

Taxonomy and distribution of freshwater *Blennothrix ganeshii* Watanabe et Komárek (Oscillatoriaceae, Cyanophyceae) from central Mexico

by

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With 12 figures and 2 tables

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Abstract: The genus *Blennothrix* belongs to the family Oscillatoriaceae (subfamily Oscillatorioidae), differing from others in the subfamily by the presence of several trichomes in a sheath. Freshwater members of *Blennothrix* have been recently divided into eight species (Komárek 1998), with the taxon *B. ganeshii* Watanabe et Komárek reported from three sites in two tropical basins of the central region of Mexico. However, information on anatomical (vegetative and reproductive) characters and environmental distribution is scarce and species determination is uncertain. This study analyses the anatomical structure of *Blennothrix* populations distributed in different sites along the central tropical region of Mexico in order to determine the taxonomic status of the studied populations. Eight populations were sampled, with concurrent environmental data recorded. Morphological characters previously considered to be of taxonomic importance, as well as complementary features such as algal mat length, filament diameter, trichome width, length of cells, and thickness and shape of the sheath were measured in several filaments of each sample. Our results showed that all the observed populations in the central region of Mexico fit within the circumscription of *B. ganeshii* (Watanabe & Komárek 1989, Komárek 1998). However, two important morphological features were observed in all populations. First, branches were rarely present and varying in frequency from 1 to 3 trichomes per filament. Second, the presence of a transverse lamellation due to constriction in the longitudinal axis of the sheath was observed. Some morphological characters described for this species were extended.

Key words: *Blennothrix*, Cyanophyceae, Mexico, Oscillatoriaceae, springs, streams.

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Introduction

The freshwater and marine family Oscillatoriaceae is distinguished by: cylindrical trichomes with short, discoid cells; facultative or obligatory production of sheaths joined to the trichome, which are firm, sometimes slightly lamellated, containing one or more trichomes; filaments occasionally or obligately false-branched in ensheathed filaments; motile hormogonia or immotile hormocytes which develop by means of necridia formation (Anagnostidis & Komárek 1988). There are four distinguishable subfamilies within Oscillatoriaceae according to the frequency of false branching and filament morphology. The subfamily Oscillatorioideae Gomont is comprised of three well-characterized genera. The freshwater and marine genus *Blennothrix* (containing the former species of *Hydrocoleum* sine typo) differs from the other genera by the presence of several trichomes in a sheath. Freshwater members of *Blennothrix* have been recently divided into eight species (Komárek 1998).

Blennothrix ganeshii Watanabe et Komárek has been reported from Mexico, from three sites in the Balsas Basin (Valadez-Cruz et al. 1996). Montejano et al. (2000) listed this same species as a common component of the Mexican Tropical Panuco Basin. However, information on anatomical (vegetative and reproductive) characters and environmental distribution is scarce and the species determination is uncertain. The purpose of this study is to analyse the morphological structures of *Blennothrix* populations distributed in different environmental and geographic sites along the central tropical region of Mexico in order to characterize the degree of morphological plasticity and determine the taxonomical status of the studied populations.

Material and methods

Eight populations of *Blennothrix ganeshii* were sampled from the central region of Mexico (18–23° N, 99–100° W) in altitudes from 30–800 m (Fig. 1). The samples were observed alive, preserved after 6 hrs in 4% formaldehyde and deposited in the herbarium FCME (Holmgren et al. 1990). Temperature, pH, depth and type of substratum were recorded at each sampling site (sensu Carmona 1997). Shading and current velocity were estimated for each sampling site following Johansson (1982) and De Nicola et al. (1992). Quantitative and qualitative morphological measurements were made in 20 replicated filaments; the number of replicates was determined using the equation: $n = (s/Ex)^2$, where s = standard deviation, E = predetermined standard error (in this case 0.05) and x = average (Southwood 1978). Staining was done with 0.3% Alcian-Blue in 3% acetic acid at pH 2.5 (Sheath & Cole 1990). A BX51 Olympus microscope equipped with a SC35 photomicrographic system was used for observations, measurements, and pictures of samples. In each filament characters previously considered to be of taxonomic importance at generic and specific levels in relevant studies were measured (Frémy 1930, Geitler 1932, Anagnostidis & Komárek 1988, Watanabe & Komárek 1989, Komárek 1998), as well as algal mat length, filament diameter, trichome width, branching and number of trichomes in a filament (generic level), length of cells and thickness and shape of the sheath. Associations among populations were determined by cluster analysis with the unweighted group average method and by principal coordinates analysis (PCO) from a standardized data matrix (Valentin 2000). The analyses were performed with the STATISTICA statistical package based on morphometric features (trichome width, filament diameter, sheath thickness and cellular length). To compare means among populations, one-way analysis of variance and Student's t -test were also calculated.

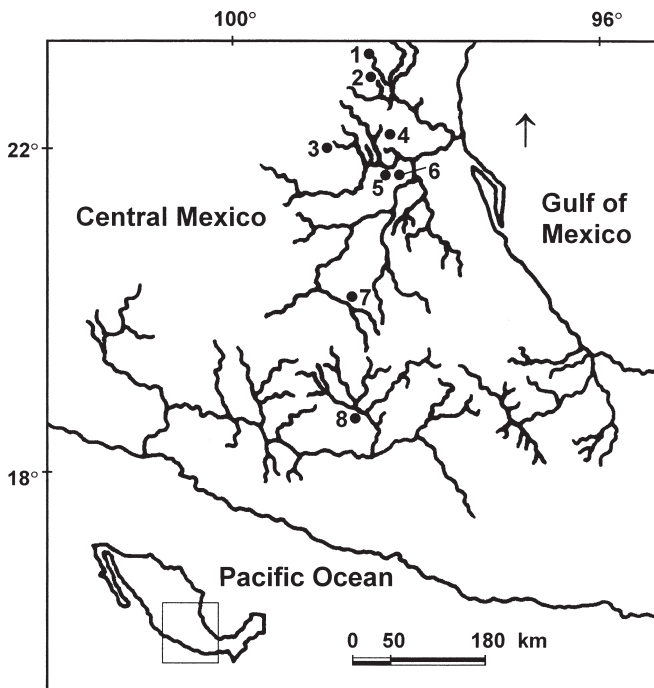


Fig. 1. Location of the study regions in Central Mexico with indications of the sites with freshwater populations of *Blennothrix ganeshii* (•).

Results and discussion

Morphological analysis

The cluster analysis and PCO show two distinct groups (Figs 2, 3). Group 1 contains seven populations with relatively large measurements often beyond the size range of previously described *B. ganeshii* and group 2 contains the Tzindejé population, which has the smallest dimensions of trichome width, filament diameter and sheath thickness (Table 1). Analysis of variance and Student's *t*-tests were not significant in one or more characters ($p < 0.05$). The general characteristics of our populations are: macroscopic violet-brownish algal mats growing on different substrates, mats composed of sparsely branched filaments with false branching (*Coleodesmium*-type) (Fig. 4). Filaments grew up to 10 cm long, and were more or less parallel. Cells had fine, homogeneously granular content and end cells were flattened and slightly rounded (Fig. 5), sometimes with slightly thicker outer cell wall, without calyptra, but occasionally with remains of necridic cell walls (Fig. 6). Sheaths were firm, colorless, usually lamellated; zones often occur with transverse lamellation due to constriction in the longitudinal axis, particularly at the ends of sheaths (Fig. 7). The lamellation in the outer sheath surface was characterized by several parallel layers which shed single layers and accumulate small detritus particles (Figs 8, 9). Trichomes

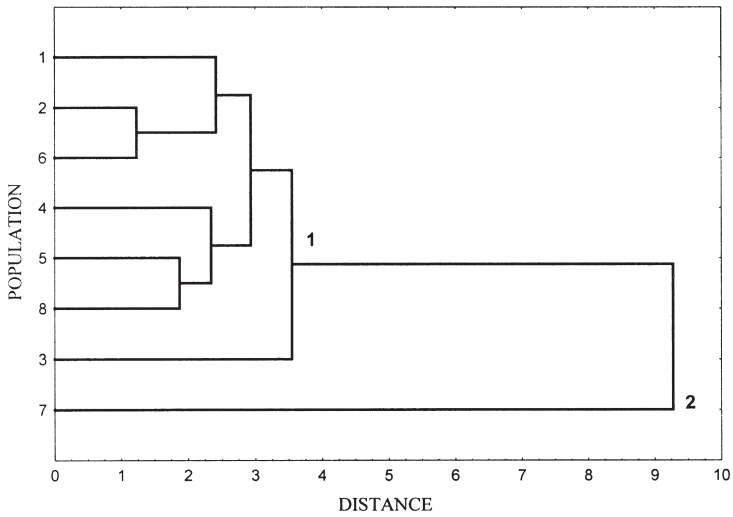


Fig. 2. Cluster diagram showing two groups of *Blennothrix* populations analysed. Group 1 contains seven populations and group 2 contain one population. The population numbers correspond to those shown in Table 1.

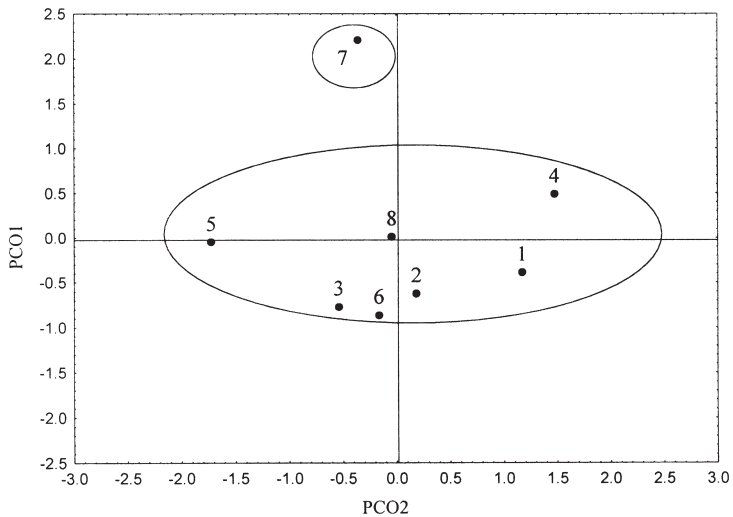
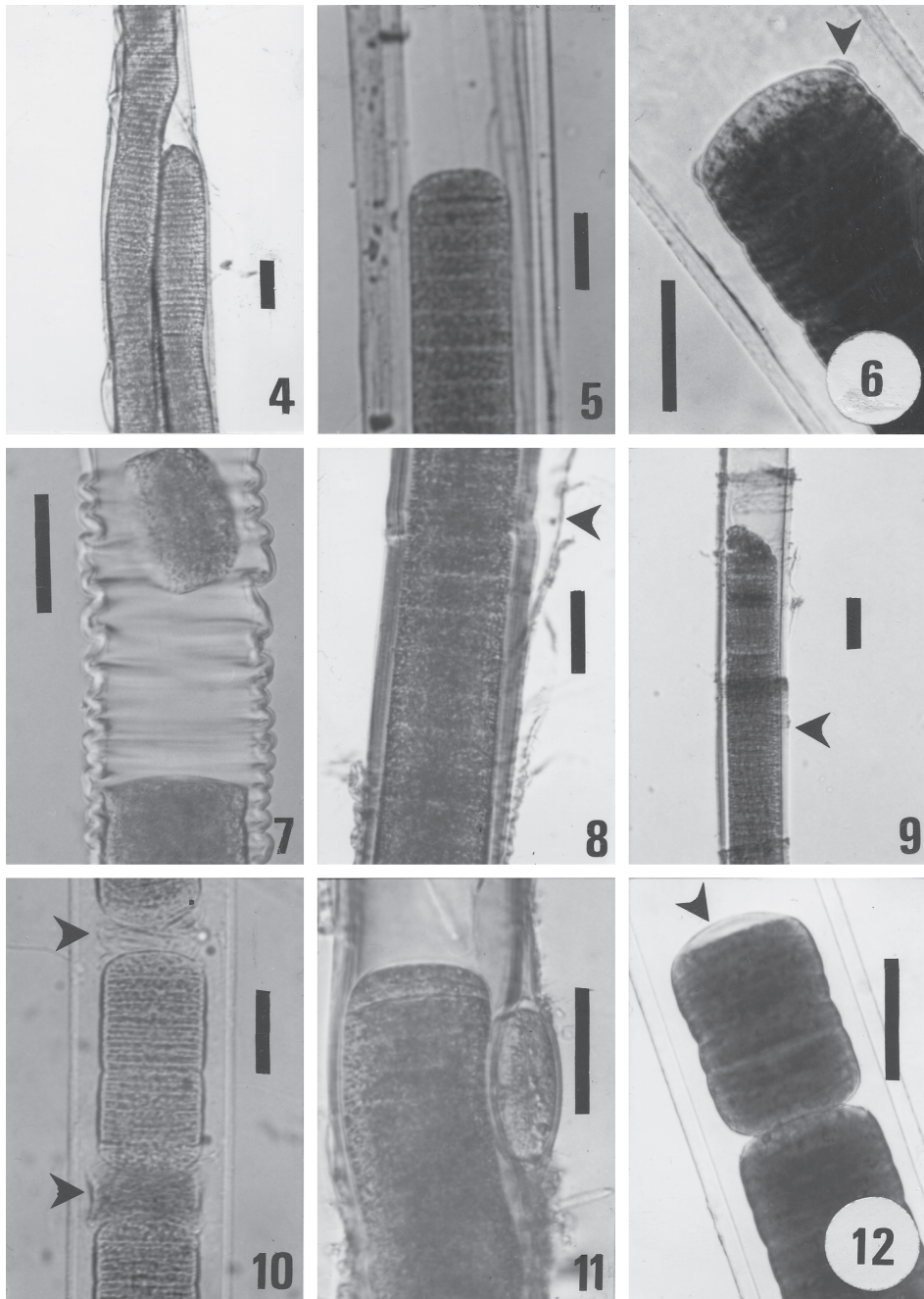


Fig. 3. Principal Coordinates Analysis (PCO) scatter plot of Mexican populations of freshwater *Blennothrix* showing two groups. The population numbers correspond to those shown in Table 1.



disintegrated into hormogonia through production of several necridic cells (Fig. 10). Hormogonia were produced either from the terminal part of trichomes or by disintegration of the whole trichome. Each hormogonium formed its own sheath within the mother sheath and at first grew parallel to the original trichome. After formation of hormogonia, *Coleodesmium*-type false branching occurred or the hormogonia were liberated (Fig. 11). Sometimes the apical hormogonial cell was empty (Fig. 12). The sheath exhibited a strong staining with Alcian-Blue, evidence of the presence of sulphated polysaccharides (Figs 4, 8, 9) which have been found in other freshwater species of the Oscillatoriaceae (De Philippis et al. 2001).

Two important morphological features were observed in all populations. First, branches were rarely present and varied in frequency from 1 to 2 branches per filament (rarely 3). Second, the presence of transversely lamellated shedding layers at the surface of the sheath (Figs 8, 9). The presence of 1 to 4 trichomes per filament was reported as an important taxonomic character for this species (Watanabe & Komárek 1989). False branching was rare; in general only one trichome was present in each filament, resembling species of the genus *Lyngbya*. Some branches were found in some filaments where two to three trichomes were present, confirming the determination of the population as members of the genus *Blennothrix*. All the observed populations (groups 1 and 2) in central Mexico fit within the species circumscription of *B. ganeshii* (Watanabe & Komárek 1989, Komárek 1998). Analysis of variance did not show significant separation of morphometric characters, and we conclude these analyses justify recognition of a single plastic species (Table 1). The variation in group 2 (population 7) might be explained by high current velocity (Table 2). However, we consider that microhabitat studies to characterize the morphological and physiological plasticity must be done in accordance with the proposals of Anagnostidis & Komárek (1985) and Komárek (1994). The presence of the transverse lamellation due to constriction of the sheath in the longitudinal axis, trichome width up to 25 µm, and filament diameter up to 62 µm distinguish the studied populations from *B. fontana* (Jao) Anagnostidis et Komárek. Filament length (> 6 cm), trichome width (> 40 µm) and cell length (> 4 µm) from Mexican populations extend the diagnosis for *B. ganeshii*.

Distribution

Blennothrix ganeshii was sampled in three streams and five springs from tropical calcareous zones, the sample sites are environmentally similar to other sites where

Figs 4-12. Morphological features of freshwater populations of *Blennothrix ganeshii*. Fig. 4. Filaments with *Coleodesmium*-type false branching and sheath stained with alcian-blue (BALE w/n). Fig. 5. End cells flattened and slightly rounded (FCME PA3874). Fig. 6. Remains of necridic cell walls on hormogonial wall (arrowhead) (BALE w/n). Fig. 7. Sheath with crosswise lamellation (FCME PA3707). Figs 8, 9. Lamellation in the outer sheath surface characterized by several parallel layers which shed single layers and sheath stained with Alcian-Blue (arrowhead) (FCME PA3874). Fig. 10. Trichomes disintegrating into hormogonia by means of several necridic cells (arrowheads) (FCME PA3880). Fig. 11. Germinating hormogonium within the mother sheath (FCME PA3707). Fig. 12. Empty apical hormogonial cell (arrowhead) (BALE w/n). Scale bars represent 40 µm.

the species has been reported in central Mexico (Valadez et al. 1996, Montejano et al. 2000). The populations of *B. ganeshii* from the central region of Mexico are the third described populations in the world, after those from Central Nepal (Watanabe & Komárek 1989).

Populations of *B. ganeshii* were found in habitats with warm temperatures (23.6-33°C), neutral pH (7.0-7.5), high conductivity (368-1530 $\mu\text{S cm}^{-1}$), shallow depth (5-50 cm) and low to fast flowing waters, in shaded or open river segments, and on rocky substrates or fallen tree trunks (Table 2). The pH was similar to that reported by Watanabe & Komárek (1989), however, temperature values were higher than those recorded by the same authors (17°C) and also higher than those found in cold mountains from central Mexico by Komárek et al. (1996). Filaments of *B. ganeshii* were used as substrate by several epiphytic Cyanophyceae species such as *Chamaecalyx swirenkoi*, *Stichosiphon sansibaricus*, *Xenococcus bicudo*, and *X. willei*. Populations of *B. ganeshii* were found mixed with *Chara canescens*, *Cladophora* sp., *Vaucheria* sp., *Spirogyra* sp. and were associated with several red algae, such as *Thorea hispida*, *Hildenbrandia angolensis* and the *Chantransia* stage of *Batrachospermum*.

In summary, our study has shown that there is one infrageneric taxon of *Blennothrix* in central Mexico. Trichome width, false branching, and sheath constriction were the features that distinguish this species. However we have found a wide variation that needs to be resolved by means of cultures, microhabitat, morphometric and physiological studies, as well as molecular analyses that indicate genetic separation and not necessarily ecophenotypic variation.

Description and taxonomic proposal

Blennothrix ganeshii Watanabe et Komárek. - Bull. Natl. Sci. Mus., Tokyo, B 15 (3): 74, 1989 (Figs 4-12)

Macroscopic violet-brownish algae, sparsely branched filaments (1 to 2, rarely 3 trichomes) with false branching *Coleodesmium*-type. Filaments up to 10 cm long, 45.2-75.0 μm diameter; trichome width of 33.9-50.0 μm . Cells 2.2-6.7 μm length; end cells are flattened and slightly rounded, sometimes with slightly thicker outer cell wall, without calyptra, but occasionally with remains of necridic cell walls in hormogonial wall. Sheath firm and colorless, 3.2-11.0 μm thickness, usually lamellated and with transverse lamellation due to constriction in the longitudinal axis, particularly at the ends of sheaths; the surface sheath sheds layers. Trichomes disintegrate into hormogonia of different cell number by several necridic cells and are produced from the terminal part of trichomes or by the disintegration of the whole trichome; hormogonia are liberated from sheaths, or grow parallel or perpendicular within the mother sheath.

Specimens examined: (1) Tamaulipas, Ciudad Mante, Las Playitas, coll. J. Carmona, 12.v.1997 (FCME PA3707); (2) Río Frío, coll. J. Carmona, 12 v. 1997 (FCME PA3717); (3) San Luis Potosí, Tamasopo, Puente de Dios, coll. E. Cantoral, 10.i.1998 (FCME PA3780); (5) Aquismón, Tambaque, coll. M. Ramírez, 07.xii.1999 (FCME

PA3874); (6) La Garita, coll. M. Ramírez, 07.xii.1999 (FCME PA3880); (7) Hidalgo, Ixmiquilpan, Tzindejéh, coll. J. Carmona, 17.xi.1994 (FCME PA3554); (8) Morelos, Tlaquiltenango, Los Manantiales, coll. Y. Beltrán, 15.v.2001 (BALE w/n).

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