



Two new species of *Mallomonas* (Chrysophyceae: Synurales): *Mallomonas temonis* and *Mallomonas divida*

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Abstract

This paper describes two new species of *Mallomonas*. *Mallomonas temonis* was identified in the Table Mountain Plateau, South Africa; *Mallomonas divida* was identified in Aquitaine, France. The descriptions are based on silica-scale morphology studied by means of transmission and scanning electron microscopy. *Mallomonas temonis* belongs to the section *Heterospinae*. A scale is surrounded by a submarginal rib connected at lateral parts by a strong, more or less transversal rib. *Mallomonas divida* belongs to the section *Torquatae*. The shield of scales is covered by regularly arranged, delicate papillae and marked by one or several rows of more distinctive papillae.

Introduction

Synurales, including *Synura* Ehrenberg 1834: 314 and *Mallomonas* Perty 1852: 170, are often important components of the phytoplankton community of freshwater lakes and reservoirs. *Mallomonas* represents the most diversified genus of the group (Siver 1991). Currently, it is comprised of approximately 280 taxa; however, only 163 taxa are accepted taxonomically (Guiry & Guiry 2012). The cell body of the flagellates is covered with an armor of imbricated silica scales. The scales represent flat, bilateral, almost symmetrical objects with dimensions of 2.5–6.0 × 1.5–4.0 μm in most species. In *Mallomonas*, some or all scales may harbor a bristle. The oldest record for the genus dates to ~40 million years ago, and is represented by numerous *Mallomonas* scales and bristles discovered in Eocene maar lake sediments (Siver & Wolfe 2005). Siver & Wolfe (2005) reported several extinct species along with scales significantly similar to contemporary taxa, and they concluded that the scale morphology is an old and reliable morphological character. *Mallomonas* also represents the flag ship genus to study biogeography and to evaluate the neutral dispersal model in protists (Řezáčová & Neustupa 2007).

Early classification of *Mallomonas* was based on characteristics like cell size and shape, colonial versus solitary habitat, number of visible flagella, number of chloroplasts, scale outline, and position of bristles visible under a light microscope (Starmach 1985). The introduction of electron microscopy into silica-scaled chrysophyte research was initially performed by Fott (1955) and Asmund (1955), and it revolutionized the *Mallomonas* taxonomy. *Mallomonas* is considered to have one of the best morphological species concepts within Stramenopiles. The taxonomically diagnostic morphology of scales and bristles enables this concept of morphospecies to be applied reproducibly. Intensive investigation resulted in description of numerous new species and several recently published monographs (Siver 1991, Kristiansen 2002, Kristiansen & Preisig 2007). Several analyses using molecular markers were performed to set a baseline for understanding of synurophyte phylogeny (Lavau *et al.* 1997, Andersen 2007); however, the genus *Mallomonas* was shown to be polyphyletic in the *rbcL* phylogenetic tree, and it was not resolved based on nuclear SSU rDNA data (Andersen 2007). The most recent study examined the phylogenetic relationship among *Mallomonas* species isolated from Korean localities using scale morphology combined with nuclear SSU and LSU rDNA and *rbcL* genes (Jo *et al.* 2011).

Although new species of *Mallomonas* are continuously described (e.g., Kim & Kim 2008, Němcová *et al.* 2011), the real diversity of the genus is unknown and probably strongly underestimated. The purpose of this study is to describe two additional species, *Mallomonas temonis* and *M. divida*, from Western Cape (South Africa) and Aquitaine (France), respectively. Two new species, *Mallomonas solea-ferrea* Němcová & Kristiansen in Němcová *et al.* (2011: 378) and *M. siveri* Němcová & Kristiansen in Němcová *et al.* (2011: 379), have been described recently from Western Cape (Němcová *et al.* 2011). Water-bodies in Aquitaine have been investigated recently and a high percentage of all known synurophyte species, including the rare ones, was revealed (Němcová *et al.* 2012).

Materials and Methods

This paper is based on either surface sediment samples fixed with Lugol's solution (*Mallomonas temonis*) or on freshly collected plankton and surface sediment samples (*M. divida*). Surface sediment samples were obtained by syringe suction from the upper sediment layers. Plankton samples (20 µm mesh net) were combined with water squeezed from the submerged vegetation. Selected environmental variables were measured at the collection site of *M. divida* (pH, conductivity and surface water temperature; pH-conductometer WTW 340). The Lugol-fixed samples were washed by repeated centrifugation in deionized water. Drops of the fresh or washed samples were dried onto Formvar-coated transmission electron microscopy (TEM) grids. The TEM grids were examined with a JEOL 1011 transmission electron microscope. Photomicrographs were obtained using a Veleta CCD camera equipped with image analysis software (Olympus Soft Imaging Solution GmbH). For scanning electron microscopy (SEM), the Formvar-coated grid (already observed in TEM) was mounted onto an SEM stub with double-sided adhesive carbon tape, coated with gold for 5 min (forming a 3 nm layer) with a Bal-Tec SCD 050 sputter coater, and observed with a JEOL 6380 LV scanning electron microscope. Typification of the new species is based on the illustrations hereby published, as it was impossible to preserve specimen showing the features attributed to the taxon (ICN art. 40.5).

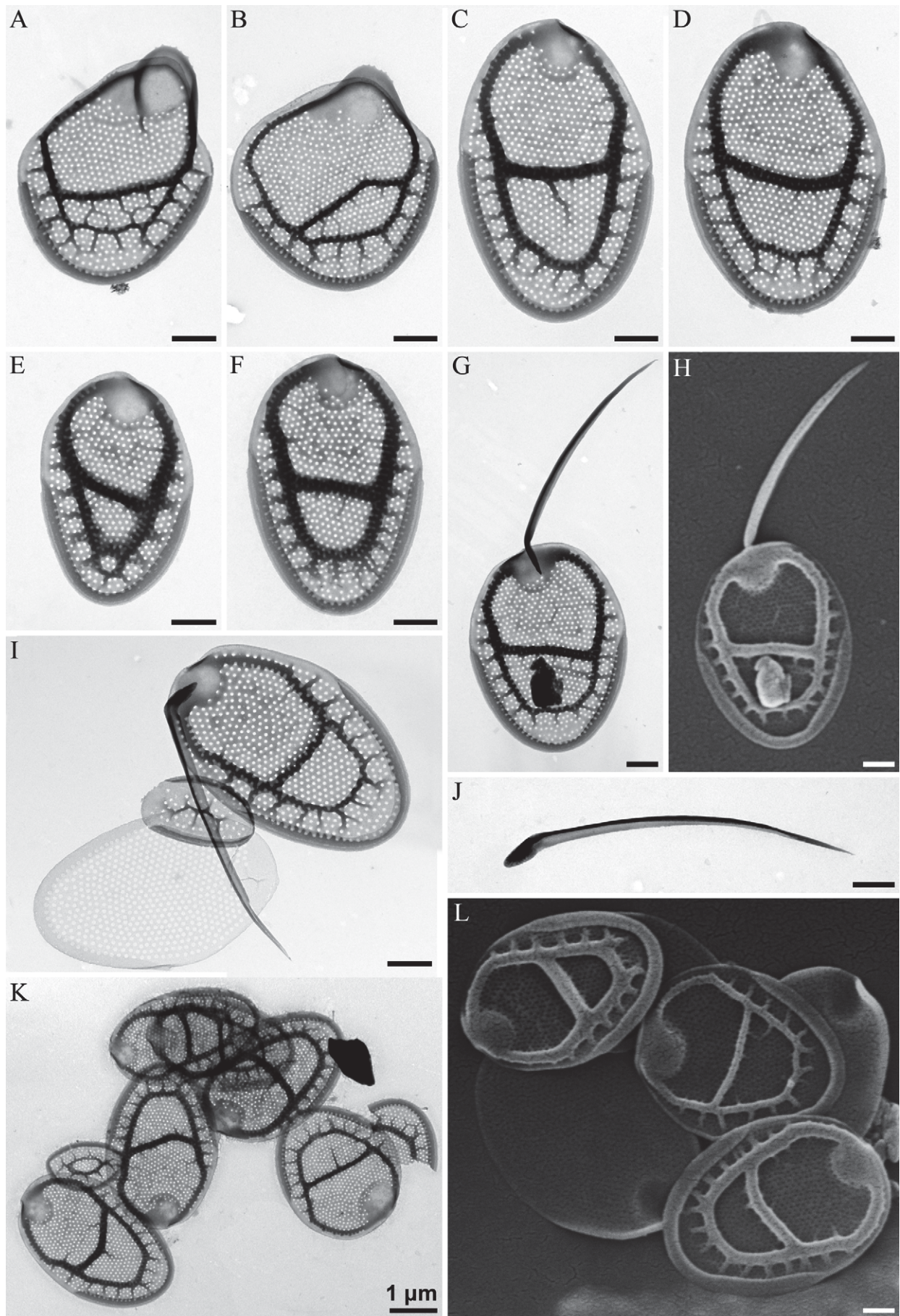
Taxonomy

Mallomonas temonis Němcová *sp. nov.* (Figs 1A–L)

Type:—SOUTH AFRICA. Western Cape: unnamed small pool at Table Mountain, 33° 57.499' S, 18° 24.282' E, 1064 m a.s.l., sample collected by Y. Němcová, 20 August 2010. Figure 1D.

Three types of scale may be discerned including apical, body, and rear scales; however, a continuous transition between body and rear scales exists. Apical scales (2.6–3.2 × 1.9–2.5 µm) are broad and remarkably asymmetric, with one lateral edge shorter than the other. A large, broad, flat dome is eccentrically situated and its posterior part is extended, which forms a lip. The base plate is marked with closely spaced pores, which are clearly visible on the less silicified scale (Fig. 1I). A scale is surrounded by a submarginal rib curving toward

FIGURE 1. *Mallomonas temonis*. A–B, asymmetric apical scales with an eccentrically situated dome; C–D, oval body scales; E–F, rear scales; G–H, a body scale bearing a bristle; G, a scale viewed in a transmission electron microscope; H, the same scale viewed in a scanning electron microscope (SEM); I, a body scale with a bristle anchored to the dome, a rounded rearmost scale and a less silicified scale marked with closely spaced pores; J, a slightly curved bristle with a sharp tip; K, a group of body scales including one rearmost scale; L, a group of body scales viewed in SEM. Note that the side of the scale adjacent to the plasma membrane is smooth. Bar = 0.5 µm, if not stated otherwise. A–F and I are shown at the same magnification.



the dome. At the posterior part of the scale, a strong, more or less 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. 1A). The submarginal areas possess struts (occasionally bifurcated) radiating from the submarginal rib; the struts do not reach the proximal border. The proximal border surrounds approximately half of the scale outline (Figs 1A–B). Body scales ($3.2\text{--}3.9 \times 2.1\text{--}2.7 \mu\text{m}$) are oval and essentially bilaterally symmetrical (Figs 1C–D). The submarginal rib fuses with the dome forming the continuous entity (Figs 1H, 1L). The strong transversal rib crosses the shield in approximately the lower half to third (Figs 1K–L). Remnants of the reticulum of smaller meshes are sometimes visible on the shield (Fig. 1G). Rear scales ($2.1\text{--}2.7 \times 1.4\text{--}1.9 \mu\text{m}$) are smaller and more rhomboidal compared to body scales (Figs 1E–F). The rearmost scales ($1.4\text{--}1.6 \times 0.8\text{--}1.1 \mu\text{m}$) are oval, lack a dome, and have a rudimentary reticulum formed by delicate ribs. Bristles ($3.2\text{--}3.5 \mu\text{m}$) are smooth, slightly curved and attenuated, with a sharp tip. The foot of the bristle is thin (Figs 1G–J). Only one type of bristle was observed. Cyst unknown. Cell shape and dimensions unknown.

Habitat:—Small, humic pool.

Distribution:—South Africa, the species was only found in the type locality.

Etymology:—The epithet refers to the strong transversal rib, from Latin “temo” (pole or beam).

Mallomonas divida Němcová & Kreidlová *sp. nov.* (Figs 2A–K)

Type:—FRANCE. Aquitaine: unnamed lake south of the Reserve Naturelle Marais d'Orx, $43^\circ 34.398' \text{ N}$, $1^\circ 23.464' \text{ W}$, 23 m a.s.l., sample collected by Y. Němcová & J. Kreidlová, 27 March 2011. Figure 2E.

Collar, body, and rear scales are discerned. Collar scales, surrounding the flagellar pore ($3.1\text{--}3.3 \times 1.9\text{--}2.1 \mu\text{m}$), are of trapezoid shape and relatively broad at the base (Figs 2A–D). The large and suboval dome has a small peak that rises as the extension of the dorsal submarginal rib. The dome is partly reticulated by circular meshes and possesses a ridge dorsally marked with delicate struts (Figs 2B–C). The submarginal rib is distinctly developed at the dorsal part of the scale. The flange is smooth, forming a prominent elongated protrusion on the dorsal edge of the collar scale adjacent to the dome ridge. A proximal border circulates the posterior and dorsal parts of the scale. The submarginal rib is smooth and less prominent at the ventral part of the scale. The shield of the scale is covered by more or less regularly arranged delicate papillae, and divided by one or several rows of more distinctive papillae (Fig. 2D). At the articulation, the submarginal rib window (depression without secondary layer) possesses the row of 3–5 pores. Body scales ($2.6\text{--}2.9 \times 1.7\text{--}2.0 \mu\text{m}$) are rhomboid and domeless (Figs 2E–H). The posterior submarginal rib is not prominent; it is more flattened and smooth. There is no anterior submarginal rib; the anterior flange and shield are not distinctly separated. There are rows of papillae outlining the outer surface of the anterior scale part, whereas the posterior flange is smooth. The ornamentation of the shield is similar to the collar scales. Posteriorly on the shield, a window with 2–3 pores is present. Rear scales ($2.2\text{--}2.5 \times 1.2\text{--}1.5 \mu\text{m}$) are asymmetric with one anterior flange considerably longer than the other, and possessing a short spine (Figs 2I–J). The rearmost scales ($1.1 \times 0.8 \mu\text{m}$) are oval with a spine (Fig. 2J). Bristles not observed. Cyst unknown. Dimensions of the cell unknown, first row is formed by approximately 5–6 collar scales (Fig. 2K).

Habitat:—Mesotrophic lake.

Distribution:—France, the species was only found in the type locality.

Etymology:—The epithet refers to shield divided by one or several rows of more distinctive papillae.

Observations:—Selected environmental variables were measured during the sampling: pH 6.6, conductivity $395 \mu\text{S}\cdot\text{cm}^{-1}$, surface water temperature 16.1°C .

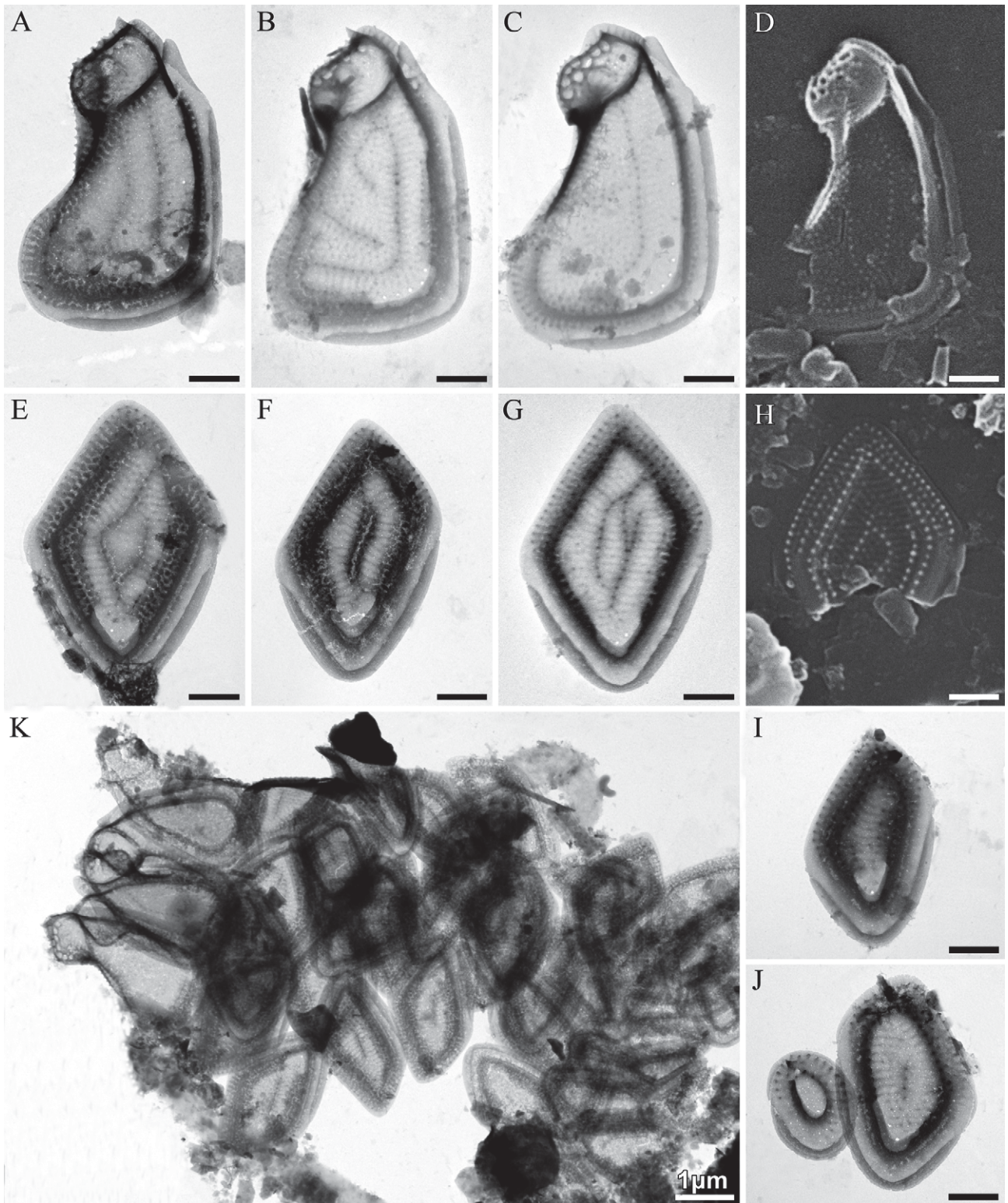


FIGURE 2. *Mallomonas dividua*. A–D, collar scales with different shield patterns. The dome is partly reticulated by circular meshes; A–C, scales viewed in a transmission electron microscope (TEM); D, a scale viewed in a scanning electron microscope (SEM). The pattern on the shield is formed by one or several rows of more distinctive papillae; E–H, body scales with different shield patterns; E–G, scales viewed in TEM; H, a scale viewed in SEM; I, a rear scale with one anterior flange considerably longer than the other; J, two rearmost scales possessing spines; K, the scales comprising the scale case. Bar = 0.5 μ m, if not stated otherwise. A–J are shown at the same magnification.

Discussion

Mallomonas temonis belongs to the section *Heterospinae* Momeu & Péterfi (1979: 19). Within this section, it is most similar to *Mallomonas canina* Kristiansen (1982: 294). All members of this section possess obovate domed scales. The base plate is perforated with closely spaced pores. Scales are marked with a more or less elaborated reticulum of ribs. In *M. canina*, the surface of the scale is ornamented with a series of prominent papillae, and except for the continuous submarginal rib, several (mostly four) primary ribs cover the shield, with the longitudinal apical rib terminating at the base of the dome (Kristiansen 1982). The scales of *M. temonis* are distinguishable from those of *M. canina*, in that they lack the prominent papillae and the reticulum is composed solely of the continuous submarginal rib connected by a single transversal rib. Moreover, only needle-shaped bristles were observed in *M. temonis*, whereas in *M. canina*, both types of bristles, needle-shaped and hooked, were documented. *Mallomonas temonis* also closely resembles *Mallomonas hindonii* Nicholls (1982: 92). The scales of both species are similar in that they lack prominent papillae on the shield and have a submarginal rib that fuses with the dome, forming a continuous entity. The scales of *M. hindonii* are ornamented with a group of primary ribs that usually originate from a central point (Nicholls 1982), whereas in *M. temonis*, only the transversal rib, in addition to the continuous submarginal rib, is present. A taxon that is similar to *M. temonis* is *Mallomonas pugio* Bradley (1964: 325). The scales of both species are similar, with a continuous submarginal rib connected by a single transversal rib. However, in *M. pugio*, well developed posterior flange ribs and a longitudinal apical rib are present (Bradley 1964). The elongated dome is ornamented with a group of thick ribs, whereas in *M. temonis*, the dome is oval and smooth. The scales of *Mallomonas harrisiae* Takahashi (1975: 41) are distinguishable from those of *M. temonis* in that the system of primary ribs of the shield is more elaborated and their rounded dome is marked with longitudinal ribs (Takahashi 1975). Moreover, apical scales of *M. harrisiae*, *M. hindonii* and *M. pugio* possess very broad and highly asymmetric domes, with one side drawn into a pointed apex and the longitudinal apical rib crossing the dome. By contrast, the dome of *M. temonis* is round, and the anterior part is extended and forms a lip. To conclude, the most distinctive scale character, discerning *M. temonis* from the other species of the section *Heterospinae*, is the transversal rib which leaves both the apical and posterior parts of the shield almost without any further structures. *Mallomonas temonis* was found in an unnamed small pool at Table Mountain together with the recently described *M. siveri* (Němcová *et al.* 2011), *Mallomonas temonis* was relatively frequent in the sample.

Mallomonas divida belongs to the section *Torquatae* Momeu & Péterfi (1979: 16), series *Mangoferae* Asmund & Kristiansen (1986: 110). The taxon that is most similar to *M. divida* is *Mallomonas mangofera* var. *gracilis* (Dürschmidt 1983: 186) Kristiansen in Kristiansen & Preisig (2007: 82). The scales of both taxa are similar in having the shield ornamented by numerous delicate papillae. In *M. mangofera* var. *gracilis*, the papillae are irregularly scattered and form a dense secondary layer (Dürschmidt 1983). By contrast, in *M. divida*, the papillae are arranged more regularly and the shield is ornamented by one to several rows of more distinctive papillae. Moreover, all types of scales are slightly smaller in *M. mangofera* var. *gracilis* than in *M. divida*. *Mallomonas villosa* Dürschmidt (1986: 101) differs from *M. divida* by the presence of the mesh pattern on the shield formed by groups of papillae, and in having depressions of thinner scale material on the anterior flange. Moreover, the dome of collar scales in *M. villosa* is marked with patches of minute pores and papillae and is terminated by a sharply pointed peak. The rear scales of *M. villosa* may also possess one to several spines (Dürschmidt 1986), whereas only a single spine was observed in *M. divida*. *Mallomonas divida* also resembles *Mallomonas madagascariensis* P. Hansen in Hansen & Kristiansen (1995: 216), but this last differs in that it has the shield papillae arranged in a regular hexagonal way and an almost unornamented dome. The body scales of *Mallomonas crocodilorum* P. Hansen in Hansen & Kristiansen (1995: 222) are distinguishable from those of *M. divida* in the shield pattern that comprises distinctive papillae with even spacing but with no apparent symmetry. Moreover, approximately two rows of shield papillae continue into one side of the anterior flange (Hansen & Kristiansen 1995). All types of scales are slightly larger than those of *M. divida*. Finally, rear scales of *M. crocodilorum* are spine-less. A similar taxon to *M. divida* is also

Mallomonas inornata Nicholls (1989: 293) from the series *Eoae* Asmund & Kristiansen (1986: 106) (section *Torquatae*). Several features may be used to distinguish these two species. *Mallomonas inornata* differs in that there is only a single prominent pore posteriorly on the shield window, the shield is unpatterned, and collar scales possess an apically positioned rounded dome with a prominent beak-like protrusion (Nicholls 1989). *Mallomonas divida* shared an unnamed lake south of the Reserve Naturelle Marais d'Orx (Aquitaine) with a diverse silica-scaled community and was relatively rare.

Future research will reveal whether *Mallomonas temonis* and *M. divida* are restricted to warm areas with mild winters, or whether their distribution is wider and they have been unnoticed because of their rare occurrence and/or low population numbers. Description of new species often initiates new reports from other parts of the world. Further studies on silica-scaled chrysophytes may help to evaluate the distribution patterns and environmental requirements of these newly described species.

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References

- Andersen, R.A. (2007) Molecular systematics of the Chrysophyceae and Synurophyceae. In: Brodie, J. & Lewis, J. (eds.) *Unravelling the algae: The past, present, and future of algal systematics*. CRC Press, Boca Raton, pp. 283–311. <http://dx.doi.org/10.1201/9780849379901.ch15>
- Asmund, B. (1955) Electron microscope observations on *Mallomonas caudata* and some remarks on its occurrence in four Danish ponds. *Botanisk Tidsskrift* 52: 163–168.
- Asmund, B. & Kristiansen, J. (1986) The genus *Mallomonas* (Chrysophyceae). A taxonomic survey based on the ultrastructure of silica scales and bristles. *Opera Botanica* 85: 1–128.
- Bradley, D.E. (1964) A study of the *Mallomonas*, *Synura* and *Chryso-sphaerella* of Northern Iceland. *Journal of General Microbiology* 37: 321–333. <http://dx.doi.org/10.1099/00221287-37-3-321>
- Dürschmidt, M. (1983) A taxonomic study of the *Mallomonas mangofera* group (Mallomonadaceae, Chrysophyceae), including the description of four new taxa. *Plant Systematics and Evolution* 143: 175–196. <http://dx.doi.org/10.1007/BF00986377>
- Dürschmidt, M. (1986): New species of the genus *Mallomonas* (Mallomonadaceae, Chrysophyceae) from New Zealand. In: Kristiansen, J. & Andersen, R. A. (eds), *Chrysophytes. Aspects and Problems*. Cambridge University Press, Cambridge, pp. 87–106.
- Ehrenberg, C.G. (1834) Beiträge zur physiologischen Kenntniss der Corallenthiere im allgemeinen, und besonders des rothen Meeres, nebst einem Versuche zur physiologischen Systematik derselben. 1. *Abhandlungen der Königlichen Akademie Wissenschaften zu Berlin, Physikalische Klasse* 1832(1): 225–380.
- Fott, B. (1955) Scales of *Mallomonas* observed in the electron microscope. *Preslia* 27: 280–282.
- Guiry, M.D. & Guiry, G.M. (2012) AlgaeBase. World-wide electronic publication, National University of Ireland, Galway. Available from <http://www.algaebase.org> (accessed on 11-13-2012).
- Hansen, P. & Kristiansen, J. (1995) *Mallomonas madagascariensis*, *M. lemuriocellata* and *M. crocodilorum* (Synurophyceae), three new species from Madagascar. *Nordic Journal of Botany* 15: 215–223. <http://dx.doi.org/10.1111/j.1756-1051.1995.tb00146.x>
- Jo, B.Y., Shin, W., Boo, S.M., Kim, H.S. & Siver, P.A. (2011) Studies on ultrastructure and three-gene phylogeny of the genus *Mallomonas* (Synurophyceae). *Journal of Phycology* 47: 415–425. <http://dx.doi.org/10.1111/j.1529-8817.2010.00953.x>
- Kim, H.S. & Kim, J.H. (2008) *Mallomonas koreana* sp. nov. (Synurophyceae), a new species from South Korea. *Nova Hedwigia* 86: 469–476. <http://dx.doi.org/10.1127/0029-5035/2008/0086-0469>
- Kristiansen, J. (1982) *Mallomonas canina* sp. nov. (Chrysophyceae), a new member of sect. *Heterospinae*. *Nordic*

- Journal of Botany* 2: 293–296.
<http://dx.doi.org/10.1111/j.1756-1051.1982.tb01192.x>
- Kristiansen, J. (2002) The genus *Mallomonas* (Synurophyceae) - A taxonomic survey based on the ultrastructure of silica scales and bristles. *Opera Botanica* 139: 1–218.
- Kristiansen, J. & Preisig, H.R. (2007) Chrysophyte and Haptophyte algae. Part 2: Synurophyceae. In: Büdel, B., Gärtner, G., Krienitz, L., Preisig, H.R. & Schagerl, M. (eds.): *Süßwasserflora von Mitteleuropa. Freshwater Flora of Central Europe*. Spektrum Akademie Verlag, Heidelberg, pp. 1–252.
- Lavau, S., Saunders, G.W. & Wetherbee, R. (1997) A phylogenetic analysis of the Synurophyceae using molecular data and scale case morphology. *Journal of Phycology* 33: 135–151.
<http://dx.doi.org/10.1111/j.0022-3646.1997.00135.x>
- Momeu, L. & Péterfi, L.S. (1979) Taxonomy of *Mallomonas* based on the fine structure of the scales and bristles. *Contribuții Botanice Cluj-Napoca* 1979: 13–20.
- Němcová, Y., Bulant, P. & Kristiansen, J. (2011) *Mallomonas solea-ferrea* and *Mallomonas siveri* (Stramenopiles): two new taxa from the Western Cape (South Africa). *Nova Hedwigia* 93: 375–384.
<http://dx.doi.org/10.1127/0029-5035/2011/0093-0375>
- Němcová, Y., Kreidlová, J., Kosová, A. & Neustupa, J. (2012) Lakes and pools of Aquitaine region (France) - a biodiversity hotspot of Synurales in Europe. *Nova Hedwigia* 95: 1–24.
<http://dx.doi.org/10.1127/0029-5035/2012/0036-0001>
- Nicholls, K.H. (1982) *Mallomonas* species (Chrysophyceae) from Ontario, Canada, including descriptions of two new species. *Nova Hedwigia* 36: 89–124.
- Nicholls, K.H. (1989) Descriptions of four new *Mallomonas* taxa (Mallomonadaceae, Chrysophyceae). *Journal of Phycology* 25: 292–300.
<http://dx.doi.org/10.1111/j.1529-8817.1989.tb00125.x>
- Perty, M. (1852) *Zur Kenntniss kleinster Lebensformen: nach Bau, Funktionen, Systematik, mit Specialverzeichniss der in der Schweiz beobachteten*. Jent & Reinert, Bern, 228 pp.
- Řezáčová, M. & Neustupa, J. (2007) Distribution of the genus *Mallomonas* (Synurophyceae) - Ubiquitous dispersal in microorganisms evaluated. *Protist* 158: 29–37.
<http://dx.doi.org/10.1016/j.protis.2006.06.004>
- Siver, P.A. (1991) *The biology of Mallomonas. Morphology, taxonomy, ecology*. Kluwer Academic Publishers, Dordrecht. 230 pp.
<http://dx.doi.org/10.1007/978-94-011-3376-0>
- Siver, P.A. & Wolfe, A.P. (2005) Eocene scaled chrysophytes with pronounced modern affinities. *International Journal of Plant Sciences* 166: 533–536.
<http://dx.doi.org/10.1086/428702>
- Starmach, K. (1985) Chrysophyceae und Haptophyceae. In: Ettl, H., Gerloff, J., Heynig, H., Mollenhauer, D. (eds.): *Süßwasserflora von Mitteleuropa I*. Gustav Fischer Verlag, Stuttgart, pp. 1–515.
- Takahashi, E. (1975) Studies on genera *Mallomonas* and *Synura*, and other plankton in freshwater with electron microscope IX. *Mallomonas harrisae* sp. nov. (Chrysophyceae). *Phycologia* 14: 41–44.
<http://dx.doi.org/10.2216/i0031-8884-14-1-41.1>