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***Gloeothece hindakii* (Cyanoprokaryota, Synechococcaceae) – a New Planktonic Species from Lake Tanganyika (Africa)**

By

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With 17 Figures

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Summary

STOYNEVA M. P., GÄRTNER G. & VYVERMAN W. 2009. *Gloeothece hindakii* (Cyanoprokaryota, Synechococcaceae) – a new planktonic species from Lake Tanganyika (Africa). – *Phyton* (Horn, Austria) 48 (2): 199–209, with 17 figures.

During the recent (2002–2004) phytoplankton investigations in the tropical Lake Tanganyika, an abundant cyanoprokaryote alga was found. It is described as a new species – *Gloeothece hindakii* STOYNEVA, GÄRTNER & VYVERMAN (*Synechococcaceae*), which is characterised by: 1) gelatinous lamellated sheaths around single cells and around small colonies of 2–4–6 (–8–16) cells; 2) aerotopes in the cell content; 3) eccentric position of cells within the mucilage before and after transverse cell divisions; 4) daughter cells in one plane; 5) planktonic mode of life.

Zusammenfassung

STOYNEVA M. P., GÄRTNER G. & VYVERMAN W. 2009. *Gloeothece hindakii* (Cyanoprokaryota, Synechococcaceae) – a new planktonic species from Lake Tanganyika (Africa) [*Gloeothece hindakii* (Cyanoprokaryota, Synechococcaceae) – eine neue

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planktonische Art aus dem Tanganyikasee (Afrika)]. – *Phyton* (Horn, Austria) 48(2): 199–209, mit 17 Abbildungen.

Bei Untersuchungen von Phytoplanktonproben aus dem Tanganyikasee 2002 bis 2004 wurde eine häufig vorkommende cyanoprokaryotische Alge gefunden und als neue Art – *Gloeothece hindakii* STOYNEVA, GÄRTNER & VYVERMAN (*Synechococcaceae*) beschrieben. Die neue Art ist durch folgende Merkmale charakterisiert: 1) geschichtete Gallerthüllen um jede Zelle sowie um Zellkolonien aus 2–4–6 (–8–16) Zellen; 2) Aerotope in den Zellen vorhanden; 3) exzentrische Lage der Zellen in der Gallerte vor und nach der transversalen Teilung; 4) Tochterzellen in einer Ebene; 5) planktische Lebensweise.

1. Introduction

Representatives of *Cyanoprokaryota* could be found in almost all habitats and play an important role in the freshwater phytoplankton scenario, where they participate in different functional groups depending on their morphology, ecological and physiological properties (REYNOLDS & al. 2002). One of the most striking features of phytoplankton is the variability in the investment of cells and coenobia in mucilage, which regulates the cell buoyancy and is an efficient protection against grazing and digestion (REYNOLDS 2007). The same is valid of most chroococcal species, which live in colonies of different cell aggregations and various mucilage shapes. The structure of the mucilage and the resulting colony form are diagnostically characteristic of these genera (KOMÁREK & ANAGNOSTIDIS 1999). However, there are still many unresolved taxonomic problems concerning infra-generic and infraspecific taxa of genera like *Gloeothece* NÄG., *Gloeocapsa* KÜTZ. and *Chroococcus* NÄG., where cells are embedded in distinct gelatinous sheaths. Many of their representatives, which look quite similar in different developmental and seasonal stages, undergo taxonomical transformations and have been synonymised in quite different ways by various authors (e.g., GETTLER 1932, GOLLERBAKH & al. 1953, KOMÁREK & ANAGNOSTIDIS 1999, WHITTON 2002). According to the most modern system of cyanoprokaryotes published in 'Süßwasserflora von Mitteleuropa' (KOMÁREK & ANAGNOSTIDIS 1999) the main differences between these genera lie in the mode of cell division and colony formation. The same authors give particular emphasis to the habitat, which means that the species limits are often more restricted than in other floras. The detailed checking of material found recently (2002–2004) in planktonic samples from different sites, depths and seasons from the large tropical, ancient, tectonic Lake Tanganyika in Africa allowed us to follow morphological changes and different developmental stages of one of its most important species, which lives in small mucilaginous colonies.

Cell division by binary fission in a single plain, usually transverse to the longitudinal axis of the cells together with growth to the original size and shape before the next division clearly indicate the affiliation of the material to the family *Synechococcaceae* ANAGNOSTIDIS & KOMÁREK (KO-

MÁREK & ANAGNOSTIDIS 1999). According to the cell length/width ratio, which is less than 3:1, the alga belongs to its subfamily *Aphanothecoideae* KOMÁREK & ANAGNOSTIDIS 1999. The reference to the genus *Gloethece* NAG. is based on the permanent presence of distinct and concentrically lamellate, colorless cell envelopes in the small colonies with low numbers of cells, combined with the possibility for reproduction by disintegration of colonial aggregates and liberation of cells after gelatinization of envelopes (KOMÁREK & ANAGNOSTIDIS 1999, RIPPKA & al. 2001). However, the species' ecology and diagnostic features do not fit to any of the descriptions found in the available literature. The planktonic mode of life in small colonies in different depths in the oligotrophic, tropical lake together with the peculiar eccentric position of daughter cells just after division at the periphery of the distinct and vesicle-enlarged, wide, lamellate but colorless mucilage allow us to describe a new species.

2. Study Site

Lake Tanganyika is situated between 3° 30' and 8° 50' S and 29° 05' and 31° 15' E in a deep narrow trough of the western branch of the Rift Valley of East Africa (COULTER 1994). This tectonic lake with an area of 31 900 km² and a mean depth over 500 m is 650 km long and 50 km wide, and contains three distinct basins – Kigoma, Kungwe and Kipili with maximum depths of 1310, 885 and 1410 m (PLISNIER & al. 1999). Lake Tanganyika is meromictic with anoxic monimolimnion, containing the second largest volume of anoxic water in the world after the Black Sea (PLISNIER & al. 1999). The main limnological characteristics (as yearly medians for the 0–100 m water column) of the lake at both sampling stations Kigoma (Tanzania) in the north and Mpulungu (Zambia) in the south are provided according to PLISNIER & al. 1999 and DESCY & al. 2005:

- 1) water temperature: 25.7 °C at Kigoma and 24.5 °C at Mpulungu, with stronger seasonality in the southern part of the lake;
- 2) pH: ca. 8.9, generally similar at each station;
- 3) conductivity: 654 µS at Kigoma and 662 µS at Mpulungu;
- 4) turbidity: 0.25 NTU at Kigoma and 0.33 NTU at Mpulungu;
- 5) transparency: 12.8 m at Kigoma and 11.9 m at Mpulungu;
- 6) total phosphorus: 16 µg l⁻¹ TRP in PO₄-P, with major increase in upwelling periods in Mpulungu (May–September).

According to DESCY & al. 2005 the average euphotic depth was 38.3 m (range: 25.2 to 55.7 m) in Kigoma compared to 35.2 (range: 13.8 to 65.1 m) in Mpulungu, and was positively correlated with water temperature and water stability.

3. Material and Methods

From February 2002 to January 2005, water column samples were taken fortnightly from two offshore and two littoral stations of Lake Tanganyika: Kigoma

(Tanzania) in the north (04°51.26' S, 29°35.54' E) and Mpulungu (Zambia) in the south (08°43.98' S, 31°02.43' E). In addition, three cruises were organized from Kigoma to Mpulungu, two in the dry season (July 10–17, 2002; July 7–13, 2003) and one in the rainy season (January 30 – February 7, 2004) with eight sampling sites (DESCY & al. 2005). One-litre samples for phytoplankton counts were taken at each 20 m depth and were afterwards settled for sedimentation during 48 hours in the laboratory of TAFIRI at Kigoma and DOF at Mpulungu. The supernatant was removed and the concentrated samples were transferred to 100 ml bottles for transportation. Before counting, the samples were reconcentrated in order to transfer the sample in a 10 ml sedimentation chamber. A drop of Rose Bengal was added to enhance the visibility of the cell content (DESCY & al. 2005). Plankton-net (10 µm mesh width) samples from the upper water column (0–50 m) in the same pelagial and littoral sites were collected also. All samples were stored in the Laboratory of Aquatic Ecology and Protistology of Ghent University.

A Zeiss Axiovert 135 inverted microscope was used to count phytoplankton by the standard method after UTERMÖHL. Detailed investigations of *Gloeotheca* specimens on non-permanent slides were carried-out with a Leitz Diaplan microscope, equipped with Differential Interference Contrast at a magnification of 1000. The standard staining by Indian ink, Lugol's solution, Gentian Violet and Methylene Blue was applied. Digital photographs were taken with an Olympus DP 50 camera.

4. *Gloeotheca hindakii* STOYNEVA, GÄRTNER & VYVERMAN spec. nova

Diagnosis: Cellulae in familias 2-4-6 et raro pluricellulares aggregatae, raro etiam agglomerationes symmetricae e 2-3-5 coloniis formatae. Cellulae 3.5–12 µm latae, 4-14.5 µm longae, late rotundato-ovatae, ellipsoideae vel post divisionem semiglobosae, contentu protoplasmatico cellularum pallide aerugineo-coeruleo vel olivaceo, vesiculis gaseosis (aerotopes). Tegumento proprio gelatinoso, achromatico, homoganeo, distincte lamelloso, etiam colonias (ad 8–12 cellulas) circumdato. Multiplicatio cellularum divisione transversali successive vel raro simultanea ad directionem unam et diffluentia coloniarum. Cellulae post divisionem in tegumento hyalino excentricae locatae.

Habitatio: Species planctonica vivens, laco Tanganyika, Africa.

Iconotypus: Figura nostra 13, ex collectione lacus Tanganyikae (Mpulungu), Africa.

Collectio conservata: deposita in herbario universitatis Gandavae (GENT), sub numero Climcnet 060503.

Description: Small colonies composed of 2-4-6 cells, or rarely more-celled; possible but rare formation of symmetrical agglomerations of 2-3-5 colonies. Cells are 3.5–12 x 4-14.5 µm, broadly ovoid, ellipsoidal or hemispherical (immediately after division), pale blue-green or olive-green, with aerotopes. The mucilage envelope is colorless, firm, distinct and lamellate and is formed around each cell or around colonies smaller than 12-16 cells. Cell division is transverse to the longitudinal cell axis, mainly successive

and rarely simultaneous. Daughter cells occupy a distant and eccentric position at the poles of the mucilage. Daughter cells of the subsequent divisions lie in one plane. Reproduction by disintegration of colonies is common.

Eponymy: The species epitheton *hindakii* is given in honour to the prominent phycologist František HINDÁK from the Slovakian Academy of Sciences (Bratislava).

5. Observations and Discussion

The species appears in almost all investigated quantitative and net phytoplankton samples collected from Tanganyika phytoplankton in the period 2002–2004, but its most pronounced development is during the rainy season (middle of October – beginning of June) in the upper layers (0–20 m, rarely – 40 m). It dominated in the processed quantitative phytoplankton samples from Kigoma at 16.09.2003, 16.10.2003, 28.10.2003, 11.11.2003, 25.11.2003, 11.12.2003, 11.03.2003, 20.05.2003 (at depth 20 m), and was found abundantly in net samples from 08.04.2003, 06.05.2003 and 10.06.2003. It dominated also in the quantitative samples taken during the transversal Cruise nearby Kigoma at Site 1 on 17.02.2004 (at depth 40 m). On 6.05.2003 it was subdominant of *Nitzschia* sp., on 30.09.2003 it was subdominant of the co-dominants *Anabaenopsis tanganyikae* (G. S. WEST) WOLOSZ. & MILLER and *Lobocystis planctonica* (TIFF. & AHLSTR.) FOTT, on 17.04.2004 (at 0 m) it was subdominant of *Closteriopsis petkovii* STOYNEVA & al. In July 2003 it was abundant in net samples from sites 2 and 3 (nearby Kigoma) of the transversal Cruise. The alga was dominant in the processed quantitative samples from Mpulungu on 11.03.2003, 25.03.2003, 10.05.2004 (20 m) and subdominant with *Anabaena* cf. *spiroides* KLEB. on 26.11.2002 (at depth 0 m). In net samples from Mpulungu it was most abundant also on 08.04.2003, 23.04.2003 (at 0 and 20 m), 06.05.2003, 15.07.2003, 20.04.2005 and 04.05.2004. In spite of being small-sized and abundant, this alga was never observed ingested in zooplankters, in contrast with other mucilage-bearing algae in Tanganyika (STOYNEVA & al. 2008).

After processing of phytoplankton counts in the inverted microscope, this alga was published for the Tanganyika plankton under the name *Gloeocapsa decorticans* (A. BRAUN) RICHTER in WILLE (DESCY & al., 2005) due to fitting of the features of small (2–4 celled) colonies with the description provided in KOMÁREK & ANAGNOSTIDIS 1999 and with their figure 319. In the notes on the occurrence it was indicated: ‘Subaerophytic, on wet rocks, walls, known also from caves, data from ‘stagnant water’ should be checked...’ (KOMÁREK & ANAGNOSTIDIS 1999: 246). Exactly this un-conformity to ecology was the reason to rework the material on qualitative slides with higher magnifications and to check again all our notes, mea-

surements, drawings and photos. As a result of this work, due to confirming the different mode of cell division in one plane it was decided to correct the previous identification.

The alga forms colonies, which are usually small – 15-22-(38.5) μm , composed of sheathed cells or groups of cells (Fig. 1-13, 15-17). They consist mainly of 2-4-6 cells, or very rarely are more-celled (Fig. 8, 9). Agglomerations of 2-3-5 colonies with irregular or very rarely regular shape (Fig. 8) have been observed but generally colonies appeared separately (Fig. 3, 6).

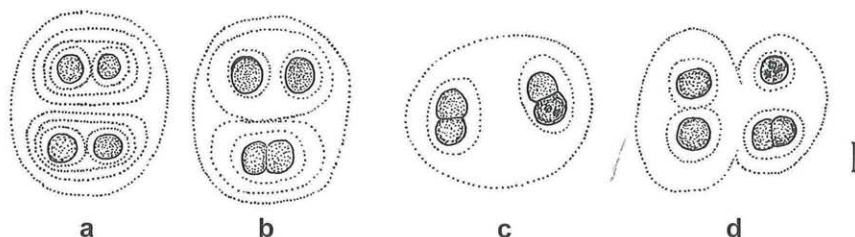


Fig. 1. *Gloeotheca hindakii* STOYNEVA, GÄRTNER & VYVERMAN – colonies in different stages of division. – Scale bar – 5 μm .

There is no common distinct mucilage envelope around colonies bigger than 12-16 cells or around the agglomerations (Fig. 7, 8), which, most probably is due to rapid reproduction by disintegration of colonies. The staining by Gentian Violet, Methylene Blue and Indian ink did not reveal any diffuent gelatinous envelope around the same colonies and aggregations (Fig. 2, 3, 5, 6, 7).

The mucilage is colorless, but firm, distinct and lamellate. New individual sheaths are formed immediately after the separation of the daughter cells. In colonies the number of layers usually is 2-4 (Fig. 1a, b, 2, 3, 5, 12, 14). Coloration by Gentian Violet and Methylene Blue makes the layers more pronounced and better visible, but does not reveal any other peculiarities in their structure (Fig. 2, 3, 5). Empty lamellate sheaths remained in the plankton long after cell death.

The development starts from a sheathed solitary, spherical or broadly oval cell (Fig. 5, 14), which elongates before division. Cells are (3.5)-3.8-7-(12) $\times$ (4-5)-7-9.5-(14.5) μm , spherical to broadly oval, ellipsoidal or hemispherical (immediately after division), pale blue-green or olive-green. The length/width is always less than 3:1 and is mainly 1.3-1.4 (e.g. $5.1/3.8 = 1.3$, $6.4/4.8 = 1.3$, $9/6.4 = 1.4$). The presence of aerotopes is typical but is generally seen after application of immersion oil or staining (Fig. 7, 9, 11, 15).

The cell division is transverse to the longitudinal cell axis and is running mainly in a successive way (Fig. 1b, d, 3, 4, 16, 17), but could appear also simultaneously (Fig. 1a, c, 2, 6, 9, 11, 15). Most peculiar is the distant

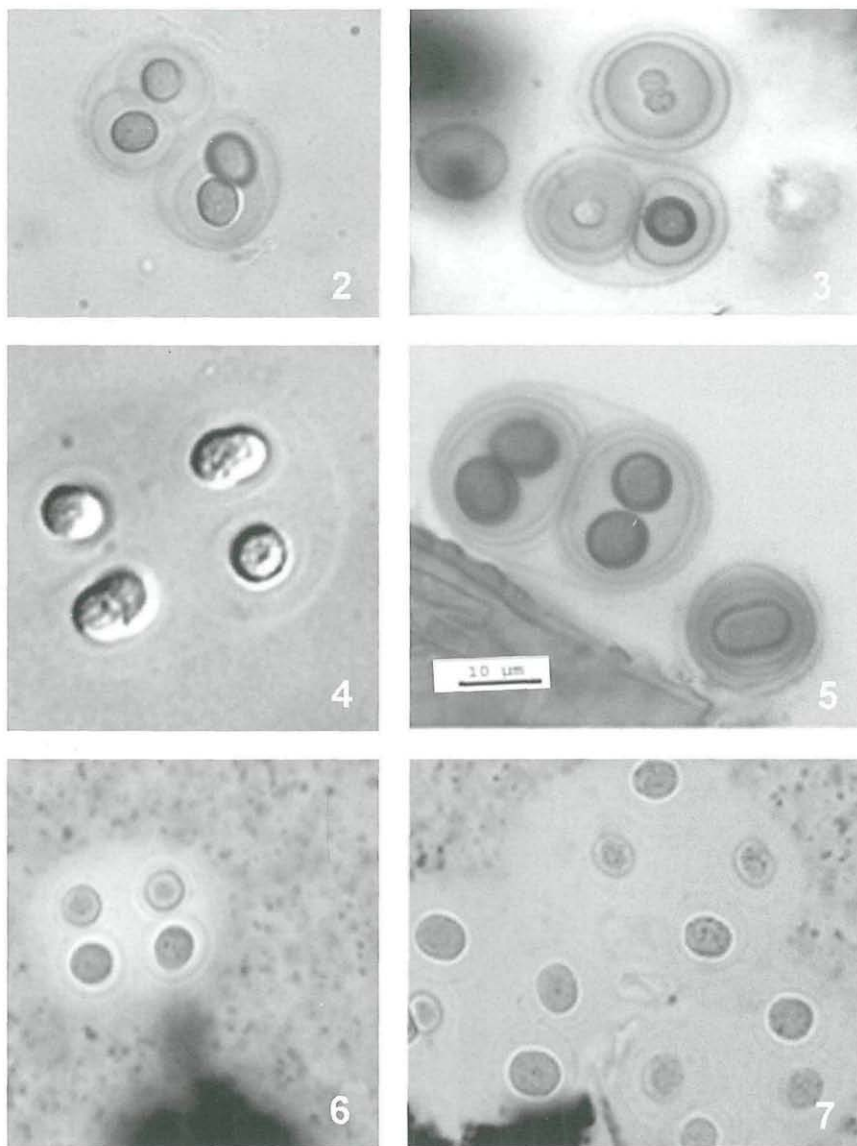


Fig. 2-7. *Gloeotheca hindakii* STOYNEVA, GÄRTNER & VYVERMAN in quantitative samples prestained by Bengal Rose (except 4): Fig. 2 after additional staining by Gentian Violet; Fig. 3, 5 after additional staining by Methylene Blue; Fig. 4 transversal successive cell division; Fig. 6, 7 after additional staining by Indian ink. – Scale bar – 10 µm.

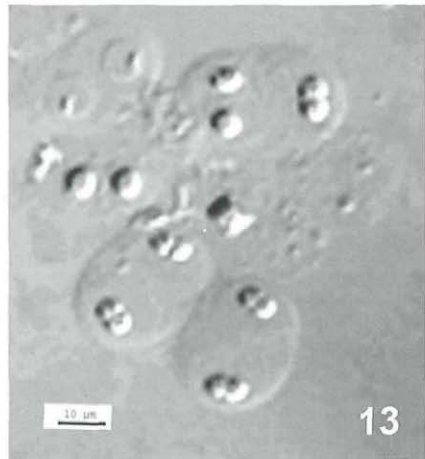
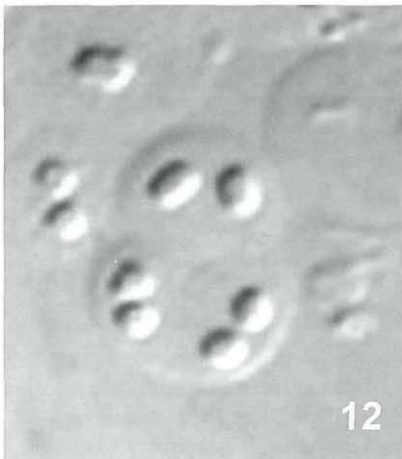
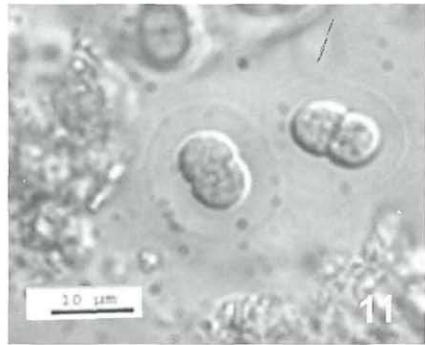
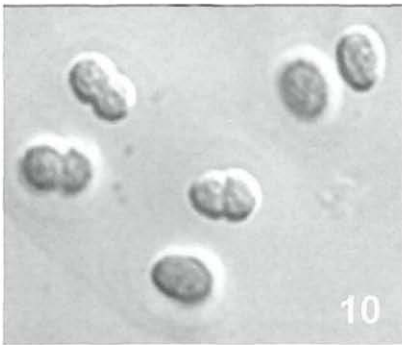
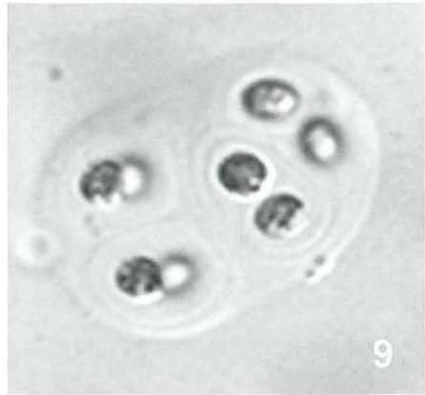
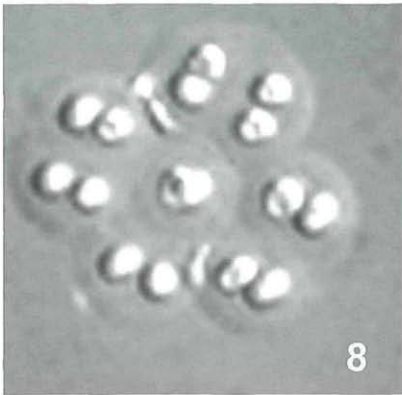


Fig. 8-13. Agglomerations of colonies of *Gloeotheca hindakii* STOYNEVA, GÄRTNER & VYVERMAN in net samples (Fig. 9 – after staining by Lugol's solution, cells with visible aerotopes). Fig. 8-11 taken with magnification 1000x and oil immersion and Fig. 12 and 13 taken at magnification 400x. – Scale bar equal 10 µm in Fig. 11 also valid for Fig. 8-10, in Fig. 13 also for Fig. 12.

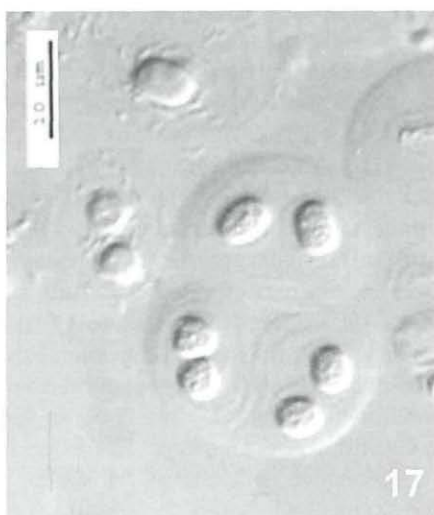
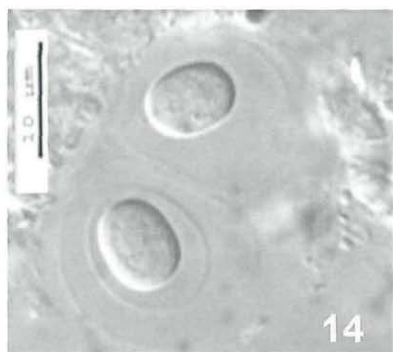


Fig. 14–17. *Gloeotheca hindakii* STOYNEVA, GÄRTNER & VYVERMAN in quantitative and net samples with eccentric position of cells in the common mucilage before and during cell division. Fig. 14 taken with magnification 1000x and oil immersion and Fig. 15–17 taken at magnification 400x. – Scale bar equal 10 μ m in Fig. 17 also valid for Fig. 15 and 16.

and eccentric position of the daughter cells at the poles of the mucilage (Fig. 1c, 13, 16, 17). It is preceded by the eccentric position of both initial cells before division (Fig. 14). On first glimpse, when seen separately on smaller magnifications (Fig. 13), such single or double colonies with daughter cells even resemble some stages of *Epigloeosphaera glebulenta* (ZALESSKY) KOMÁRKOVÁ-LEGNEROVÁ or *Aphanothece microscopica* NÄGELI (KOMÁREK & ANAGNOSTIDIS 1999: Fig. 46d, e, 72e). However, all other features and careful investigation of the material clearly separate the new taxon from the representatives of these genera.

The presence of wide, distinct, lamellate and vesicle-enlarged sheaths with sharply delimited margins resembles *Gloeocapsa* and particularly the two-cell stages of *Gloeocapsa rupestris* KÜTZING as illustrated on Fig. 337e in KOMÁREK & ANAGNOSTIDIS. The clearly distinguishing characters are the lack of common sheath for bigger colonies in *Gloethece hindakii*, the mode of reproduction (binary fission in three perpendicular planes in successive generations in *Gloeocapsa*) and the subaerophytic way of life of *G. rupestris* on periodically wetted rocks and walls (KOMÁREK & ANAGNOSTIDIS 1999). The ecological peculiarities comprise one of the most important diagnostic features according to the most modern cyanoprokaryote system of KOMÁREK & ANAGNOSTIDIS 1999. Additionally, the lack of obvious gas vesicles (and hence aerotopes) in the genus *Gloeocapsa* is noted by KOMÁREK 2003.

The initial stages and colonies of 2–4 cells of *Gloethece hindakii* resemble the same stages of *Gloethece rupestris* (LYNGBYE) BORNET in WITTRÖCK & NORDSTEDT, *Chroococcus obliterated* RICHTER and *Chroococcus minutus* (KÜTZING) NÄGELI as illustrated in KOMÁREK & ANAGNOSTIDIS 1999 on Fig. 97b, 391j and 394c. However, there are clear differences with *Gloethece rupestris*, which lie in the presence of a distinct lamellate mucilage around colonial agglomerations and in the ecological peculiarities and habitat characteristics (aerophytic and atmophytic, on wet rocks and wall, sometimes on mosses, mainly in mountains) – KOMÁREK & ANAGNOSTIDIS 1999. The differences to *Chroococcus obliterated*, which is known from littoral of lakes and ponds, lie in the eventual presence of common diffluent wide mucilage around the colonies with thin, usually non-lamellate (or slightly lamellate) envelopes, following the outline of inner cells and, most important, in the mode of division – in at least three planes in successive generations (KOMÁREK & ANAGNOSTIDIS 1999). Additionally, KOMÁREK 2003 noted the possibility for irregular division for the whole genus *Chroococcus* but did not indicate it in the description of *Gloethece*. All differences mentioned above are valid for a clear distinction from *Chroococcus minutus*, for which the usual lack of lamellate and delimited envelopes is outlined (KOMÁREK & ANAGNOSTIDIS 1999). Taking into account all the outlined peculiarities, a misinterpretation of *Gloethece hindakii* among these species is likely impossible.

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