, who casts obstacles aside, we would like to be in permanent contact with him in our Cyanophyte/Cyanobacteria research.

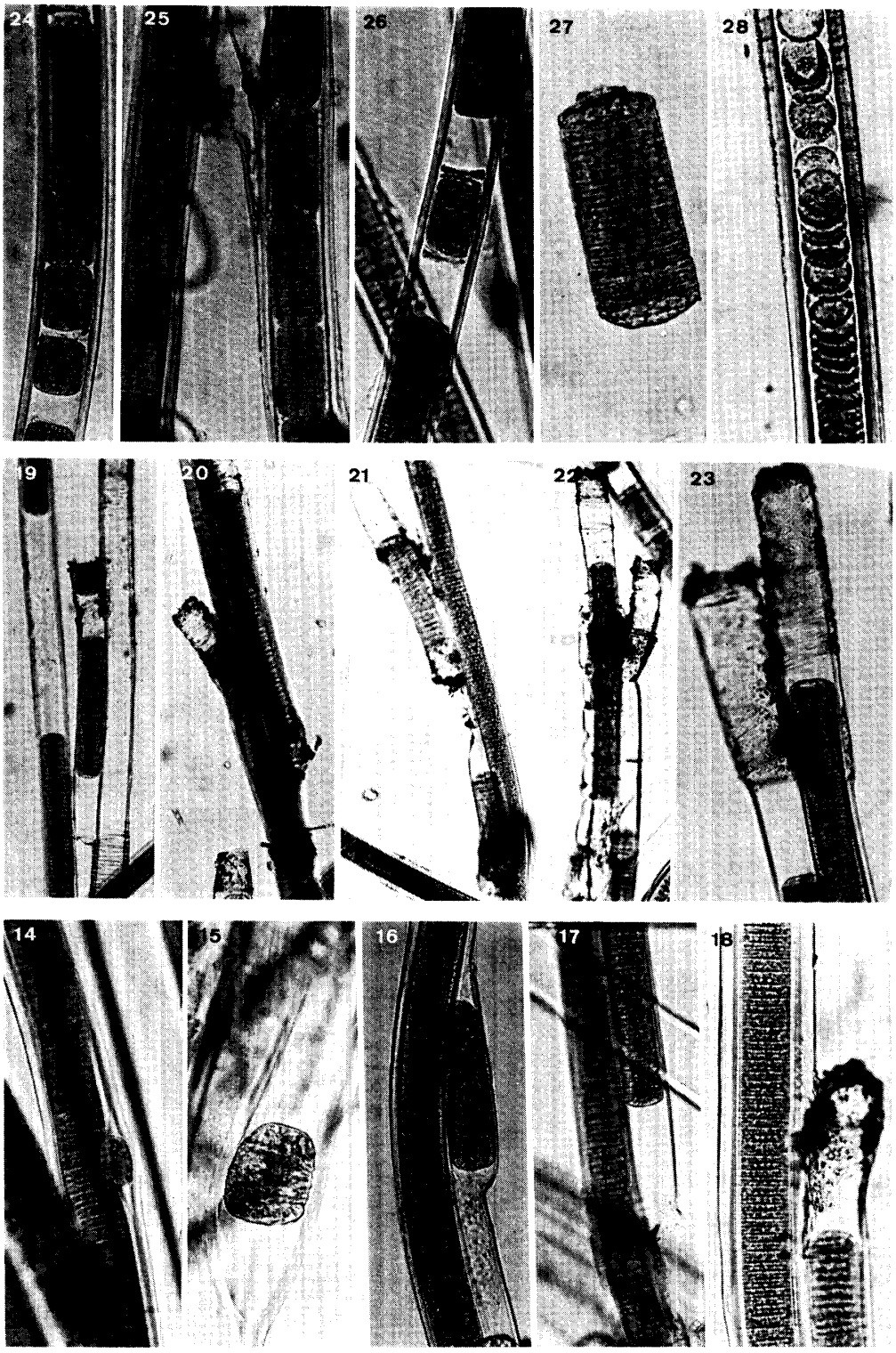
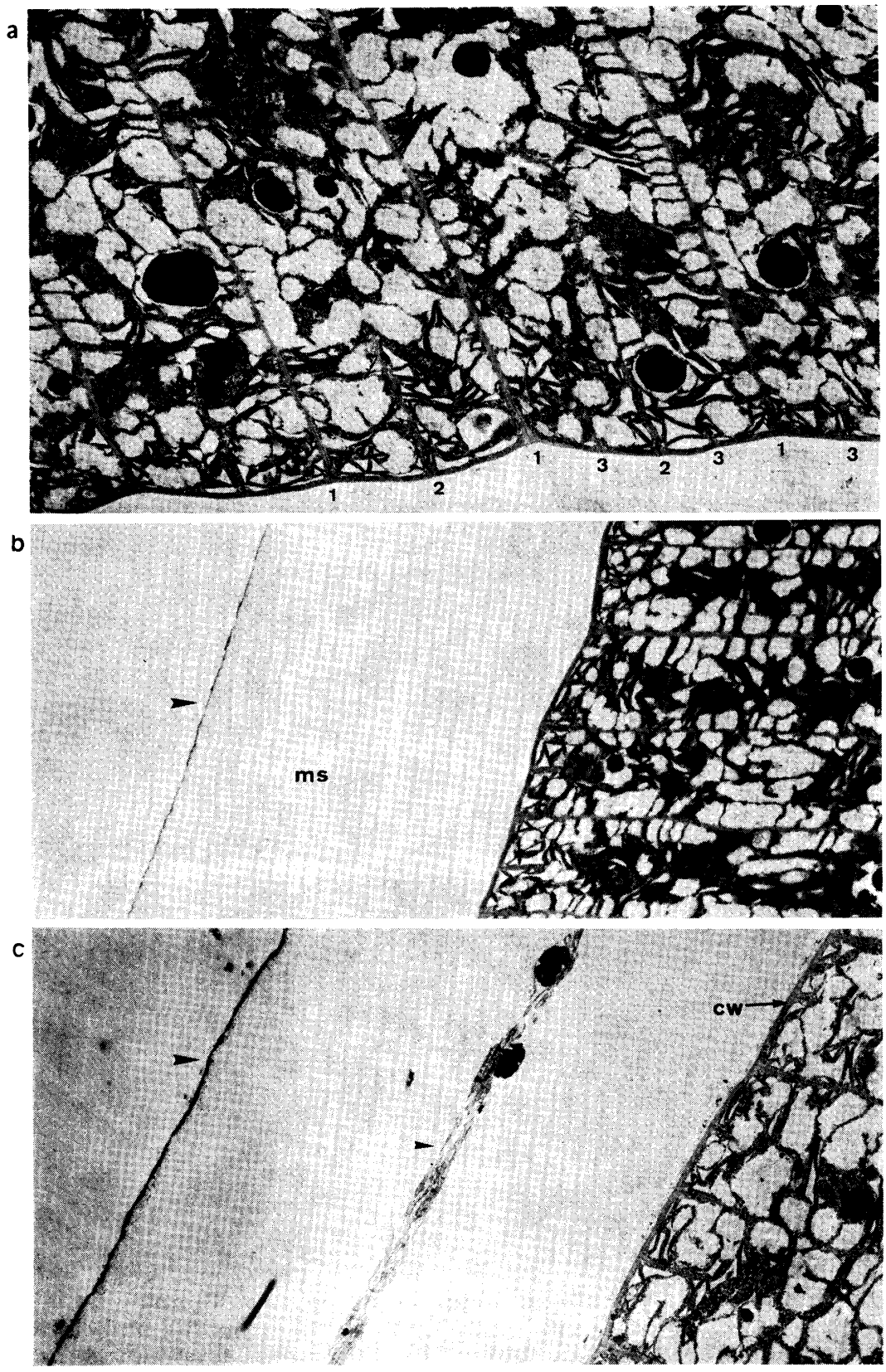


Fig. 5. *Blennothrix ganeshii*; D – germinating hormogonia within the mother sheath and initiation of branching, E – branching of filaments, F – disintegration of trichomes in hormogonia (27 = liberated hormogonium, 28 = disintegration of trichome in solitary cells).



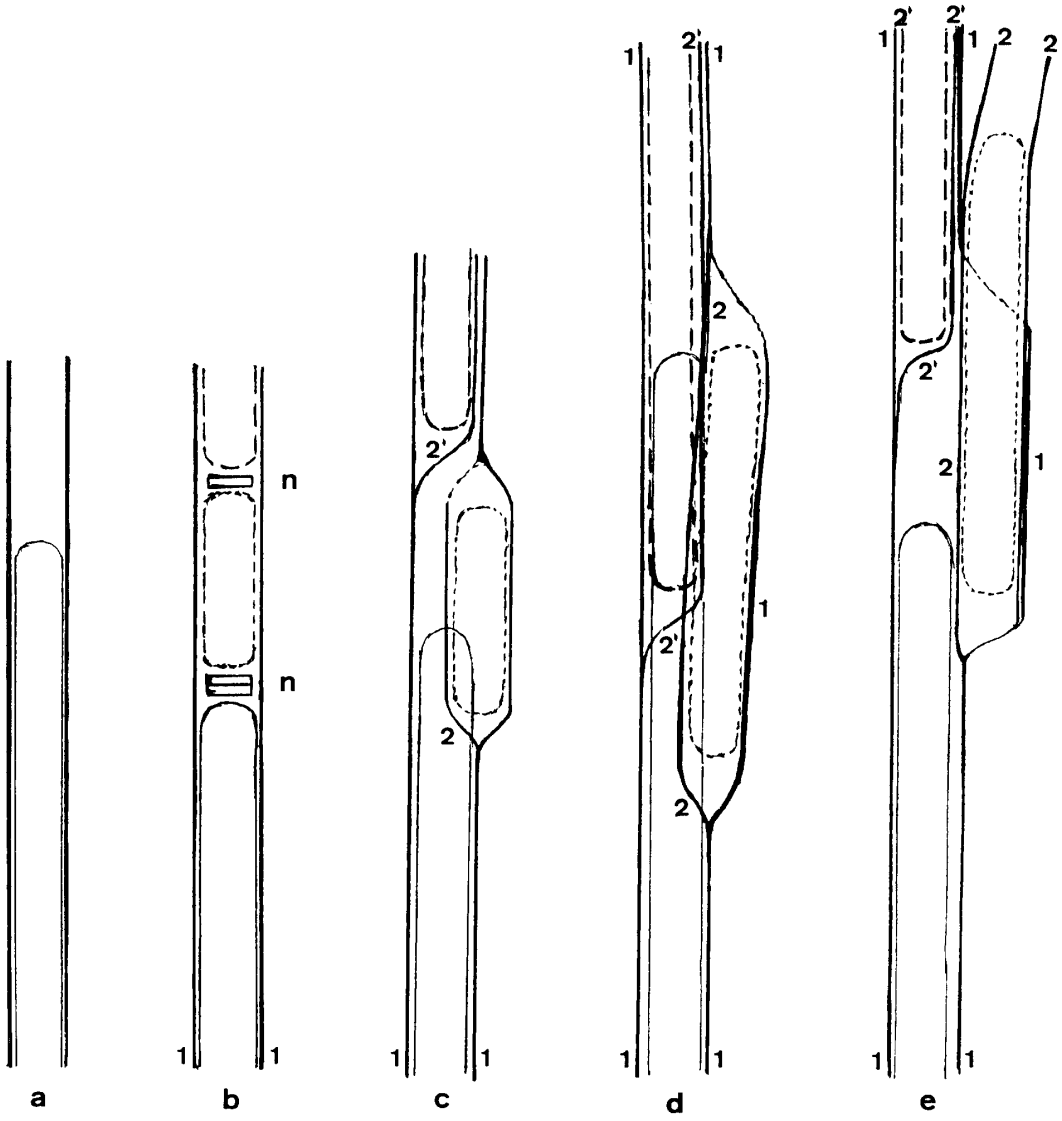


Fig. 7. Scheme of branching in the genus *Blennothrix* (derived from *B. ganeshii*); n = necridic cells, 1 = mother sheath, 2 = sheaths of germinating hormogonia within mother sheath. (Orig.)

Acknowledgments

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Fig. 6. *Blennothrix ganeshii*, EM-sections; a – lengthwise section through a part of a trichome (1 = closed cross walls, 2 = not closed cross walls, 3 = initiating cleavage of cross walls), b – mucilaginous sheaths (ms) of a simple filament with one trichome, c – double-layered mucilaginous sheath (arrows, cw = cell wall). (Orig. NIKI.)

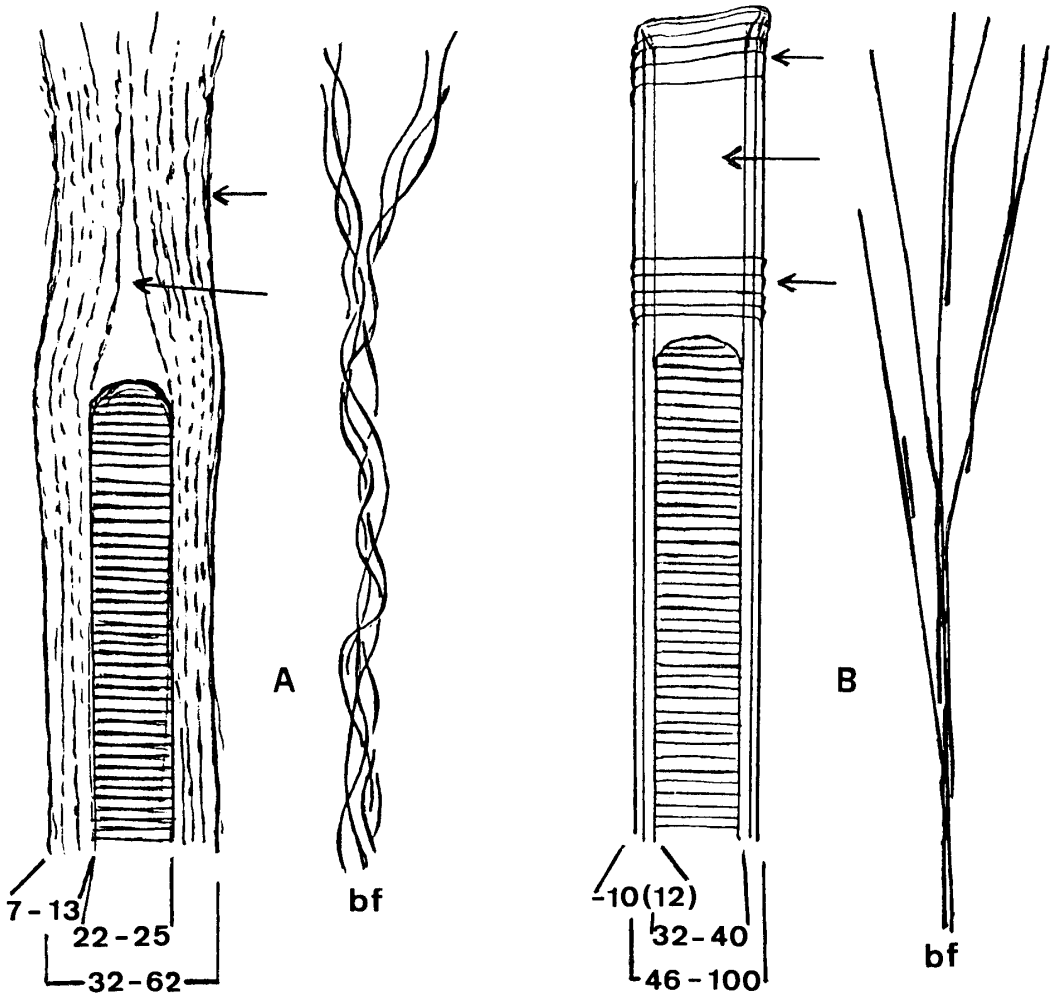

Fig. 8. Main differential features between *Blennothrix fontana* (A) and *B. ganeshii* (*B*); the numbers represent the variation in width of sheaths, trichomes and filaments in μm, bf= branching of filaments. (Orig.)

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