

Received: September 3, 2024  
Accepted: October 24, 2024

ISSN 1857–9027  
e-ISSN 1857–9949  
UDC:  
DOI: 10.20903/masa/nmbsci.2023.44.26

*Original scientific paper*

## NEW AND INTERESTING *MAYAMAEA* SPECIES (BACILLARIOPHYCEAE) FROM OSOGOVO MOUNTAIN

Zlatko Levkov\*, Dušica Zaova, Danijela Mitić-Kopanja

Institute of Biology, Faculty of Natural Sciences, Ss Cyril and Methodius University in Skopje,  
RN Macedonia

\*e-mail: zlevkov@pmf.ukim.mk

Observations of a sample from a small spring on Mountain Osogovo revealed the presence of several interesting and previously unknown small-celled diatom species. Among them, the genus *Mayamaea* shows relatively high diversity, with at least eight species. Four species are presented in this study, two of which are described as new: *Mayamaea matevskiana* sp. nov. and *Mayamaea stafilovii* Levkov & Zaova sp. nov. *Mayamaea matevskiana* is characterized by elliptical to elliptic-lanceolate valves, relatively coarse and strongly radiate striae, an indistinct central area bordered by more distantly positioned central striae, slightly expanded proximal raphe ends, and simple distal raphe ends. *Mayamaea stafilovii* has a similar valve morphology and shape of the central area to *M. matevskiana* but differences can be observed in the valve size (which is considerably smaller than *M. matevskiana*) and the shape of the external distal raphe ends (which are long, slightly deflected, and extend onto the valve mantle). The new species are described based on detailed light and scanning electron microscopic observations.

**Key words:** diatoms; diversity; morphology; springs; taxonomy

### INTRODUCTION

The use of electron microscopy, particularly scanning electron microscopy (SEM), in diatom research enables the observation of fine frustule structures and, especially, the morphology of small-cell species which are often difficult to visualize with a light microscope (LM). In recent decades, many new species and genera have been described based on ultrastructural features observed with SEM [1–4]. Numerous new genera have been separated from large and morphologically diverse genera, such as *Navicula* Bory, *Achnanthes* Bory, *Cymbella* C. A. Agardh, *Amphora* Ehrenberg, and others [1–4]. The genus *Mayamaea* Lange-Bertalot [2] was separated from *Navicula* Bory based on *Navicula atomus* (Kützinger) Grunow (= *Amphora atomus* Kützinger). The genus comprises small-celled diatoms with elliptical to lanceolate valves, and striae composed of individual areolae occluded by hymen, located toward the outer areola opening [5–7]. Recent molecular studies show that the ge-

nus is monophyletic [8], although only a few members have been genetically studied [9].

Representatives of the genus *Mayamaea* can be found in almost all types of water and wet habitats, including oligotrophic habitats [10, 11], eutrophic streams [5], pools [6], oxbow lakes [12], and soils [7]. Many species are known only from their type locality or have restricted distributions, such as in Antarctica or Hawaii [13, 14], while a few are considered widely distributed in Europe [15], mostly found in eutrophic and polluted rivers [16]. However, this could be a consequence of sampling bias, as wet or subaerial habitats (e.g., wet mosses, temporary springs, soils) are not frequently surveyed. The recent increased awareness of climate change and global warming has led to more detailed studies on springs, including temporary springs [17–19]. These studies have expanded the distribution range of some previously known rare species [20, 21] and have resulted in the description of new species [22].

More recently, specific habitats, including halomorphic soils, hypersaline springs and subaerial habitats, have been studied in N. Macedonia [23–26], resulting in the description of several new diatom species. However, temporary or small springs have rarely been investigated for diatoms, although they host many interesting and potentially new species [27]. During the observations of a sample from a small spring near the monastery Sv. Pantelejmon, near the village of Pantelej, Mountain Osogovo, several small-cell naviculoid species of different genera such as *Chamaepinnularia* Lange-Bertalot & Krammer, *Craticula* Grunow, *Muelleria* (Frenguelli) Frenguelli, etc. were recorded. At least eight different species of *Mayamaea* have been observed, and at least two species had morphological features that differed from previously known species. These two species are described here as *Mayamaea matevskiana* sp. nov. and *Mayamaea stafilovii* Levkov & Zaova sp. nov.

#### MATERIAL AND METHODS

Osogovo Mountain, located in the north-eastern part of the Republic of N. Macedonia, with a small portion extending into Bulgaria, is a massive mountain range with relatively homogeneous geology, consisting primarily of Precambrian and Paleozoic crystalline rocks. The site of Pantelej is located in the southwestern part of the mountain, at an altitude of approximately 770 m. Geologically, it is characterized by Neogene andesite rocks, and hydrologically by a number of small rock springs. The sampling site for this study is a small spring near the Sv. Petka fountain, close to the Sv. Pantelejmon Monastery, near the village of Pantelej (41.969376 N, 22.310405 E). Samples were collected from the mud of a small, temporary wet pond formed by the spring. The field sampling campaign was conducted twice, in January 2021 and May 2021.

Diatom samples for LM and SEM observations were preserved in the laboratory with a 4% formaldehyde solution. In the laboratory, the samples were treated with  $\text{KMnO}_4$  and left overnight. To remove the organic matter, 37 % HCl was added and the samples were boiled at 80 °C for 45 minutes [25]. The samples were then centrifuged and rinsed five times with distilled water. Permanent diatom slides were mounted with Naphrax (Brunel Microscopes Ltd). LM observations were conducted using a Nikon Eclipse 80i microscope, under oil immersion at 1000 $\times$  magnification with differential interference contrast (DIC), while diatom images were captured with a Nikon Coolpix P6000 camera. For SEM observations, aliquots of the diatom sample were dried on a coverslip at-

tached to stubs with carbon tape and coated with a thin gold-palladium layer (approximately 20 nm) for 120 seconds with a sputtering current of 20 to 25 mA using a Polaron SC7640 Sputter (Quorum Technologies, Ashford, UK). Scanning electron microscopy was performed at 5 kV and a 5 mm working distance using a Zeiss Gemini Ultra Plus FEM (Cambridge Instruments Ltd, Cambridge, UK).

#### RESULTS

Four *Mayamaea* species are presented in this study. Two species are already known from a few localities in Europe, but are rarely illustrated using LM and SEM, while two species exhibit characteristics that differ from other known species and are described here as new species.

##### *Mayamaea edaphica* C.E. Wetzel nom. prov.

(Plate 1: Figs 1–54, Plate 2: Figs 1–6)

**Morphology LM:** Valves small, strictly elliptical with rounded apices. Valve length 8.0–11.5  $\mu\text{m}$ , valve width 4.0–5.0  $\mu\text{m}$  ( $n = 54$ ). Axial area very narrow, linear. Central area broad unilateral fascia widening towards valve mantle. Raphe filiform with indistinct proximal raphe ends. Striae fine, radiate throughout, 30–35 in 10  $\mu\text{m}$ . Central striae coarser and irregularly shortened.

**SEM:** Externally, valve face flat, transition to valve mantle gradual. Central area broad unilateral fascia. Raphe linear with tear-drop-shaped and slightly unilaterally deflected proximal raphe ends. Distal raphe ends simple and slightly expanded, located on the valve face. Striae uniseriate, composed of round areolae occluded with hymens. Areolae on valve mantle elongated. Striae bordering central area, irregularly shortened. Internally, central area slightly inwardly elevated, axial area slightly thickened sternum. Proximal raphe ends simple, distally raphe terminates in small helictoglossae. Areolae on valve face with round to elongated openings with visible external occlusions, especially towards valve mantle.

**Remarks:** This species was recently described by Wetzel (pers. comm.) from soils in Luxembourg. A previous record of this species is provided by Reichardt [28] under *Mayamaea* spec. The type population consists of elliptic-lanceolate to linear-elliptic and slightly narrower valves (3.0–4.0  $\mu\text{m}$ ) with densely spaced striae (40–45 in 10  $\mu\text{m}$ ). Such a stria density leads to an almost hyaline appearance of the valve. SEM observations reveal a large central area with a unilateral fascia, which is a unique feature of this species. The significance of the differences in valve shape, width, and stria density between the type population and

the population from Mountain Osogovo remains uncertain, as only two populations of this species are known and illustrated to date. Further studies and records of *M. edaphica* from different localities will help elucidate the broader morphological variability and stability of particular features.

***Mayamaea excelsa* (Krasske) Lange-Bertalot**  
(Plate 3, Figs 1–22)

**Morphology LM:** Valves strictly elliptical with rounded ends. Valve length 14.0–17.5 µm, valve width 5.5–6.5 µm ( $n = 30$ ). Axial area relatively narrow, linear. Central area almost absent, not separated from axial area. Central striae more distantly spaced. Raphe filiform with slightly expanded proximal and distal raphe ends. Striae fine, strongly radiate throughout, 15–21 in 10 µm. Individual areolae not recognizable with LM.

**SEM:** Externally, valve face flat, transition to valve mantle gradual. Axial area broad, lanceolate, weakly expanding towards mid-valve. Central area absent and not differentiated from axial area. Raphe linear with tear-drop-shaped proximal raphe ends. Distal raphe ends very short and weakly curved, located on the valve face. Striae uniseriate, composed of round areolae occluded with hymens. Areolae on valve mantle elongated. Few striae bordering central area irregularly shortened. Internally, central and axial area flat. Proximal raphe ends simple and slightly unilaterally deflected, distally raphe terminates in small helictoglossae. Areolae on valve face with round to elongated openings with visible occlusions.

**Remarks:** The lectotype of *Mayamaea excelsa* (= *Navicula excelsa* Krasske) was designated by Lange-Bertalot *et al.* [31]. According to the protologue [32], the valves are elliptical, 16 µm long and 7.0 µm wide with 12 striae in 10 µm in the middle. Lange-Bertalot [15] provide a slightly wider range of valve size: 12.0–16.0 µm long and 5.5–7.0 µm wide with 16–18 striae in 10 µm.

So far, only two valves of *M. excelsa* illustrated with SEM have been published. The valve depicted in Lange-Bertalot [15, pl. 103, figs 4, 5] is characterized by a very broad central area, coarsely punctate striae and long and deflected distal raphe ends passing on the valve mantle. LM images of *M. excelsa* and description [15, p. 138, pl. 104, figs 35–40] indicates that central area is almost absent in this species. Probably this valve does not belong to *M. excelsa* *sensu stricto*. The valve illustrated in Barragán *et al.* [7] is slightly narrower (4.8 µm) and has a distinct central area composed of irregularly shortened central stria and prolonged distal raphe ends strongly unilaterally bent. However, the observed specimen from the small spring near the

Sv. Petka fountain differs considerably from the aforementioned valve in the shape of the axial area, central area, proximal and distal raphe ends, as well as in the stria and areola density. The conspecificity of these two specimens remains uncertain.

*Mayamaea excelsa* is for the first time recorded for diatom flora of N. Macedonia. According to Lange-Bertalot [15], this species is presumably cosmopolitan, but always with a small number of specimens.

***Mayamaea matevskiana* sp. nov.**  
(Plate 4: Figs 1–29, Plate 5: Figs 1–6)

**Description LM:** Valves small, elliptical to elliptic-lanceolate with narrowly rounded ends. Valve length 9.0–12.0 µm, valve width 4.0–4.5 µm ( $n = 35$ ). Axial area very narrow, linear to lanceolate. Central area almost absent, not separated from axial area. Central striae more distantly spaced. Raphe filiform with slightly expanded proximal and distal raphe ends. Striae fine, strongly radiate throughout, 18–22 in 10 µm. Individual areolae not recognizable with LM.

**SEM:** Externally, valve face flat, transition to valve mantle gradual. Axial area moderately broad, linear, weakly expanding towards mid-valve. Central area absent and not differentiated from axial area, bordered with more distantly spaced central striae. Raphe linear with tear-drop-shaped and weakly bent proximal raphe ends. Distal raphe ends simple, located on the valve face. Striae uniseriate, strongly radiate. Striae absent at valve ends. Areolae round occluded with hymens, ca. 50 in 10 µm. Internally, central and axial area thickened and slightly inwardly elevated. Proximal raphe ends simple and slightly unilaterally deflected, distally raphe ends slightly unilaterally deflected and terminates in small helictoglossae. Areolae on valve face with round to elongated openings with visible occlusions.

**Type:** Slide Acc. No. 013814A, small spring near Sv. Petka fountain, near village Pantelej, Mountain Osogovo, N. Macedonia, 41.969376 N, 22.310405 E. 805 m a.s.l. sediment, Coll. date: 07.01.2021, Leg. D. Zaova.

**Isotype:** BM 92456

**Etymology:** The species is named in honor of Prof. Dr. Vlado Matevski, a prominent botanist and member of the Macedonian Academy of Sciences and Arts for his 70<sup>th</sup> birthday.

**Ecology and distribution:** The species was observed in sediment from a small spring. The highest abundance was recorded during the winter months, indicating a preference for cold, oligo-

trophic waters. So far, this species is known only from the type locality.

**Comparison with similar taxa:** The most similar species to *M. matevskiana* is *M. excelsa* in terms of the valve outline and morphology of the axial and central areas. With LM, differences can be observed in the valve size (length 12.5–16.5  $\mu\text{m}$ , width 5.5–8.5  $\mu\text{m}$  in *M. excelsa* vs. length 9.0–12.0  $\mu\text{m}$ , width 4.0–4.5  $\mu\text{m}$  in *M. matevskiana*). With SEM, the main differences can be noticed in the morphology of the raphe (the distal raphe ends in *M. excelsa* are short and bent, whereas the raphe ends in *M. matevskiana* are simple) and in the areola size (smaller in *M. excelsa*) and density of the areolae (approximately 60 areolae in 10  $\mu\text{m}$  in *M. excelsa*). A comparable valve size is present in *M. atomus* (Kützing) Lange-Bertalot (length 8.5–13.0  $\mu\text{m}$ , width 4.0–5.5  $\mu\text{m}$ ), but differences are noticeable in the shape of the central area (elliptical, bordered by irregularly shortened striae in *M. atomus*) and the distal raphe ends (long, strongly deflected and extending onto the valve mantle in *M. atomus*). *Mayamaea alcimonia* (E. Reichardt) C.E. Wetzel, Barragán & Ector has a well-defined central area, with unilaterally deflected proximal raphe ends and long, strongly deflected distal raphe ends that extend onto the valve mantle [33]. *Mayamaea permitis* (Hustedt) K. Bruder & Medlin has smaller valves (length 6.0–9.0  $\mu\text{m}$ , width 3.0–4.0  $\mu\text{m}$ ) with a higher stria density (30–36 in 10  $\mu\text{m}$ ). Compared to other known *Mayamaea* species [15], *M. matevskiana* can be differentiated by the shape of the central area (which is well defined and elliptical to irregularly shaped in many *Mayamaea* species), the orientation of the striae (many species have parallel or slightly radiate striae), and the stria density (which is higher in many other *Mayamaea* species).

**Diatom composition** is typical for subaerial habitats and dominated by *Adlafia minuscula* (Grunow) Lange-Bertalot, *Hantzschia abundans* Lange-Bertalot, *H. amphioxys* (Ehrenberg) Grunow, several *Luticola* species such as *Luticola nivalis* (Ehrenberg) D. G. Mann, *Luticola pseudonivalis* (Bock) Levkov, Metzeltin & A. Pavlov, *L. levkovii* E. Reichardt, *Nitzschia* aff. *aurariae* Chohnoky, *Surirella angusta* Kützing and several unknown *Mayamaea* species.

***Mayamaea stafilovii* Levkov & Zaova sp. nov.**

(Plate 6: Figs 1–46, Plate 7: Figs 1–6)

**Description LM:** Valves small, elliptic-lanceolate, lanceolate to linear-lanceolate with narrowly rounded ends. Valve length 8.0–11.5  $\mu\text{m}$ , valve width 3.0–4.0  $\mu\text{m}$  ( $n = 51$ ). Axial area very

narrow, linear. Central area small, but distinct, irregularly shaped bordered 5–6 irregularly shortened central striae. In some specimens central striae more distantly spaced. Raphe filiform with slightly expanded proximal and distal raphe ends. Striae fine, strongly radiate throughout, 26–30 in 10  $\mu\text{m}$ . Individual areolae not recognizable with LM.

**SEM:** Externally, valve face flat, transition to valve mantle gradual. Axial area narrow, linear, weakly expanding towards central area. Central area small, round bordered with irregularly shortened central striae. Raphe linear with tear-drop-shaped and weakly bent proximal raphe ends. Distal raphe ends long, unilaterally deflected simple, located on the valve face. Striae uniseriate, strongly radiate. Areolae round occluded with hymens, ca. 40–50 in 10  $\mu\text{m}$ . Internally, central and axial area slightly thickened and inwardly elevated. Proximal raphe ends simple and slightly unilaterally deflected, distally raphe terminates in small helictoglossae. Areolae on valve face with large round to elongated openings with visible occlusions.

**Type:** Slide Acc. No. 013814B, small spring near Sv. Petka fountain, near village Pantelej, Mountain Osogovo, N. Macedonia, 41.969376 N, 22.310405 E. 805 m a.s.l. sediment, Coll. date: 07.01.2021, Leg. D. Zaova.

**Isotype:** BM 92456

**Etymology:** The species is named in honor of Prof. Dr. Trajce Stafilov, Faculty of Natural Sciences, Ss. Cyril and Methodius University for his significant contribution in freshwater ecology.

**Ecology and distribution:** Similar to the previous species, *M. stafilovii* was observed during the winter months, indicating a preference for cold, oligotrophic waters with low flow. It was found on sediment near the spring. So far, it is known only from the type locality.

**Comparison with similar taxa:** *Mayamaea stafilovii* is morphologically similar with *M. atomus*. Differences between these two species can be noticed in the valve shape (elliptical in *M. atomus*), valve width (4.0–5.5  $\mu\text{m}$  in *M. atomus*), stria density (19–22 in 10  $\mu\text{m}$  in *M. atomus*) and shape of the central area (indistinct and slightly separated from the axial area in *M. atomus*). *Mayamaea stafilovii* was observed in the same sample as *M. matevskiana*, but these two species can be easily differentiated by the valve shape (elliptic in *M. matevskiana* vs. elliptic-lanceolate, lanceolate to linear-lanceolate in *M. stafilovii*), valve size (4.0–5.5  $\mu\text{m}$  in *M. matevskiana* vs. 3.0–4.0  $\mu\text{m}$  in *M. stafilovii*), stria density (18–22 in 10  $\mu\text{m}$  vs. 26–30 in 10  $\mu\text{m}$  in *M. stafilovii*). *Mayamaea alcimonia* has more elliptical valves with coarser stria (24–26 in

10  $\mu\text{m}$ ) and internally the raphe sternum is strongly thickened and inwardly elevated [33]. Comparable valve size has *M. permitis*, but the latter species can be easily differentiated by the valve shape and stria density (30–36 in 10  $\mu\text{m}$ ) giving more hyaline appearance of the valve [15, 34].

## DISCUSSION

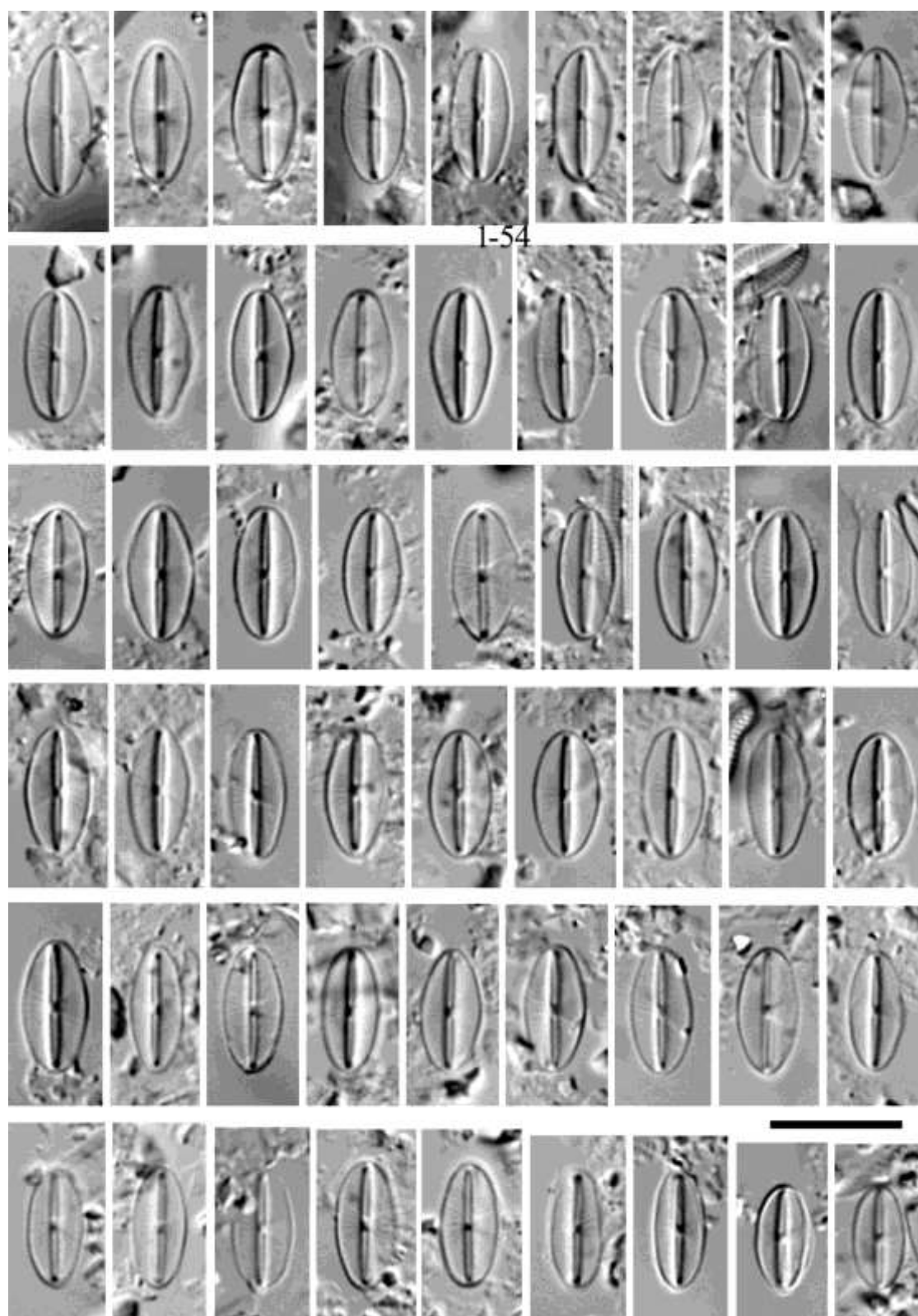
The genus *Mayamaea* is considered as relatively small genus with about 30 species [7]. It comprises small-celled taxa with elliptical to lanceolate outlines and usually with fine striae. There are few morphological characters visible with LM that might be used for differentiation of species, such as valve shape, size, central area and stria orientation and density. In combination with the valves' small size, identification and differentiating species becomes challenging. In such cases, some species may be overlooked and/or misidentified. Hustedt [35] illustrated many small-celled species that were recently transferred to *Mayamaea*. Some of them were later considered as conspecific, by several authors. For instance, *Navicula peratomus* Hustedt [36] was considered as a later synonym of *Navicula permitis* Hustedt [10] by Krammer & Lange-Bertalot [37]. The latter species was described from Plitvice Lakes, Croatia, and represents typical aerophytic species inhabiting wet mosses [35]. However, by many authors is considered as one of the most tolerant species to pollution [15]. On the other side, *N. peratomus* was described from Weser, Germany, and considered a pollution-tolerant species [35]. Similarly, *M. atomus* was considered tolerant to high levels of saprobity [16]. The latter species is most likely also an aerophytic species, and many of the records could actually refer to other species. Similarly, Lange-Bertalot et al. [38] expressed concerns about the identity of some taxa recorded under the name *Mayamaea spirans* (Hustedt) Lange-Bertalot. Molecular analyses could provide support for species differentiation [8], but there are two major challenges: the limited number of species for which sequences have been published, and the small genetic differences between these sequences [9].

In more recent studies, the genus *Mayamaea* is considered predominantly aerophytic [7, 39]. Wet habitats (such as soils, rocks, mosses, and temporary springs) are not included in monitoring schemes and are rarely studied. These habitats often host a relatively small number of species, but in many cases, they also contain unknown and probably new species [40]. Most of the studies have

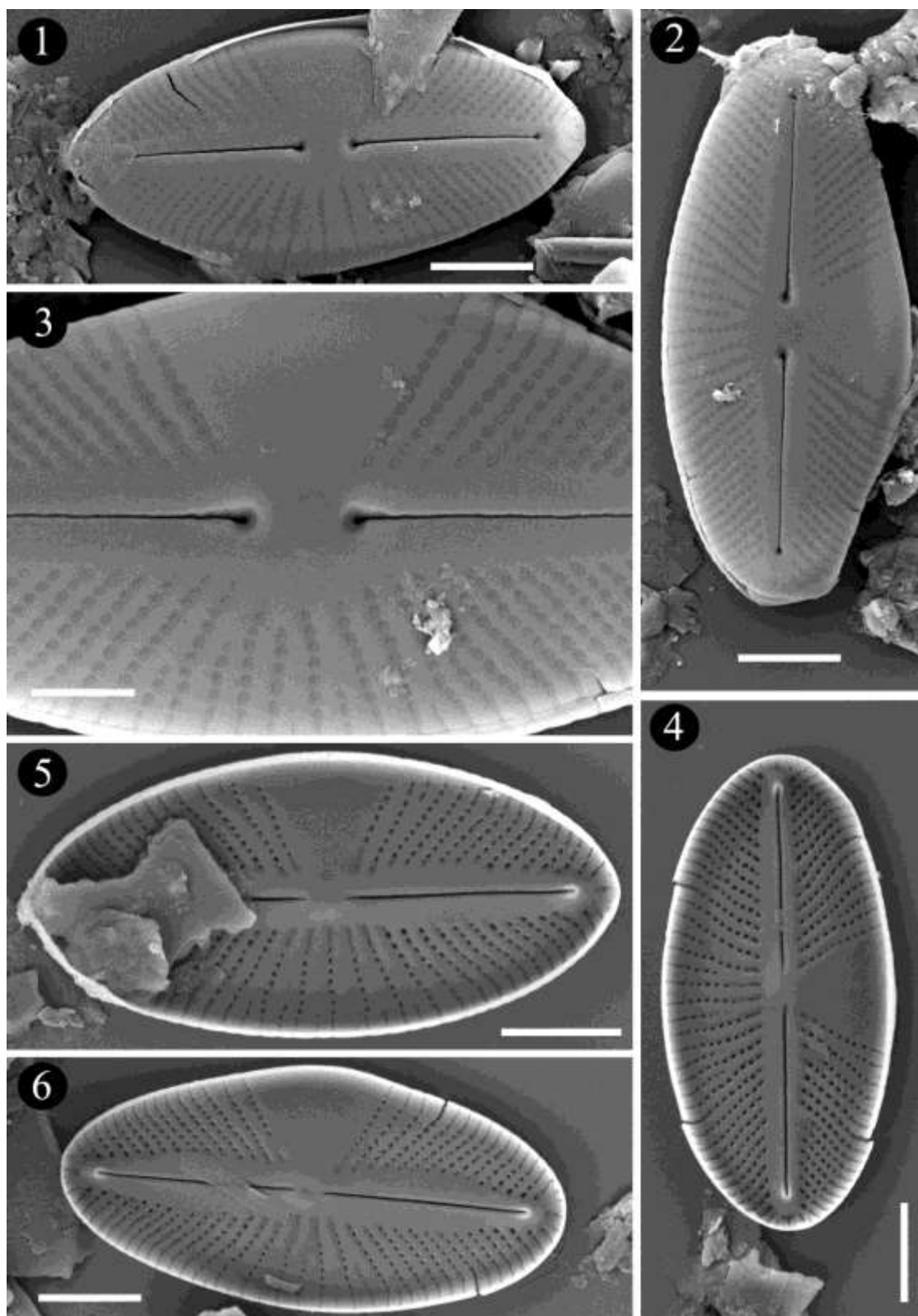
been conducted in the (sub)Antarctic region [e.g., 11, 14], while studies in Europe are relatively rare [e.g., 41, 42]. More recently, unusual habitats for diatoms, such as lampenflora, have been studied, including wet walls in caves, dams, and metro stations [43–45]. Observations in these habitats have led to the description of several new species [46, 47]. Detailed studies of such habitats reveal the existence of many interesting, rare, and likely many unknown species (Levkov, unpub. data).

Wet habitats may be considered highly endangered due to climate change and human impact. An increase in temperature and a decrease in precipitation [48, 49] will reduce humidity and water flow, directly affecting the vegetation period. Higher temperatures will influence snowmelt patterns, leading to a reduction in spring flow during the summer. Additionally, higher temperatures will increase evaporation rates, which will further reduce available water and extend dry periods. The effects of climate change on small springs are complex and interlinked, resulting in reduced water availability and quality, altered ecosystems, and posing major challenges to diatom communities. Furthermore, many small springs have been completely or partially captured and/or modified as fountains, which has led to the destruction of natural habitats for diatoms. These human activities will contribute to a significant decline in the distribution of subaerial species and, ultimately, a loss of biodiversity. Further studies on small springs are needed to understand their responses to climate change, particularly the responses of diatoms. Long-term studies on species composition, shifts in communities, changes in biodiversity, and alterations in habitat structure are essential to understanding the importance and vulnerability of these habitats. Future research will provide valuable insights for protecting small springs as biological hotspots.

The sample used as type material for the description of the new species in this study was collected during the winter months (January). Samples from the same site were also collected in late spring, but the abundance of *Mayamaea* species was significantly lower. This suggests that the species observed in this study prefer cold, oligotrophic, and wet habitats. Such preferences imply that these species may be highly sensitive to climate change and could be considered endangered. However, their distribution is not yet well understood, and further studies in similar habitats are needed to more precisely define their biogeography, ecology, and sensitivity.

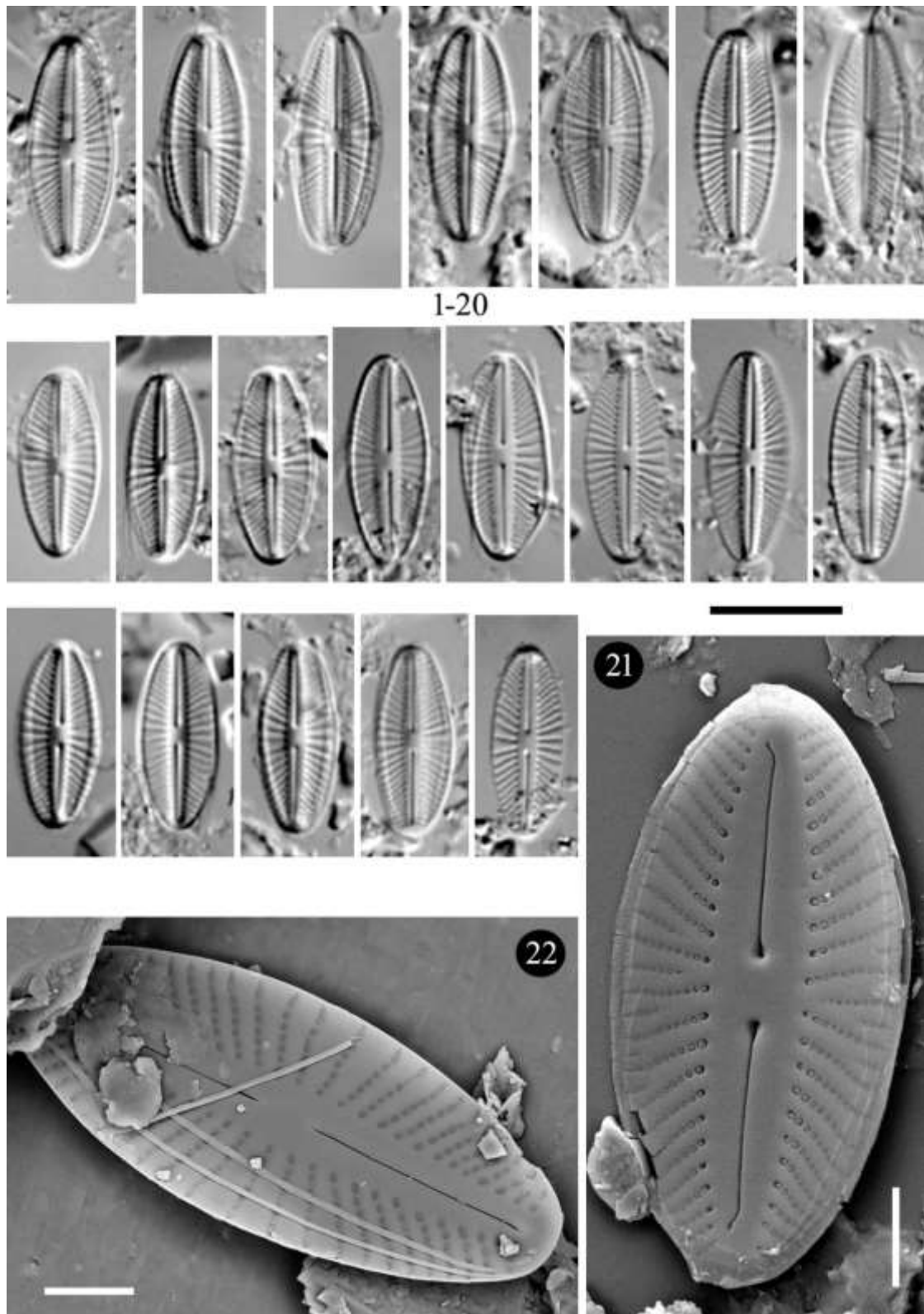


**PLATE 1: Figs 1–54.** LM micrographs of *Mayamae edaphica* from the spring near fountain Sv. Petka, Mountain Osogovo. Scale bar = 10  $\mu$ m.



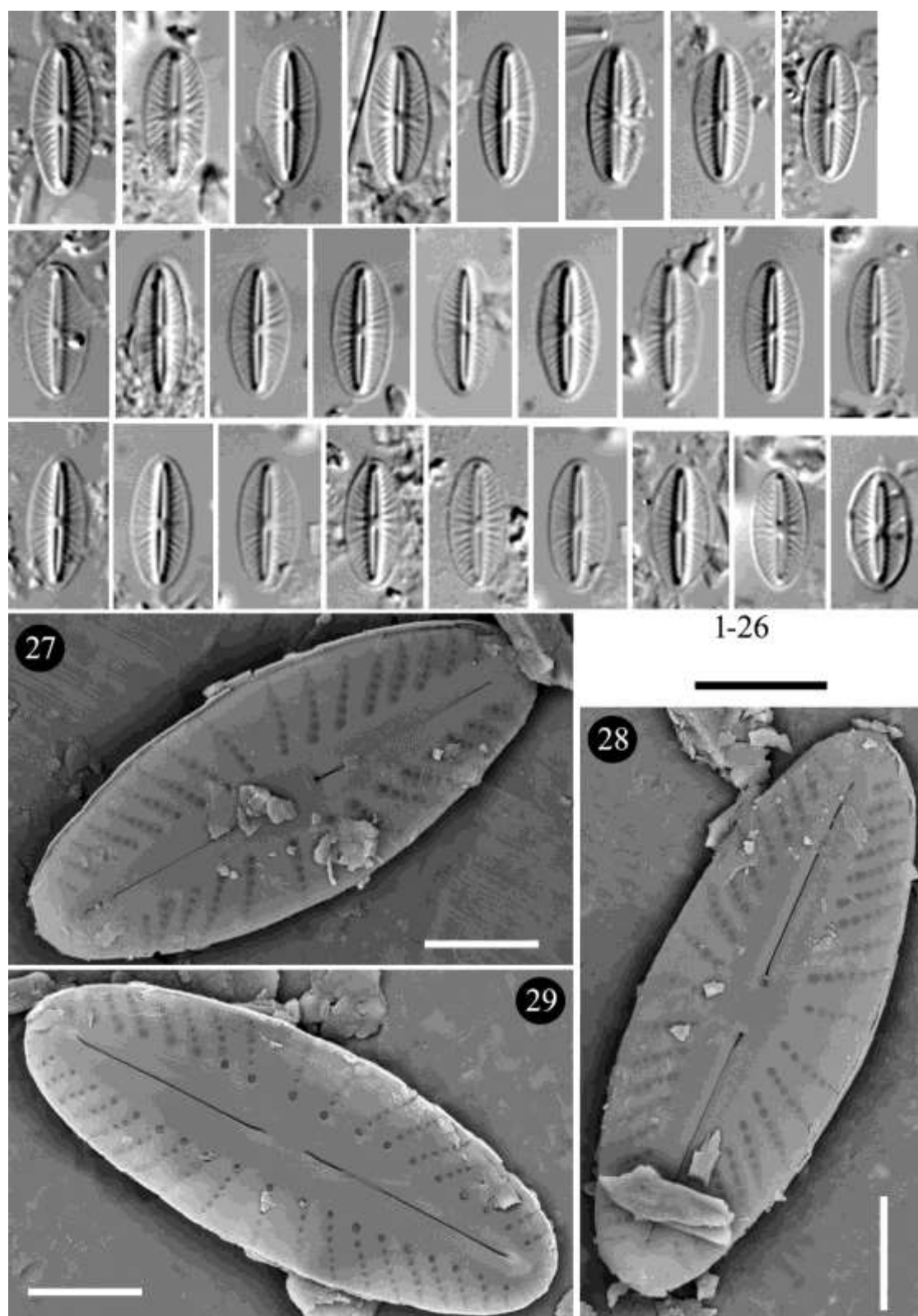
**PLATE 2: Figs 1–6.** SEM micrographs of *Mayamaea edaphica*. Figs 1, 2. External view of the whole valve showing the raphe and stria morphology. Fig. 3. Detailed external view of the mid-valve showing the unilateral central area, proximal raphe ends and striae composed of round and occluded areolae. Figs 4–6. Internal view of the whole valve showing the raphe with simple proximal raphe ends and distal ends terminating in small helictoglossae. Scale bar = 2  $\mu\text{m}$  (Figs 1, 2, 4–6); 1  $\mu\text{m}$  (Fig. 3).



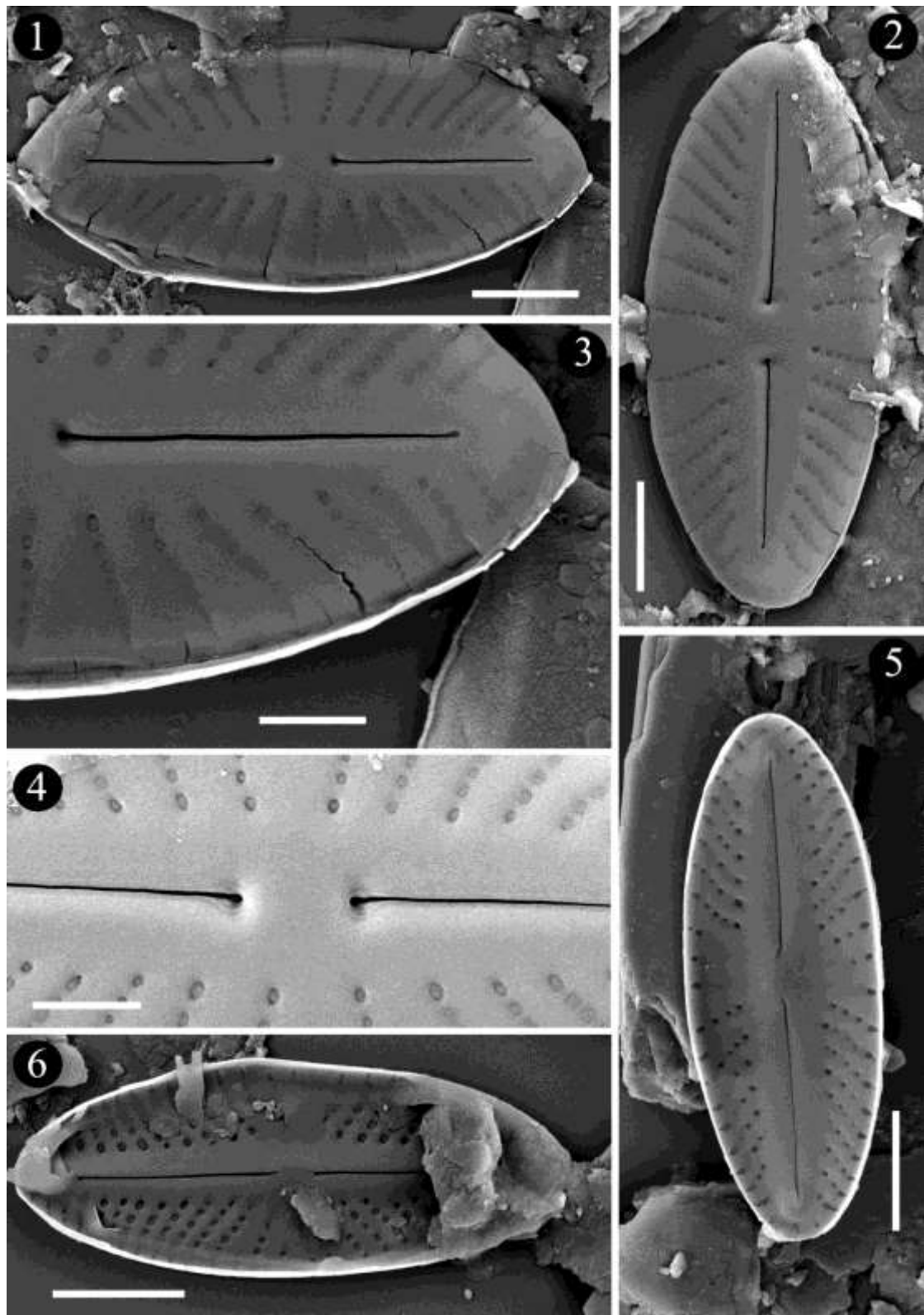


**PLATE 3: Figs 1–22.** LM micrographs of *Mayamaea excelsa*. **Figs 1–20.** LM micrographs of *Mayamaea excelsa* from the spring near fountain Sv. Petka, Mountain Osogovo. **Figs 21, 22.** SEM micrographs. Fig. 21. External view of the whole valve showing the stria and raphe morphology. Fig. 22. Internal view of the whole valve. LM Scale bar = 10  $\mu\text{m}$  (Figs 1–20). SEM scale bar = 2  $\mu\text{m}$  (Figs 21, 22).

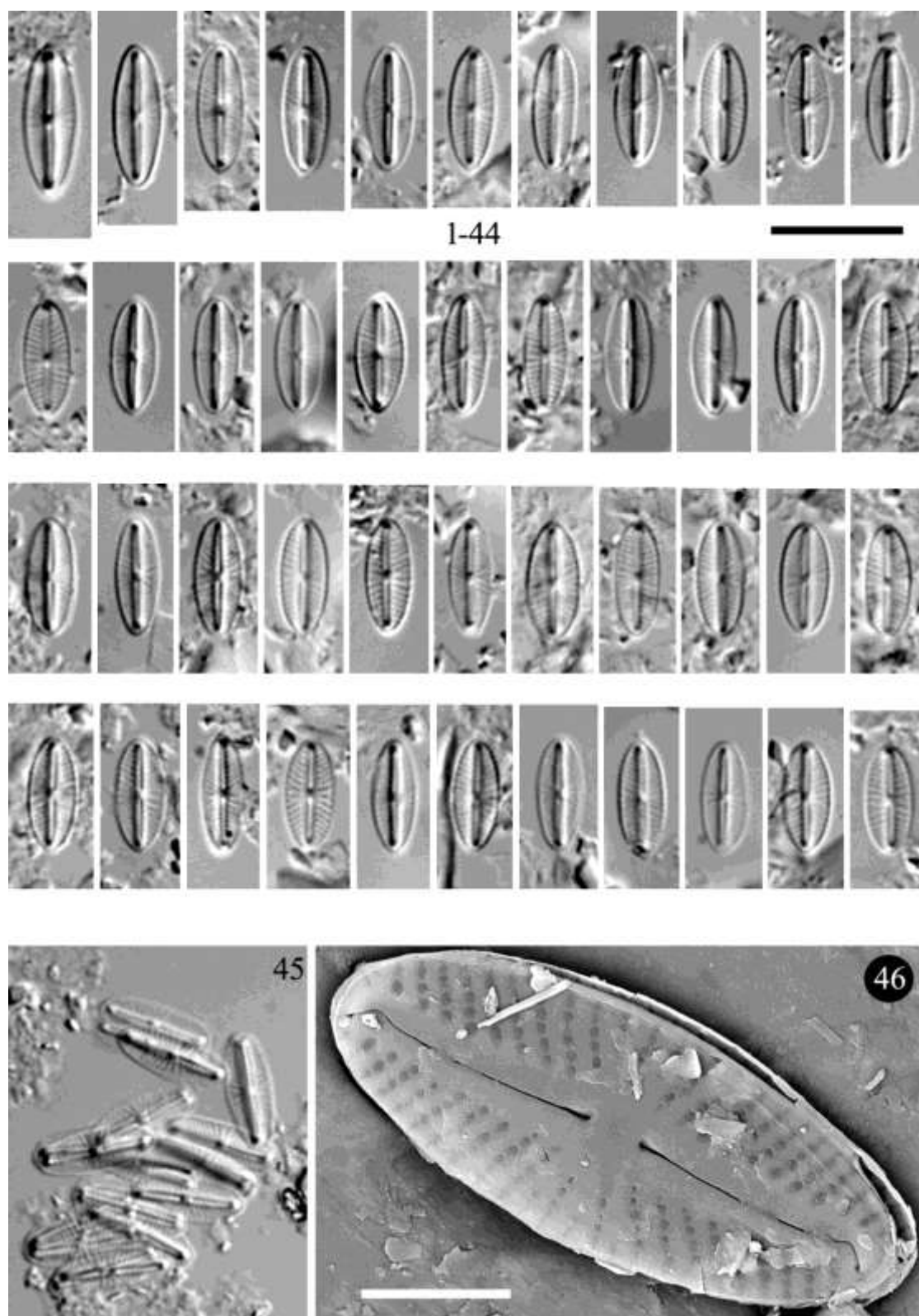




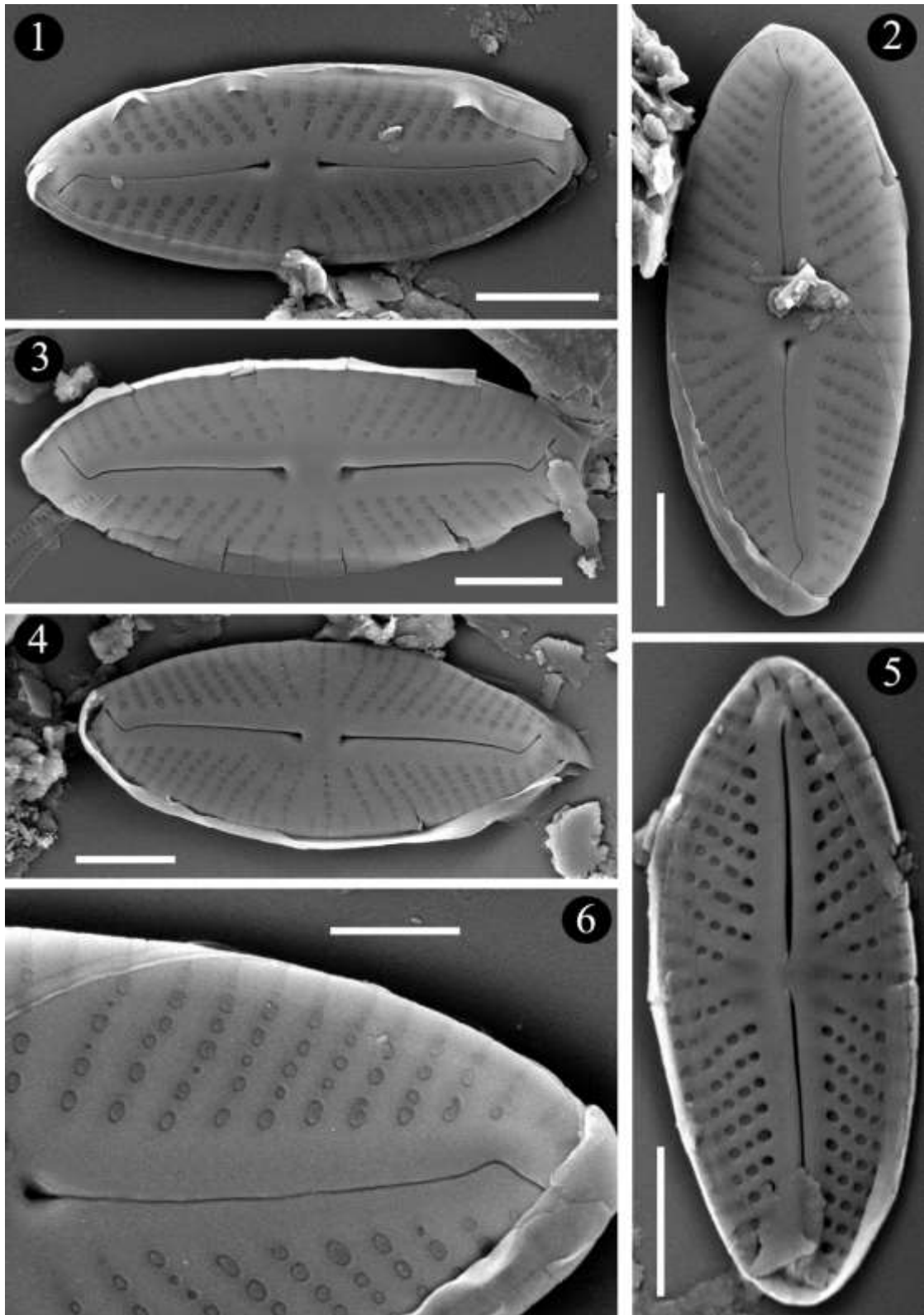
**PLATE 4: Figs 1–29.** LM and SEM micrographs of *Mayamaea matevskiana*, from type material (Acc. No. MKNDC 013814). **Figs 1–26.** LM micrographs of *M. matevskiana* showing the morphological variation. **Figs 27–29.** SEM micrographs. **Figs 27, 28.** External view of the whole valve. **Fig. 28.** Internal view of the whole valve. LM scale bar = 10  $\mu\text{m}$  (Figs 1–26); SEM scale bar = 2  $\mu\text{m}$  (Figs 27–29).



**PLATE 5: Figs 1–6.** SEM micrographs of *Mayamaea matevskiana*, from type material (Acc. No. MKNDC 013814). Figs 1, 2. External view of the whole valve. Fig. 3. Detailed external view showing the proximal and distal raphe ends and areolae occluded by hymens. Fig. 4. Detailed external view of the mid-valve showing the proximal raphe ends. Figs 5, 6. Internal view of the whole valve showing the slightly deflected proximal raphe ends and distal ends terminating in small helictoglossae. Scale bar = 2  $\mu$ m (Figs 1, 2, 5, 6); 1  $\mu$ m (Figs 3, 4).



**PLATE 6: Figs 1–46.** LM and SEM micrographs of *Mayamaea stafilovii* from type material (Acc. No. MKNDC 013814). **Figs 1–45.** LM micrographs of *M. stafilovii* showing the morphological variation. **Fig. 46.** SEM micrograph showing the external valve characters of *M. stafiloviana*. LM scale bar = 10  $\mu$ m (Figs 1–45); SEM scale bar = 2  $\mu$ m (Fig. 46).



**PLATE 7: Figs 1–7.** SEM micrographs of *Mayamaea stafilovii*, from type material (Acc. No. MKNDC 013814). Figs 1–4. External view of the whole valve showing the axial and central area, as well morphology of the raphe. Fig. 5. Internal view of the whole valve showing the slightly deflected proximal raphe ends and distal ends terminating in small helictoglossae. Fig. 6. External detailed view showing the proximal and distal ends, which are long and curved and run on the valve mantle. Areolae externally occluded by hymens. Scale bar = 2  $\mu\text{m}$  (Figs 1–5), 1  $\mu\text{m}$ . (Fig. 6).

**Acknowledgements.** This work was supported by the SYNTHESIS+ Project <http://www.synthesys.info/> which is financed by European Community Research Infrastructure Action under the H2020 Integrating Activities Programme, Project number 823827. The authors want to express their gratitude to Dr. David M. Williams the Natural History Museum, London, for his support and possibility to use SEM facilities in NHM. We thank Alex Ball and Innes Clatworthy, Imaging and Analysis Centre, the Natural History Museum, London, for their support with scanning electron microscopy.

## REFERENCES

- [1] F. E. Round, R. M. Crawford, D. G. Mann, *Diatoms: biology and morphology of the genera*, Cambridge University Press, 1990.
- [2] H. Lange-Bertalot, *Frankophila*, *Mayamaea* und *Fistulifera*: drei neue gattungen der klasse Bacillariophyceae, *Arch. Protistenk.*, **148** (1997), pp. 65–76.
- [3] K. Krammer, Die cybelloiden Diatomeen, Eine Monographie der weltweit bekannten Taxa, Teil 2, *Encyonema* Part, *Encyonopsis* und *Cymbellopsis*, *Bibliotheca Diatomologica*, **37** (1997), pp. 1–469.
- [4] K. Krammer, *Cymbopleura*, *Delicata*, *Navicymbula*, *Gomphocymbellopsis*, *Afrocybella*, *Diatoms of Europe*, *Diatoms of the European Inland waters and comparable habitats*, **4** (2003), pp. 1–529.
- [5] E. A. Morales, K. M. Manoylov, *Mayamaea cahabaensis* sp. nov. (Bacillariophyceae), a new freshwater diatom from streams in the southern United States, *Proc. Acad. Nat. Sci. Philadelphia*, **158** (2009), pp. 49–59.
- [6] B. Van De Vijver, E. J. Cox, New and interesting small-celled naviculoid diatoms (Bacillariophyceae) from a lava tube cave on Île Amsterdam (TAAF, Southern Indian Ocean), *Cryptogam.*, **34** (2013), pp. 37–47.
- [7] C. Barragán, L. Ector, C. E. Wetzel, *Mayamaea petersenii* sp. nov., a new diatom from European aerial habitats and a brief appraisal on the morphological diversity of the genus, *Algological Studies*, **153** (2017), pp. 71–87.
- [8] E. Kezlya, A. Glushchenko, J. P. Kociolek, Y. Maltsev, N. Martynenko, S. Genkal, M. Kulikovskiy, *Mayamaea vietnamica* sp. nov.: a new, terrestrial diatom (Bacillariophyceae) species from Vietnam, *Algae*, **35** (2020), pp. 325–335.
- [9] D. Vidaković, Z. Levkov, J. Krizmanić, B. Beszteri, B. Gavrilović, M. Ćirić, A new small-celled naviculoid diatom species, *Mayamaea panonica* sp. nov. (Bacillariophyceae) from soda pans in Serbia, *Phycologia*, **62** (2023), pp. 268–276.
- [10] F. Hustedt, Diatomeen aus Seen und Quellgebieten der Balkan-Halbinsel, *Arch. Hydrobiol.*, **40** (1945), pp. 867–973, 12 pls.
- [11] B. Van de Vijver, K. Kopalová, R. Zidarova, E. J. Cox, New and interesting small-celled naviculoid diatoms (Bacillariophyta) from the Maritime Antarctic Region, *Nova Hedwigia*, **97** (2013), pp. 189–208.
- [12] T. Bíró, M. Duleba, A. Földi, K. T. Kiss, P. Orgován, Z. Trábert, E. Vadkerti, C. E. Wetzel, É. Ács, Metabarcoding as an effective complement of microscopic studies in revealing the composition of the diatom community—a case study of an oxbow lake of Tisza River (Hungary) with the description of a new *Mayamaea* species, *Metabarcoding Metagenom.*, **6** (2022), pp. e87497.
- [13] F. Hustedt, Aërophile Diatomeen in der nordwestdeutschen Flora, *Ber. Dtsch. Bot. Ges.*, **60** (1942), pp. 55–73.
- [14] R. Zidarova, K. Kopalová, B. Van de Vijver, Ten new Bacillariophyta species from James Ross Island and the South Shetland Islands (Maritime Antarctic Region), *Phytotaxa*, **272** (2016), pp. 37–62.
- [15] H. Lange-Bertalot, *Navicula* sensu stricto. 10 genera separated from *Navicula* sensu lato, *Frustulia. Diatoms of Europe*, **2** (2001), pp. 1–526.
- [16] G. Hofmann, M. Werum, H. Lange-Bertalot, *Diatomeen im Süßwasser-Benthos von Mitteleuropa. Bestimmungsflora Kiesalgen für die ökologische Praxis, Über 700 der häufigsten Arten und ihre Ökologie*, A. R. G. Gantner Verlag K. G., Rugell, 2011.
- [17] M. H. Novais, M. M. Morais, J. Rosado, L. S. Dias, L. Hoffmann, L. Ector, Diatoms of temporary and permanent watercourses in Southern Europe (Portugal), *River Res. Appl.*, **30** (2014), pp. 1216–1232.
- [18] A. Dedić, A. Plenković-Moraj, K. Kralj Borjević, D. Hafner, The first report on periphytic diatoms on artificial and natural substrate in the karstic spring Bunica, Bosnia and Herzegovina, *Acta Bot. Croat.*, **74** (2015), pp. 393–406.
- [19] M. Cantonati, C. Casoria, R. Gerecke, O. P. Bilous, G. Maisto, S. Segadelli, D. Spitale, A. Steinbauer, S. Vogel, A. A. Saber, Diatom Indicators of Fluctuating/Intermittent Discharge from Springs in Two Bavarian Nature Conservation Areas, *Diversity*, **15** (2023), p. 915.
- [20] N. Angeli, M. Cantonati, D. Spitale, H. Lange-Bertalot, A comparison between diatom assemblages in two groups of carbonate, low-altitude springs with different levels of anthropogenic disturbances, *Fottea*, **10** (2010), pp. 115–128.
- [21] A. Beauger, E. Allain, O. Voldoire, C. E. Wetzel, L. Ector, B. Van de Vijver, Temporal evolution of diatoms in a temporary pond situated in the Massif du Sancy Mountains (Massif Central, France) and description of a new *Pinnularia* Species, *Diversity*, **12** (2020), p. 367.
- [22] C. Delgado, L. Ector, M. H. Novais, S. Blanco, L. Hoffmann, I. Pardo, Epilithic diatoms of springs and spring-fed streams in Majorca Island (Spain) with the description of a new diatom species *Cymbopleura margalefii* sp. nov., *Fottea*, **13** (2013), pp. 87–104.

- [23] A. Pavlov, Z. Levkov, Observations on the genus *Pinnularia* section *Distantes* (*Bacillariophyta*) from Macedonia; diversity and distribution, *Contributions, Sec. Math. Tech. Sci., MANU*, **34** (2013), pp. 33–57.
- [24] Z. Levkov, S. Tofilovska, C. E. Wetzel, D. Mitić-Kopanja, L. Ector, Diversity of *Luticola* D.G. Mann (*Bacillariophyceae*) species on halomorphic soils from Gladno Pole, Central Macedonia, *Nova Hedwig. Beih.*, **146** (2017), pp. 175–196.
- [25] Z. Levkov, D. Zaova, I. Jüttner, Three new cymbelloid species in the genera *Cymbella*, *Cymbopleura* and *Delicatophycus* from the mineral spring Crvena Voda near Skopje, Republic of North Macedonia, *Diatom Res.*, **38** (2023), pp. 165–179.
- [26] Z. Levkov, D. Zaova, *Halamphora ectorii* sp. nov. – A new diatom species from saline spring Solenica in North Macedonia, *Nova Hedwigia*, **38** (2023), pp. 165–179.
- [27] Z. Levkov, D. Vidaković, A. Cvetkoska, D. Mitić-Kopanja, S. Krstić, S., B. Van de Vijver, P. B. Hamilton, Observations of the genus *Muelleria* (*Bacillariophyceae*) from the Republic of North Macedonia, *Plant Ecol. Evol.*, **152** (2019), pp. 293–312.
- [28] E. Reichardt, *Die Diatomeen im Gebiet der Stadt Treuchtlingenn*, Bayerische Botanische Gesellschaft, 2018.
- [29] A. Schmidt, *Atlas der Diatomaceen-kunde*, Leipzig, O. R. Reisland Series VIII (Heft 99–100): pls. 393–400, 1934.
- [30] G. Hofmann, H. Lange-Bertalot, M. Werum, M. R. Klee, Rote Liste und Gesamtartenliste der limnischen Kieselalgen (*Bacillariophyta*) Deutschlands, In: *Rote Liste Gefährdeter Tiere, Pflanzen und Pilze Deutschlands, Band 7: Pflanzen*, D. H. N. Metzger, G. Ludwig, G., Matzke-Hajek, (Eds), Landwirtschaftsverlag: Münster, Germany, Naturschutz und Biologische Vielfalt, 2018, pp. 601–708.
- [31] H. Lange-Bertalot, K. Kulbs, T. Lauser, M. Norpel-Schempp, M. Willmann, Diatom taxa introduced by Georg Krasske: Documentation and revision, *Iconographia Diatomologica*, **3** (1996), pp. 1–358.
- [32] G. Krasske, Die Bacillariaceen-Vegetation Niederhessens, *Abhandl. Ber. Ver. Naturk.*, **56** (1925), pp. 1–119.
- [33] E. Reichardt, Die Diatomeen der Altmühl, *Bibliotheca Diatomologica*, **6** (1984), pp. 1–169.
- [34] K. Bruder, L. K. Medlin, Morphological and molecular investigations of naviculoid diatoms, II., Selected genera and families, *Diatom Res.*, **23** (2008), pp. 283–329.
- [35] F. Hustedt, Die Kieselalgen Deutschlands, Österreichs und der Schweiz unter Berücksichtigung der übrigen Länder Europas sowie der angrenzenden Meeresgebiete, Teil 3, In: *Kryptogamen Flora von Deutschland, Österreich und der Schweiz*, L. Rabenhorst (ed.), Akademische Verlagsgesellschaft m.b.h. Leipzig, 1961–1966.
- [36] F. Hustedt, Die Diatomeenflora des Flußsystems der Weser im Gebiet der Hansestadt Bremen, *Abh. Nat. wiss. Ver. Brem.*, **34** (1957), pp. 181–440.
- [37] K. Krammer, H. Lange-Bertalot, *Bacillariophyceae 1, Teil: Naviculaceae*, In: *Süßwasserflora von Mitteleuropa*, H. Ettl, H. J. Gerloff, H. Heynig & D. Mollenhauer et al. (Eds), VEB Gustav Fisher Verlag, Jena, Vol. **2** (1986), pp. 1–876, 206 pls., 2976 figs.
- [38] H. Lange-Bertalot, P. Cavacini, N. Tagliaventi, S. Alfinito, Diatom of Sardinia. Rare and 76 new species in rock pools and other ephemeral waters, *Iconographia Diatomologica*, **12** (2003), pp. 1–438.
- [39] J. Foets, C. E. Wetzel, A. J. Teuling, Pfister, Temporal and spatial variability of terrestrial diatoms at the catchment scale: controls on communities, *Peer J.*, **8** (2020), pp. e8296.
- [40] M. Rybak, T. Noga, R. Zubel, The aerophytic diatom assemblages developed on mosses covering the bark of *Populus alba* L., *J. Ecol. Eng.*, **19** (2018), pp. 113–123.
- [41] W. Bock, Diatomeen extreme trockener Standorte, *Nova Hedwigia*, **5** (1963), pp. 199–254, 3 pls.
- [42] J. R. Johansen, Diatoms of aerial habitats, In: *The Diatoms, Applications for the Environmental and Earth Sciences*, J. P. Smol, E. F. Stoermer (Eds.), 1999, pp. 264–273.
- [43] L. Ivarsson, M. Ivarsson, J. Lundberg, T. Sallstedt, C. Rydin, Epilithic and aerophilic diatoms in the artificial environment of Kungsträdgården metro station, Stockholm, Sweden, *Int. J. Speleol.*, **42** (2013), pp. 289–297.
- [44] E. Falasco, L. Ector, M. Isaia, C. E. Wetzel, L. Hoffmann, F. Bona, Diatom flora in subterranean ecosystems: a review, *Int. J. Speleol.*, **43** (2014), pp. 231–251.
- [45] T. Noga, J. Stanek-Tarkowska, N. Kochman-Kędziora, A. Pajaczek, Ł. Peszek, The inside of a dam as an unusual habitat for two rare species of *Gomphosphenia*–*G. fontinalis* and *G. holmquistii*, *Diatom Res.*, **31** (2016), pp. 379–387.
- [46] Z. Levkov, D. Metzeltin, A. Pavlov, *Luticola* and *Luticolopsis*, *Diatoms of Europe, Diatoms of the European Inland waters and comparable habitats*, **7** (2013), pp. 1–697.
- [47] S. Tofilovska, C. E. Wetzel, L. Ector, Z. Levkov, Observation on *Achnanthes* Bory sensu stricto (*Bacillariophyceae*) from subaerial habitats in Macedonia and comparison with the type material of *A. coarctata* (Brébisson ex W. Smith) Grunow, *A. coarctata* var. *sinaensis* Hustedt and *A. intermedia* Kützing, *Fottea*, **14** (2014), pp. 15–42.
- [48] E. Falasco, E. Piano, F. Bona, Diatom flora in Mediterranean streams: flow intermittency threatens endangered species, *Biodivers. Conserv.*, **25** (2016), pp. 2965–2986.

- [49] M. Cantonati, O. Bilous, D. Spitale, N. Angeli, S. Segadelli, D. Bernabè, K. Lichtenwöhler, R. Gercke, A. Saber, Diatoms from the spring ecosys-

tems selected for the long-term monitoring of climate-change effects in the Berchtesgaden National Park (Germany), *Water*, **14** (2022), p.381.

## НОВИ И ИНТЕРЕСНИ MAYAMAEA (BACILLARIOPHYCEAE) ВИДОВИ ОД ПЛАНИНАТА ОСОГОВО

Златко Левков, Душица Заова, Данијела Митиќ-Копанџа

<sup>1</sup>Институт за биологија, Природно-математички факултет,  
Универзитет „Св. Кирил и Методиј“ во Скопје, Република Северна Македонија

Анализите на примерок од мал извор од планината Осогово покажаа присуство на неколку интересни видови, како и видови со мали димензии. Помеѓу нив, родот *Mayamaea* покажа релативно висока видова разновидност со осум видови. Во рамките на оваа студија се презентирани четири вида, од кои два се опишани како нови *Mayamaea matevskiana* sp. nov. and *Mayamaea stafilovii* Levkov & Zaova sp. nov. *Mayamaea matevskiana* се карактеризира со елиптични до елиптично-ланцентни валви, релативно груби и силно радијални стрии, незабележителни централно поле ограничено со централни стрии кои се пораздалечени, слабо проширени централни краеве на рафата и едноставни дистални краеве на рафата. *Mayamaea stafilovii* поседува слична морфологија и облик на централното поле како *M. matevskiana*, но може да се забележи разлика во големината на валвата (значително помали димензии во однос на *M. matevskiana*) и обликот на надворешните дистални краеве на рафата (долги, слабо закривени и преминуваат на страничниот дел на валвата). Новите видови се опишани врз основа на детални анализи на светлосен и електронски микроскоп.

**Клучни зборови:** дијатомеи; разновидност; морфологија; извори; таксономија