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Mallomonas teres, sp. nov. (Chrysophyceae), simultaneously revealed in two distant European peat-bog regions

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ABSTRACT
We present the description of *Mallomonas teres*, sp. nov., from three localities, two peat-bogs in Northern Sweden and a peat-bog pool in the Polessian Nature Reserve, Northern Ukraine. The sites ranged from acidic to neutral pH, were low in conductivity, and contained other pH-indifferent or low-pH chrysophyte taxa. According to the scale structure *M. teres*, sp. nov., belongs to the section *Heterospinae* within the genus. The most distinctive scale character discerning *M. teres*, sp. nov., from the other species of the section *Heterospinae* is the distal part of the scale, which has a smooth dome and numerous parallel curved ribs. *Mallomonas teres*, sp. nov., is probably distributed at least in temperate, acidic European localities. Its rare occurrence could be a result of a poor dispersal capacity combined with a low growth rate.

KEY WORDS
Chrysophyceae, *Mallomonas teres*, silica-scaled chrysophytes, Synurales, new species.

RÉSUMÉ
Mallomonas teres, sp. nov. (Chrysophyceae), découverte dans deux régions européennes distinctes de tourbières.
Nous présentons la description de *Mallomonas teres*, sp. nov., issue de trois localités, deux tourbières dans le nord de la Suède et un bassin de tourbières dans la réserve naturelle de Polessian, dans le nord de l'Ukraine. Les sites s'échelonnent de pH acide à neutre, ont une conductivité basse et contiennent d'autres taxa de chrysophytes, à pH indifférent ou à pH bas. Selon la structure de l'écaille, *M. teres*, sp. nov., appartient à la section *Heterospinae* du genre. Le caractère d'écaille le plus distinctif distinguant *M. teres*, sp. nov., des autres espèces de la section *Heterospinae* est la partie distale de l'écaille, qui présente un dôme lisse et de nombreuses côtes incurvées parallèles. *Mallomonas teres*, sp. nov., est probablement répartie au moins dans des localités européennes tempérées et acides. Ses rares occurrences pourraient être le résultat d'une faible capacité de dispersion combinée à un faible taux de croissance.

MOTS CLÉS
Chrysophyceae, *Mallomonas teres*, écaille de silice chrysophytes, Synurales, espèce nouvelle.

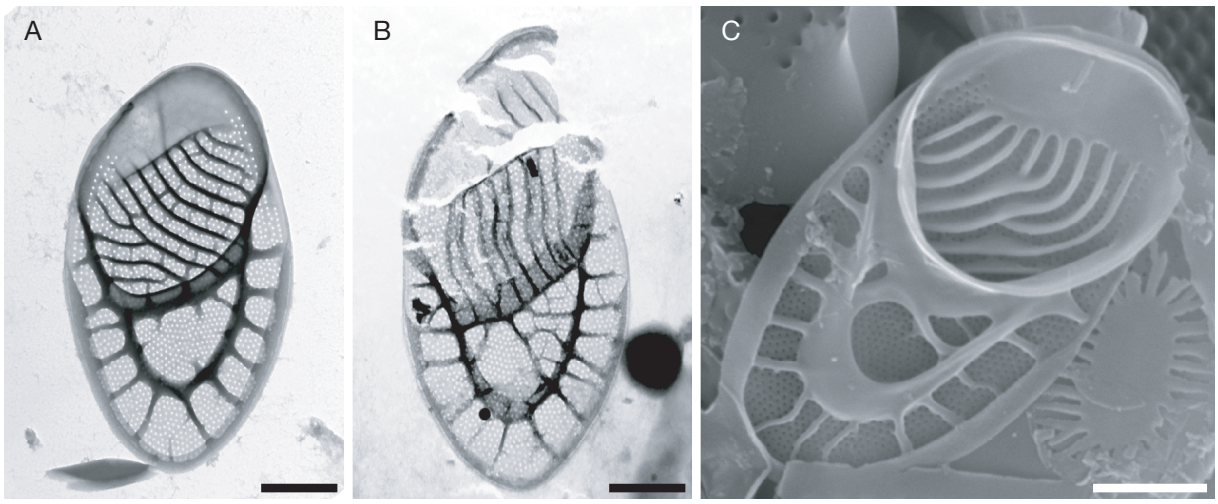

FIG. 1. — Body scales of *Mallomonas teres*, sp. nov.: **A**, **B**, scales observed in a transmission electron microscope; **C**, a scale observed in a scanning electron microscope. Scale bars: 1 µm.

INTRODUCTION

The genus *Mallomonas* was erected by Perty (1852). The suffix *-monas* was used in the early history of microbiology to denote unicellular organisms. Later, in descriptions of eukaryotic microorganisms, *-monas* was used for flagellated unicells with either one or two flagella (Boenigk 2008), e.g. *Chlamydomonas* (Chlorophyceae), *Ochromonas* (Chrysophyceae), *Cryptomonas* (Cryptophyceae). In the chrysophyte *Mallomonas*, the cell is covered by a highly organised case composed of siliceous scales (*c.* 4 µm long); some species possess also bristles attached to the scales. The taxonomic importance of scale and bristle morphology was recognised by Ivanov (1899). Almost one hundred years later electron microscopy provided an excellent tool to observe the detailed scale ultrastructure and promoted the description of numerous new species, which were classified into sections and series based on the morphology of the siliceous structures. The last updated classification was published by Kristiansen & Preisig (2007). In light of modern molecular methods, scale morphology proves to be a good marker for species delimitation, and most of the Sections have been justified (Siver *et al.* 2015). Molecular studies set the species to the broader phylogenetic frame, and time-calibrated five-gene phylogeny demonstrated that *Mallomonas* split into two major clades during the Early Cretaceous, one containing species with scales possessing a V-rib and the other with scales lacking it. The V-rib represents an evolutionary novelty that enabled more accurate spacing and organisation of scales within the scale case (Siver & Glew 1990; Siver *et al.* 2015). On the other hand, molecular data can provide a tool for the detailed differentiation of species with very similar scale morphology, e.g. pseudocryptic species within the *Mallomonas matvienkoae* group (Jo *et al.* 2013) or the *Mallomonas kalinae/rasilis* species complex (Gusev *et al.* 2018). However, only less than 20% of the described species have been sequenced to date, and we have to rely on scale morphology to place the taxa (including the fossil ones) into the modern system. Similar to the other protist groups, species diversity within *Mallomonas* is far from being fully described, and new species are constantly being recorded from all over the world (e.g. Nemcova *et al.* 2011; Gusev *et al.* 2016; 2017). Here, we introduce a new species *Mallomonas teres*, sp. nov., which was revealed almost simultaneously from two peat-bogs in Northern Sweden and a peat-bog pool in the Polessian Nature Reserve, Northern Ukraine.

MATERIALS AND METHODS

The Swedish sites were located on the western shore of the Gulf of Bothnia, near the Quark region. Both sites, Gärdvikmyrorna (63.48733°N, 19.71048°E) and Bärmyran (63.51733°N, 19.71472°E), were peat-bogs containing deeper pools, situated relatively close to the sea (2270 and 450 m, respectively). The distance between the sites was approximately 2 km. The bedrock of this area is mostly of sandstone (wacke), with metamorphic schists and paragneiss (Geological Survey of Sweden; SGU 2018). The area has a humid subarctic climate with severe winters and cool short summers, with a strong seasonality (average annual temperature 3°C, average annual precipitation 601 mm). Both sites are located in the boreal wet forest biome (ClimaTemps 2012).

The Ukrainian site was situated in the Zhytomyr Region in the territory of the Polessian Nature Reserve. It is a peat-bog pool (51.53503°N, 28.08763°E) located near a forest road in block 32 of the Selezivka forestry, overgrown with *Sphagnum* sp. The area has a humid continental climate with warm humid summers and temperate severe winters (average annual temperature 6.8°C, average annual precipitation 682 mm; Balabukh *et al.* 2013).

The Swedish material was obtained by a combination of plankton samples (20-µm mesh net) and water squeezed from the submerged vegetation. Selected environmental variables were measured at the collection site (pH, conductivity and

surface water temperature; pH-conductometer WTW 340). Drops of the fresh samples were dried onto Formvar-coated transmission electron microscope (TEM) grids. Dried material was washed by repeated transfer of the grid into drops of deionised water dispensed on the hydrophobic surface of a Parafilm stripe (see Novakova *et al.* 2004). The TEM grids were examined with a JEOL 1011 TEM. Photomicrographs were obtained using a Veleta CCD camera equipped with an image analysis software (Olympus Soft Imaging Solution GmbH).

The Ukrainian sample was obtained by squeezing water from *Sphagnum* sp. and subsequently fixed with 4% formaldehyde solution. For scanning electron microscopy (SEM), an aliquot of the sample was initially washed by repeated centrifugation in deionised water to remove fixative and digested for 4-5 minutes in sulphuric acid with potassium dichromate, followed by 3 washings. Subsequently, several drops were placed on aluminium stubs, coated with gold for 10 min and observed with a JEOL 6510 LV SEM.

RESULTS

The microphotograph of a single scale of *M. teres*, sp. nov., was published in Nemcova *et al.* (2016) as *Mallomonas* sp. The other record of the species in Ukraine strengthened our intention to formally describe this new species. *Mallomonas teres*, sp. nov., was sampled from acidic to neutral peat-bog pools low in conductivity (Gärdvikmyrorna pH 5.1, conductivity 20 µS cm^{-1}; Bärmyran pH 6.3, conductivity 25 µS cm^{-1}).

SYSTEMATICS

Mallomonas teres Nemcova & Kapustin, sp. nov.
(Fig. 1)

Holotype specimen. — Single scales on a TEM grid deposited at the Culture Collection of Algae of Charles University, Prague, H2018/1. Fig. 1A shows a representative scale from the specimen.

Type locality. — Sweden, Wästerbotten County, Gärdvikmyrorna peat-bog, (63.48733°N, 19.71048°E), 20 m a.s.l., sample collected by Y. Nemcova M. Skaloudova M. Pusztai and J. Neustupa, 3.VI.2012.

Etymology. — The epithet refers to the rounded distal part of the scale, from Latin "teres" (rounded, rotund).

Habitat. — Small peat-bog pool.

Distribution. — Sweden, in addition to the type locality, the species was also found in the Bärmyran peat-bog (63.51733°N, 19.71472°E); Ukraine, mire pool in block 32 of Selezivka forestry in the Polessian Nature Reserve (51.53503°N, 28.08763°E).

Description

Body scales 4.9-5.1 × 2.7-3.2 µm, with evenly distributed pores on the basal plate, except for the dome area (Fig. 1). Posterior rim is encircling half of the perimeter of the scale. A continuous submarginal rib originates in the posterior portion of the scale and curves toward and the dome. Near the middle of the scale, a thick, rounded transversal rib connects the lateral parts of the submarginal rib. A secondary reticulum of smaller meshes with more delicate ribs may be developed under the transversal rib (Fig. 1B). There are parallel ribs radiating at right angles from the submarginal rib. The distal part of the scale is circumscribed by the thin raised structure formed by distal parts of the submarginal rib and the rounded transverse rib (pocket-like). Within this area, there is a dome and numerous (8-12) parallel curved ribs, sometimes anastomosed or interconnected by subtle lateral struts (Fig. 1A). The dome is broad and smooth, with no typical U-shaped structure. The curved ribs of the distal portion of the scale terminate near the dome (Fig. 1C).

Bristle was not observed. Stomatocyst unknown. Cell shape and dimensions unknown.

DISCUSSION

Mallomonas teres, sp. nov., was reported from two relatively distant regions, although still within Europe. The Swedish sites are more than 1400 km away from the Ukrainian one. The same situation was observed for the rare species *M. phasma* Harris & Bradley, which was originally described from southeast England (Harris & Bradley 1960) and later reported from Aquitaine, France (Nemcova *et al.* 2012). *Mallomonas koreana* Kim & Kim was reported from only two, even more distant sites: a reservoir in South Korea and a lake in Aquitaine (Kim & Kim 2008; Nemcova *et al.* 2012).

The distribution of microbial diversity results from the interplay of a complex set of variables including historical (e.g. colonisation and extinction rates, dispersal limitation) and contemporary environmental processes (Potapova & Charles 2002; Vyverman *et al.* 2007; Siver & Lott 2012). Among the environmental variables, pH and conductivity have been considered to play important roles in structuring silica-scaled chrysophyte communities (Siver 1991; Siver 1995; Nemcova *et al.* 2016). All three sites where *M. teres*, sp. nov., was found represent mire/peat-bog pools, relatively low in conductivity and pH. Climate-related factors (e.g. precipitation or seasonability) may also be crucial in the distribution of silica-scaled chrysophytes on larger (e.g. continental) scales (Nemcova, unpublished). For example *M. tropica* Dürrschmidt & Croome represents species clearly restricted to warm tropical regions rich in precipitation (Dürrschmidt & Croome 1985; Gusev 2013).

According to the Köppen-Geiger climate classification, both studied regions (Swedish and Ukrainian) have a cold continental humid climate, differing mostly in average summer temperatures (Peel *et al.* 2007); therefore, they may host a similar pool of silica-scaled chrysophytes including *M. teres*, sp. nov., Kristiansen (2001; 2008) reported that about one third of the species described so far are endemic, i.e. restricted to a limited region (continent). Actually, most newly described species are initially considered to be endemics, due to limited sampling. It is interesting that endemic taxa are often not confined to any distant or isolated habitats with specific

environmental conditions. On the contrary, many initially endemic species have later been revealed in well-investigated areas (Kristiansen 2008), together with more common species. This was the case for *M. teres*, sp. nov., which was uncovered in peat-bog pools with other pH-indifferent or low-pH chrysophyte taxa (Nemcova *et al.* 2016, Supplementary Material). It is likely that *M. teres,* sp. nov., is not restricted to Europe, and therefore, its endemic "life span" probably depends on the intensity of chrysophyte research worldwide.

Mallomonas teres, sp. nov., is probably distributed at least in other temperate, acidic European localities. Its rare occurrence could be a result of a poor dispersal capacity combined with a low growth rate. Small populations are thus below the limit of detection in standard chrysophyte investigations. However, the possibility that *M. teres*, sp. nov., requires some special chemical substances for its life cannot be completely excluded.

According to the scale structure, *Mallomonas teres*, sp. nov., belongs to the section *Heterospinae* Momeu & Péterfi (1979). Members of this section have obovate scales without lateral incurvings, a continuous oval-shaped submarginal rib from which other secondary ribs radiate, a conspicuous posterior rim, a base plate with regularly spaced pores and a simple or complicated reticulum formed by secondary ribs. The dome is usually shallow; however, its shape and structure may vary between species (Siver 1991; Kristiansen 2002). The continuous oval-shaped submarginal rib serves to space the overlapping scales within the scale-case and to strengthen the scale; it is thought to be a homologous structure to the V-rib (Siver *et al.* 2015). The species most similar to *Mallomonas teres*, sp. nov., is *M. pugio* Bradley (1964). Scales of both species possess the continuous submarginal rib, but they differ in the appearance of a distal part of the scale. *Mallomonas pugio* has a distal part with one or two strong longitudinal ribs with struts, while *M. teres*, sp. nov., has 8-12 parallel curved ribs encircled by a pocket-like structure, formed by the rounded transverse rib. *Mallomonas pugio* has a dome with 5-8 parallel ribs, delimited proximally by a strong rib. The dome of *M. teres*, sp. nov., is broad and smooth, not bulging from the rest of the scale's distal part. *Mallomonas teres*, sp. nov., also closely resembles *M. temonis* Nemcova & Kreidlova (2013). The scales of both species are similar, with a continuous submarginal rib connected by a single transversal rib. However, the apical part of the *M. temonis* shield is almost without any further structures. The U-shaped dome is small and oval, whereas in *M. teres*, sp. nov., the dome is large, broad and smooth. The scales of *M. harrisiae* Takahashi (1975) are distinguishable from those of *M. teres*, sp. nov., in that the system of primary ribs of the shield is more elaborated and their rounded dome is marked with longitudinal ribs (Takahashi 1975). To conclude, the most important scale character discerning *M. teres*, sp. nov., from the other species of the section *Heterospinae* is a distinctive distal part with numerous parallel curved ribs, coupled with a broad and smooth dome.

Much protist diversity remains to be described. *Mallomonas teres*, sp. nov., has an unmistakable scale morphology that makes it easily recognised, and its description contributes to our knowledge of synurophyte diversity.

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