Observations on the type material of *Melosira roeseana* Rabenhorst and *Orthoseira spinosa* W.Smith (*Orthoseiraceae, Bacillariophyta*)

Natalia Kochman-Kędziora, *Department of Ecology and Environmental Protection, University of Rzeszów, Zelwerowicza 4, 35-601 Rzeszów, Poland*

Wolf-Henning Kusber, *Botanischer Garten und Botanisches Museum Berlin, Freie Universität Berlin, Königin-Luise-Str. 6-8, 14195 Berlin, Germany*

J. Patrick Kociolek, *Museum of Natural History and Department of Ecology and Evolutionary Biology, University of Colorado, Boulder, CO, USA 80309*

Bart Van de Vijver, *Meise Botanic Garden, Research Department, Nieuwelaan 38, 1860 Meise, Belgium & University of Antwerp, Department of Biology – ECOSPHERE, Universiteitsplein 1, B-2610 Wilrijk, Belgium* (correspondence: bart.vandevijver@plantentuinmeise.be)

Melosira roeseana Rabenhorst (1853: 13) was originally described from wet rocks in a waterfall in the Schnepfenthal (Thuringia, Germany). He illustrated his new species with two small drawings (Fig. 1) and added the following description: "*bis 2-100 Mm. im Durchmesser; Glieder länger als breit, mit zwei breiten Ringfurchen, an den Gelenken gestreift; Hauptseiten strahlig gestreift.*" [up to 50 µm in diameter, valves longer than wide, with two wide annular grooves, striped on the connection of two valves; valve face with radiate striae]. The description was based on material collected by August D.F.C. Röse (1821–1873) in October 1853 and published later in Rabenhorst's exsiccata series "*Die Algen Europa's, Fortsetzung der Algen Sachsens, resp. Mittel-Europa's*" as number 383 (distributed in September 1854, Decade 39, n° 383, Sayre 1969).

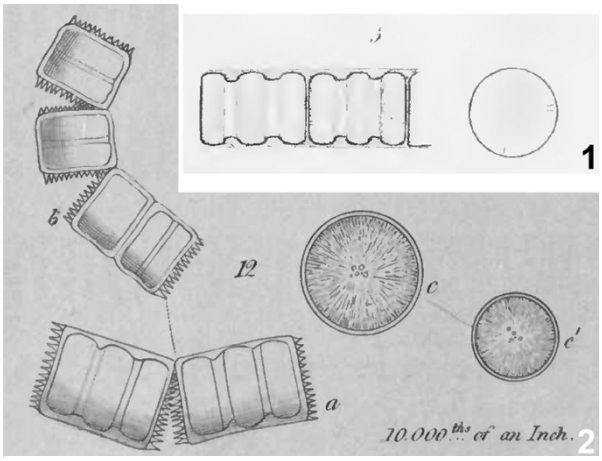


Fig. 1. Original drawing of *Melosira roeseana* taken from Rabenhorst (1853, pl. X [10], supplement: fig. 5. **Fig. 2.** Original drawing of *Orthoseira spinosa* taken from Smith (1855, fig. 12).

Three years after the description of *M. roeseana*, Smith (1856: 62) added it as a synonym to his new species *Orthos(e)ira spinosa* W.Smith (Fig. 2), described a year previously in 1855 during his trip to the Auvergne region in France (Smith 1855: 8). Rabenhorst (1853) did not mention the presence of the distinct central pores (known presently as "carinoportulae", a term introduced in 1981 by R. M. Crawford), considered one of the most conspicuous features of this species. It was Smith (1856: 62) who mentioned the presence of "*three conspicuous subcentral puncta*". Ralfs (in Pritchard 1861: 818) switched the synonyms, with *O. spinosa* now being a younger, heterotypic, synonym for *Melosira roeseana*, and added the presence of "*three or more central dots*" in the central area for *M. roeseana*. Pfitzer (1871: 134) also confirmed this conspecificity but transferred *M. roeseana* to the genus *Orthoseira* as *O. roeseana* (Rabenhorst) Pfitzer. Four years later, O'Meara (1875) proposed the isonym *O. roeseana* (Rabenhorst) O'Meara, confusing later authors such as Krammer & Lange-Bertalot (1991: 13) who used the latter. Houk & al. (2017: 33) corrected it stating that the O'Meara combination should be considered a "superfluous name".

However, the identity of the genus *Orthoseira*, described in 1848 by Thwaites, is unclear since the generitype is *Melosira americana* Kützing [≡ *Orthoseira americana* (Kützing) Round, R.M.Crawford & D.G.Mann ex Spaulding & Kociolek], a species described from South America that lacks the carinoportulae (Kützing 1844). A few months later than Thwaites (1848), Ehrenberg (1848: 217) described three new genera, *Liparogyra*, *Porocyclia* and *Stephanosira*, from some soil tree root samples collected in Venezuela and Brazil, each of them possessing "*apiculi centrali*", most likely indicating for the first time the presence of carinoportulae. However, since only the original micas Ehrenberg prepared are available of the material from which he described the genera, observations on the features of each is difficult. Danz & al. (2022) discussed in detail the history of the genus *Orthoseira* recommending the use of the name *Orthoseira* only for *M. americana*, a thorough revision of all *Orthoseira* taxa, and a phylogenetic analysis of all known taxa.

Orthoseira roeseana is probably one of the most widespread species in the genus with even recently published records showing a worldwide distribution from Asia, Europe, North America, South America, and the Antarctic realm (e.g. Houk 1993, Houk & al. 2017, Radhakrishnan & al. 2022, Rodrigues Maciel & al. 2022, Van de Vijver & al. 2002). Part of this seemingly cosmopolitan distribution most likely originates from the lumping of several (former) *Melosira* taxa described in the nineteenth century as varieties of *M. roeseana*. Grunow (in Van Heurck 1882) listed several such as *M. roeseana* var. *spiralis* (Ehrenberg) Grunow. Houk (1993) distinguished several "morphotypes" within *O. roeseana* without formally separating them (although some were later described as separate species such as *O. tatrica* Houk & al. 2017). Another reason for the force-fitting of *Orthoseira*-populations into *O. roeseana* is the incorrect interpretation of the original drawings, probably because the absence of a thorough analysis of the original Rabenhorst sample 383 material did not allow for a good comparison with what was described originally under the name *roeseana*. Crawford (1981, fig. 41) published a single SEM image of the type of *O. roeseana*, together with a large number of *O. roeseana* valves from different populations. A detailed morphological analysis of the type of *Orthoseira roeseana*, is thus lacking.

One of the many widely distributed copies of the Rabenhorst exsiccata series is kept in the Van Heurck collection, part of the Meise Botanic Garden (**BR**) herbarium. Unmounted original material for Rabenhorst sample 383 was retrieved and prepared for light and scanning electron microscopy. Additionally, unmounted material collected by William Smith during his excursion to the Auvergne in France in 1854, and used for the description of *Orthoseira spinosa*, was present in the William Smith collection, part of the Van Heurck collection also kept in **BR**. Three samples were listed as syntypes in Smith (1855): "Cave near Royat [France]; Cave under Grand Cascade, Mont Dore, elevation 4236 feet [France]. Braemar [Aberdeenshire, Scotland], Aug.1854, Dr. Balfour". All three samples were present in the Van Heurck collection and prepared for further analysis. This analysis showed that two species were present within the syntypes. The sample from Cave near Royat, collected in a small village close to the city of Clermont-Ferrand (Auvergne, France) contained a population that shows similar morphological features to the population in Rabenhorst sample 383 (Figs 36–47). However, the other two populations, Mont Dore and Braemar (Figs 22–35), present a totally different morphology, excluding conspecificity with the Royat population and the Rabenhorst type population of *Melosira roeseana*. Smith (1855: 8) provided a description for *O. spinosa* stating "*Filament fragile, often only partially cohering; valves cylindrical, spinose at the line of junction, striated ; striae moniliform, radiate, 30 in ·001". Breadth of filament ·0005" to ·0017".*" Smith also illustrated his species with several drawings of both the valve face and the girdle view (Fig. 2). The drawings correspond with the populations from the Mont Dore and Braemar in Scotland. Moreover, Greville (1855: 260) commented on the morphology of *O. spinosa*, based solely on the populations of Mont Dore and Braemar, but never mentioned the Royat population. He was also not able to see the spine-like processes in the original material ("…but I

confess that I have been unable to make out this character to my satisfaction."). Smith (1856: 62) subsequently sent material to Greville who then replied that he had observed the spines. Given these comments and observations, and comparison of the original drawings with the different analysed populations of *O. spinosa*, it is clear that the Royat population is not representative of the protologue of *O. spinosa* and therefore, a lectotype should be chosen from the other syntypes. We here designate formally the Mont Dore population as lectotype for *Orthoseira spinosa*. This choice also leads to the conclusion that *Orthoseira spinosa* and *O. roeseana* are not conspecific, contrary to what has been accepted generally for more than 150 years.

Here, we detail morphological observations on the type populations of both *Orthoseira (Melosira) roeseana* and *Orthoseira spinosa* from slides prepared from the original Rabenhorst and William Smith samples, kept in **BR**, using light and scanning electron microscopy. The material from Schnepfenthal (Rabenhorst sample 383) is designated as lectotype for *Orthoseira roeseana* and the material from Mont Dore is designated as lectotype for *O. spinosa*. The *O. spinosa*-population from Cave Royat is illustrated to show its conspecificity with *O. roeseana*.

Orthoseira roeseana (Rabenhorst) Pfitzer, 1871 (Figs 3–21, 36–47)
Basionym: *Melosira roeseana* Rabenhorst 1853: 13, pl. X [10], supplement: fig. 5
Synonyms: *Melosira roseana* var. *typica* Grunow 1880, *nom. inval., Gaillonella roeseana* (Rabenhorst) Petit 1880, *Lysigonium roeseanum* (Rabenhorst) Kuntze 1891, *Melosira dendroteres* var. *roeseana* (Rabenhorst) R.Ross 1947
Lectotype (here designated): BR-4781, slide prepared from Rabenhorst sample 383, Schnepfenthal, Thuringia, Germany, leg. August Röse, Oct. 1853, original material present in the Van Heurck collection (**BR**). The lectotype is represented by Fig. 8.
Registration (of lectotypification): http://phycobank.org/103630
Description: Frustules cylindrical, connected to each other by plate-like linking structures, forming long filaments (Fig. 14). Valve diameter 18–30 µm. Plate-like structures surrounding the entire valve face, irregularly shaped and continuing shortly onto the mantle as weakly raised ridges. Cingulum composed of a large number of broad, perforated copulae (Fig. 14). Valvocopula with thin, broad extensions, covering plate-like the valve interior leaving only the central carinoportulae uncovered (Figs 13 & 21). All copulae open bearing a long ligula and fimbriate edge (Fig. 14). Mantle moderately deep, divided into two parts (Fig. 15). Mantle edge forming a narrow hyaline band, perforated by up to 3 rows of small, rounded areolae (Figs 15 & 16). Largest part of the mantle composed of an irregular pattern of short, flattened ridges. Mantle areolae organized in uniseriate series of rounded, fairly large areolae. Valve face flat, at least the central part surrounded by a ring of deep grooves, radially arranged on the valve face/mantle junction (Figs 17 & 18). In the grooves, one series of large, rounded areolae present. Ridges separating the grooves extending as the plate-like linking structures (Fig. 16). Central area covered by irregular pattern of small siliceous plates, covering the carinoportulae (Fig. 18). When eroded, underlying carinoportula visible (Fig. 17). Areolae covered in the valve interior by individual hymenate vela. 2–4 large carinoportulae present in the central area. Between the carinoportulae, several long slits present (Fig. 20).

Orthoseira spinosa W.Smith in *Annals and Magazine of Natural History, series 2* 15: 8, pl. I: fig. 12, 1855. [*'Orthosira'*] (Figs 22–35)
Lectotype (here designated): BR-4782, slide prepared from William Smith sample Grand Cascade, Mont Dore les Bains, Auvergne, France, leg. William Smith, coll. date 13.vi.1853, original material present in the Van Heurck collection (**BR**). The lectotype is represented by Fig. 22.
Registration (of lectotypification): http://phycobank.org/103631

Description: Frustules cylindrical, connected to each other by robust, conical linking spines extending from small plate-like bases, forming short, loose filaments. Linking spines grouped in short bundles, separated by areas with larger, rimmed areolae extending from the mantle onto halfway the valve face (Figs 29–32). Cingulum composed of several broad, open, perforated, ligulate copulae (Fig. 29). Valvocopula (Figs 30, 35) with thin, hyaline pars interior lacking extensions covering the valve interior. Valvocopula edge with very short, thin fimbriae (Fig. 35). Mantle very deep with a clear step halfway its length. Mantle areolae organized in dense pattern of parallel, uniseriate striae. Mantle edge hyaline, lacking perforations but covered by irregular network of very thin ridges (Fig. 30). Valve diameter 30–45 µm. Valve face covered by irregular pattern of raised ridges forming a network in the central area and radiating towards the valve face edge (Figs 31, 32). Areolae placed in between the ridges, forming an unclear radiating pattern of striae. Up to four carinoportulae visible, externally opening via an irregular dense siliceous structure (Figs 31, 32). Internally carinoportula foramina large, rounded, filled by siliceous aggregations. Series of very small wart-like papillae present around each carinoportula (Fig. 34). Multiple small slits present between the carinoportula (Fig. 34). External dense groups of rimmed areolae also visible in valve interior as groups of small pores, located between the normal areolae (Fig. 33).

The larger valves of *Orthoseira spinosa* are differentiated from the smaller *O. roeseana* in having clusters of tapering spines (versus a ring of spines around the circumference of the valve). In *O. roeseana* areolae on the valve face are similar while in *O. spinosa* there are groups of areolae that differ in size.

This research received support (Natalia Kochman-Kędziora) from the SYNTHESYS project (www.synthesys.info), financed by the H2020 Research Infrastructures Programme, to visit Meise Botanic Garden.

Crawford, R.M. (1981). The diatom genus *Aulacoseira* Thwaites: its structure and taxonomy. *Phycologia* 20: 174–192.

Danz, A., Van de Vijver, B., Kochman-Kędziora, N., Kusber, W.-H., Guerrero, J., Vouillous, A., Salam S.E., Liu, Q. & Kociolek, J.P. (2022). The genus *Orthoseira* Thwaites: historical overview, current status and recommendations for the future. *Nova Hedwigia* 115: 1–29.

Ehrenberg, C.G. (1848). Über eigentümliche auf den Bäumen des Urwaldes in Süd-Amerika zahlreich lebende mikroskopische oft kieselschalige Organismen. *Bericht über die zur Bekanntmachung geeigneten Verhandlungen der Königlich-Preussischen Akademie der Wissenschaften zu Berlin* 1848: 213–220.

Greville, R.K. (1855). Report on a collection of Diatomaceae made in the district of Braemer by Professor Balfour and Mr. George Lawson. *The Annals and Magazine of Natural History, Series 2*, 15: 252–261.

Houk, V. (1993). Some morphotypes in the *Orthoseira roeseana* complex. *Diatom Research* 8(2): 385-402.

Houk, V., Klee, R. & Tanaka, H. (2017). Atlas of freshwater centric diatoms with a brief key and descriptions. Second emended edition of Part I and II. Melosiraceae, Liparogyraceae, Paraliaceae and Aulacoseiraceae. *Fottea* 17(Supplement): 1–616.

Krammer, K. & Lange-Bertalot, H. (1991). *Bacillariophyceae. 3 Teil: Centrales, Fragilariaceae, Eunotiaceae*. In: *Süsswasserflora von Mitteleuropa Band 2/3*. pp. [i]–xiii, [1]–576. Stuttgart & Jena: Gustav Fischer Verlag.

Kützing, F.T. (1844). *Die Kieselschaligen Bacillarien oder Diatomeen*. pp. [i–vii], [1]–152.

O'Meara, E. (1875). Report on the Irish Diatomaceae. *Proceedings of the Royal Irish Academy, Series 2* 2: 235–425.

Pfitzer, E. (1871). Untersuchungen über Bau und Entwicklung der Bacillariaceen (Diatomaceen). In: *Botanische Abhandlungen aus dem Gebiet der Morphologie und Physiologie* Bd 2. pp. [i]-vi, [1]–189.

Pritchard, A. (1861). *A history of infusoria, including the Desmidiaceae and Diatomaceae, British and foreign.* Fourth edition enlarged and revised by J. T. Arlidge, M.B., B.A. Lond.; W. Archer, Esq.; J. Ralfs, M.R.C.S.L.; W. C. Williamson, Esq., F.R.S., and the author. pp. [1]–xii, [1]–968.

Rabenhorst, L. (1853). *Die Süsswasser-Diatomaceen (Bacillarien.): für Freunde der Mikroskopie.* pp. [1]–xii, [1]–72.

Radhakrishnan, C., Yogeshwaran, M. & Karthick, B. (2022). Hanging in the air: tree moss diatoms from Indo-Burma biodiversity hot spot of India. *Aerobiologia* 38: 557–566.

Rodrigues Maciel, M.G., Pereira K. & Veiga Ludwig, T.A. (2022). Unravelling centric diatoms from the Caatinga: Coscinodiscophyceae and Mediophyceae in northwestern Ceará, Brazil. *Rodriguésia* 73: e00222021.

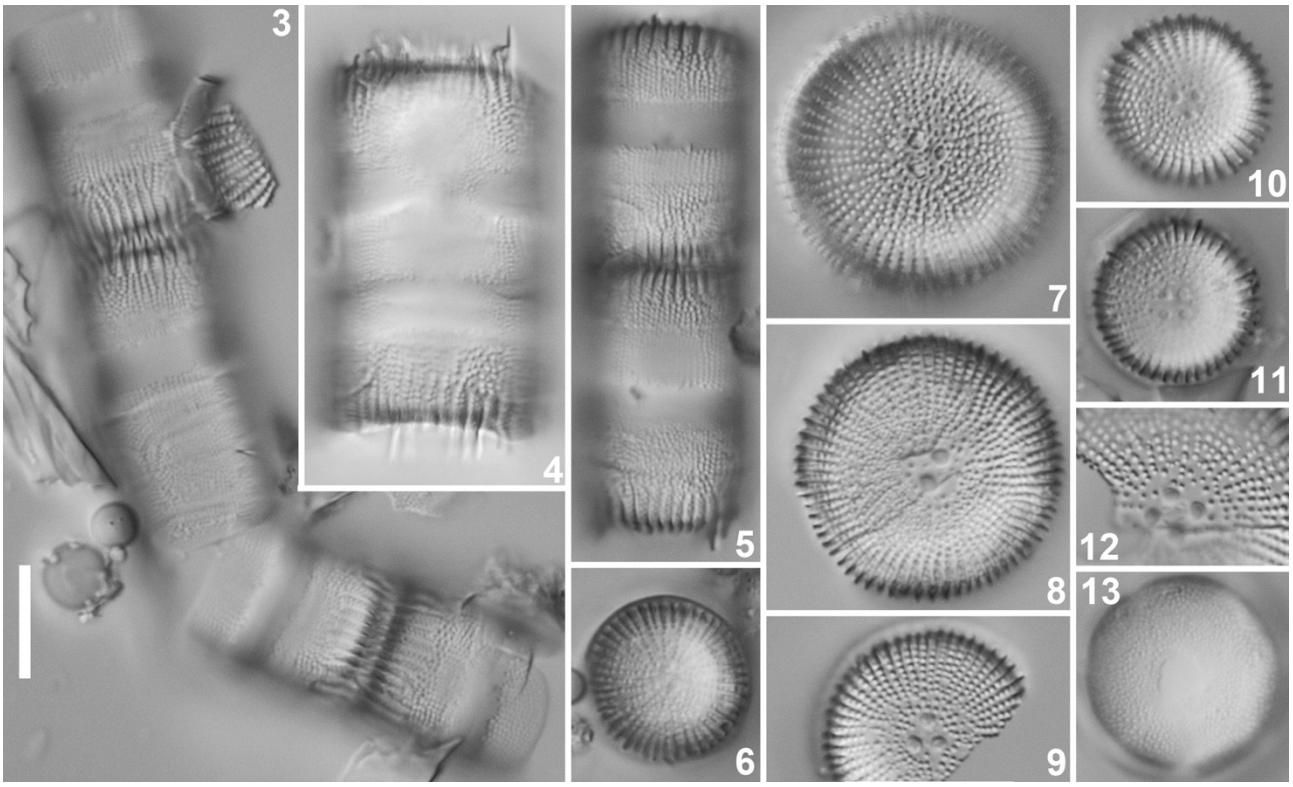
Smith, W. (1855). Notes of an excursion to the south of France and the Auvergne in search of Diatomaceae. *Annals and Magazine of Natural History, series 2* 15: 1–9.

Smith, W. (1856). *A synopsis of the British Diatomaceae; with remarks on their structure, functions and distribution; and instructions for collecting and preserving specimens.* The plates by Tuffen West. In two volumes. Vol. II. pp. [i]-xxix, 1–107.

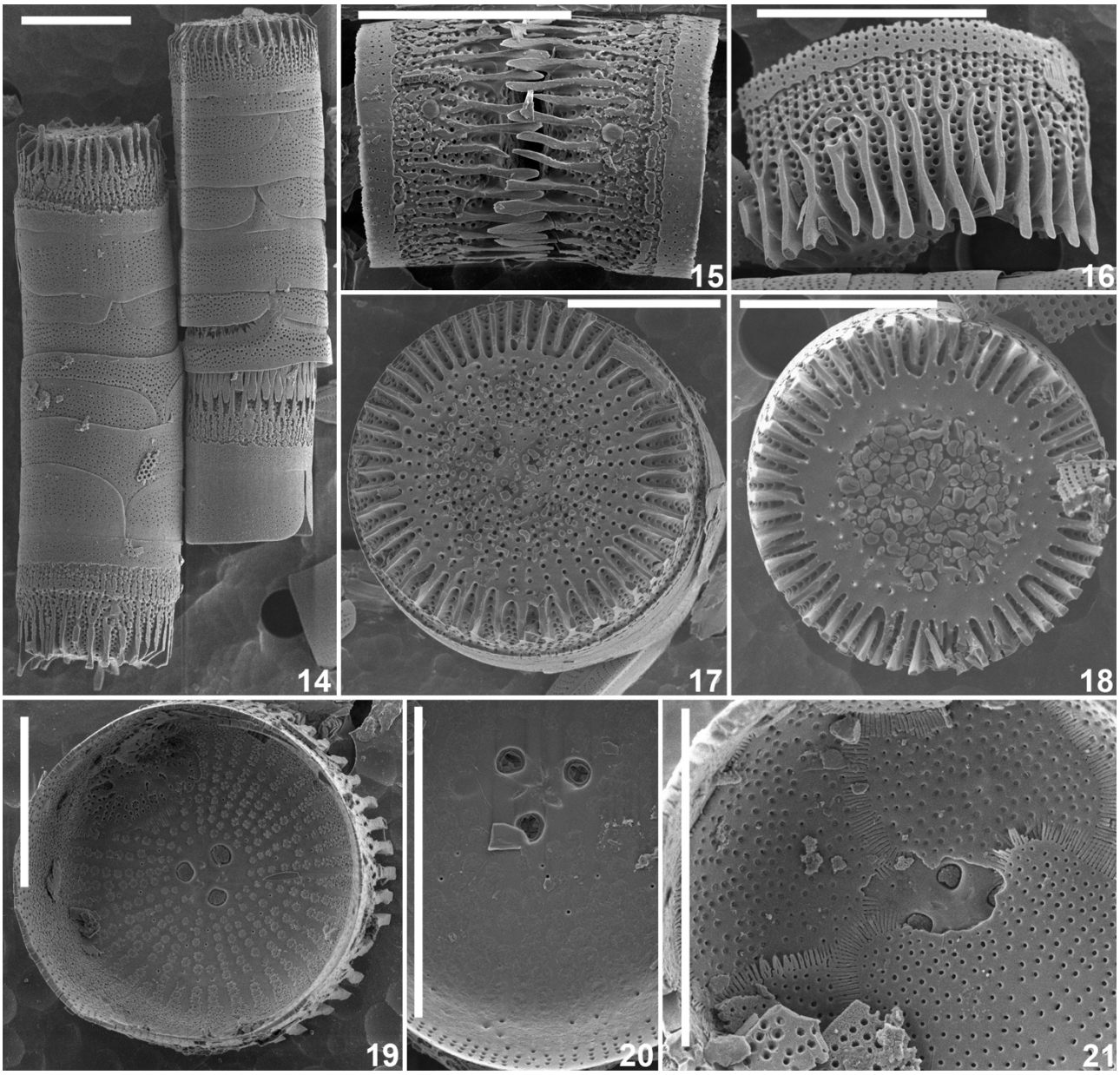
Thwaites, G.H.K. (1848). Further observations on the Diatomaceae with descriptions of new genera and species. *Annals and Magazine of Natural History, Series 2* 1: 161–172.

Van de Vijver, B., Frenot, Y. & Beyens, L. (2002). Freshwater diatoms from Île de la Possession (Crozet Archipelago, sub-Antarctica). *Bibliotheca Diatomologica* 46: 1–412.

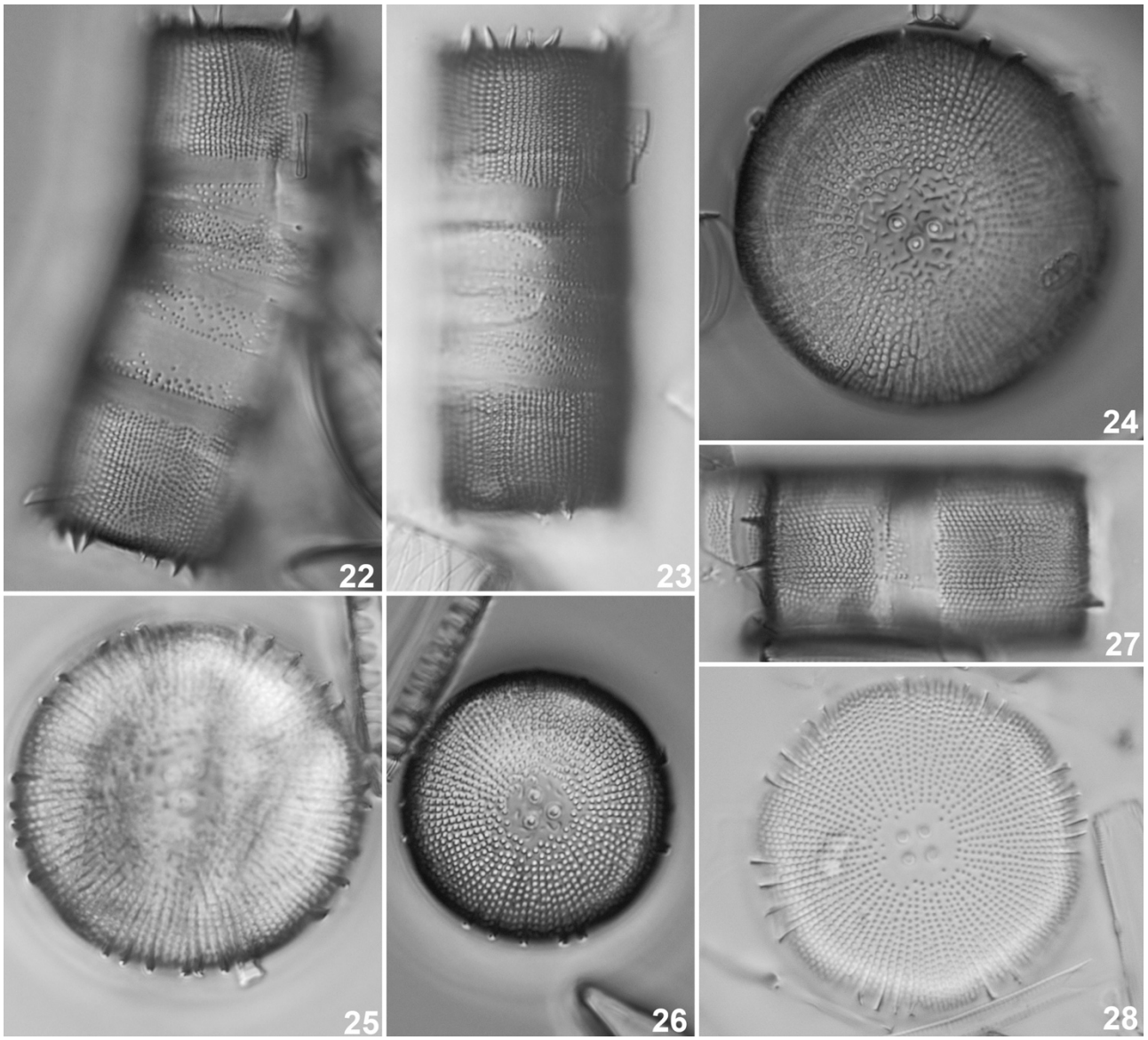
Van Heurck, H. (1882) *Synopsis des Diatomées de Belgique Atlas*. pp. pls LXXVIII-CXXXII [78-132 and suppl. pls 22 bis, 82bis, 83bis, 83ter, 95bis], tables genres. Anvers: Ducaju et Cie.



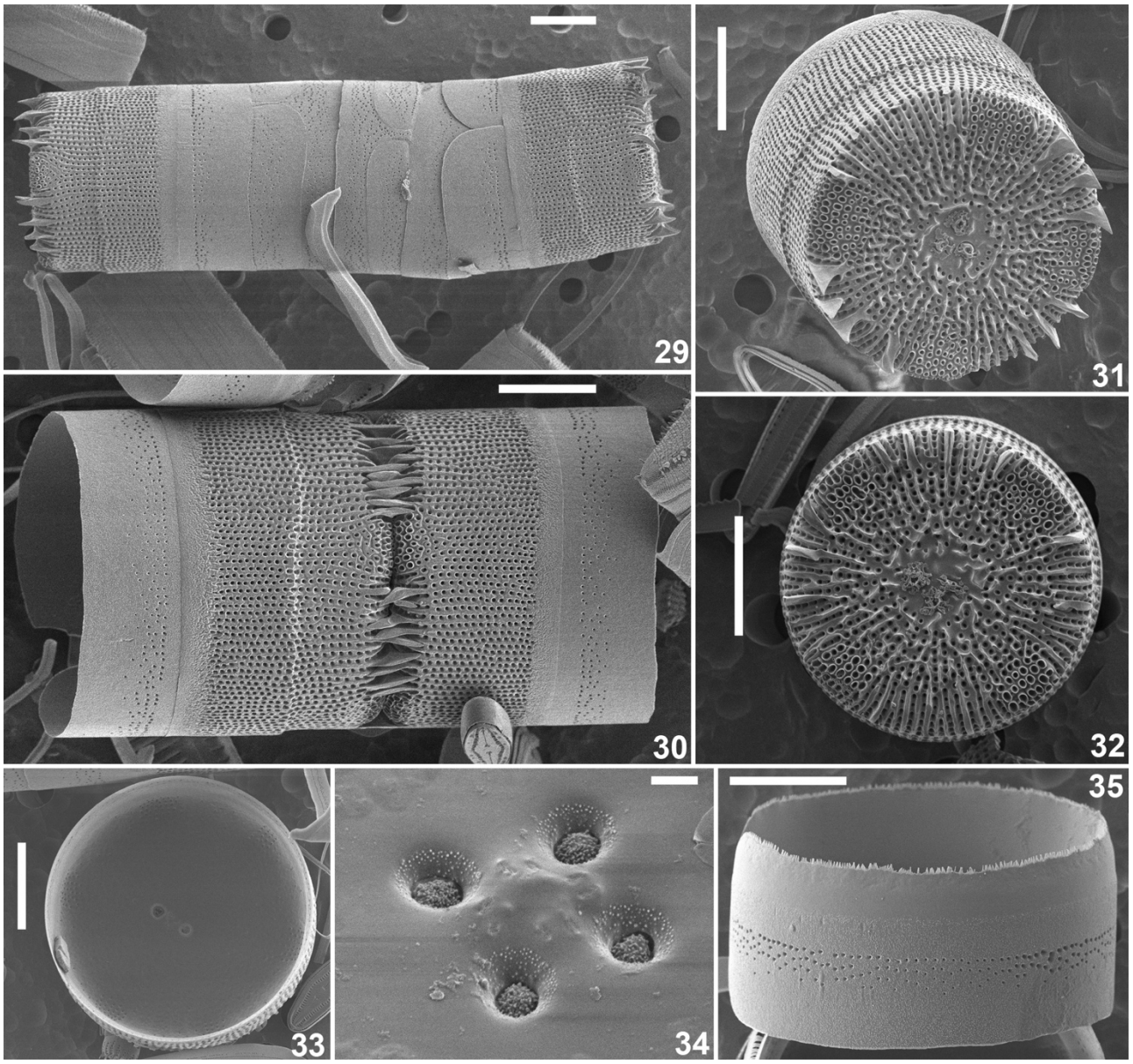
Figs 3–13. *Orthoseira roesena* (Rabenhorst) Pfitzer. LM pictures taken from the lectotype material (BR-4781, Schnepfenthal, Thuringia, Germany, Rabenhorst sample 383). Figs 3–5. Frustules in girdle view connected to each other via plate-like linking structures. Figs 6–12. LM pictures of valves in valve face view showing the variability in number of carinoportulae. Fig. 13. LM view of the valvocopula. Scale bar = 10 µm.



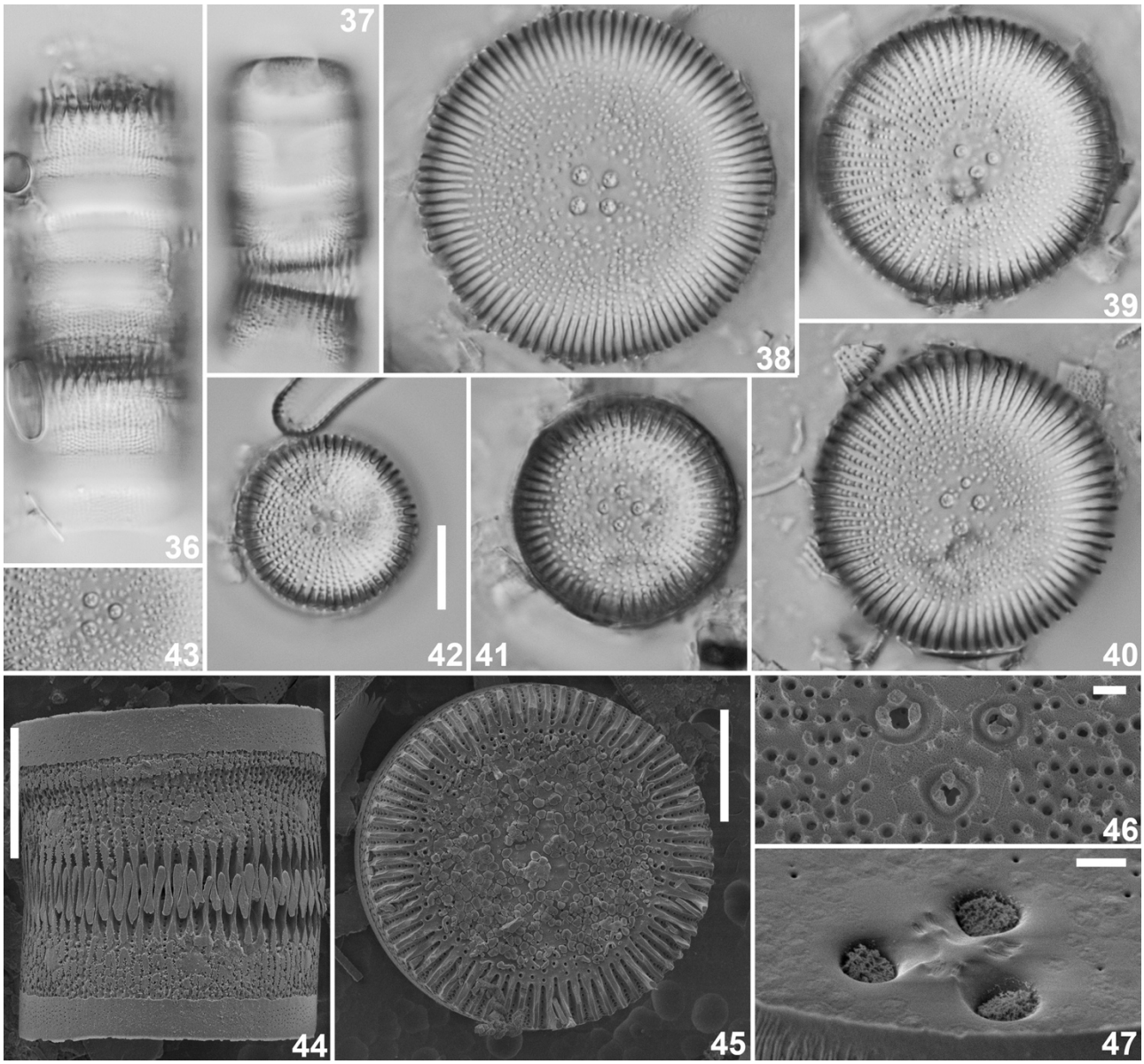
Figs 14–21. *Orthoseira roesena* (Rabenhorst) Pfitzer. SEM pictures taken from the lectotype material (BR-4781, Schnepfenthal, Thuringia, Germany, Rabenhorst sample 383). Fig. 14. Two short filaments of connected frustules showing the structure of the linking plates and the cingulum. Fig. 15. Two valves connected to each other by linking plates. Note also the hyaline mantle edge with several rows of small areolae and the irregular pattern of flattened ridges on the mantle. Fig. 16. Detail of a broken valve in oblique view highlighting the structure of the linking plates. Figs 17 & 18. SEM external views of the valve face. Fig. 19. SEM internal view of a complete valve. Fig. 20. SEM internal detail of the carinoportulae showing the slits between the carinoportulae and the areola coverings. Fig. 21. SEM view of the valvocopula. Scale bars = 10 µm.



Figs 22–28. *Orthoseira spinosa* W.Smith. LM pictures taken from the lectotype material (BR-4782, Grande Cascade, Mont Dore, Auvergne, France, leg. W. Smith, Figs 22–26) and sample Walker Arnott S142 (Braemar, leg. Prof. Balfour, Figs 27 & 28). Figs 22–23, 27. Frustules in girdle view showing the girdle structure and the marginal spines. Figs 24–26, 28. LM pictures of valves in valve face view showing the variability in number of carinoportulae. Scale bar = 10 µm.



Figs 29–35. *Orthoseira spinosa* W.Smith. SEM pictures taken from the lectotype material (BR-4782, Grande Cascade, Mont Dore, Auvergne, France, leg. W. Smith). Fig. 29. A frustule in girdle view showing the structure of the linking spines and the cingulum. Fig. 30. Two valves connected to each other by linking spines. Note also the large mantle and the dense groups of rimmed areolae separating the bundles of spines. Fig. 31. SEM view of a valve in oblique view showing clearly the valve mantle, the groups of linking spines and the group of rimmed areolae extending from mantle to valve face. Figs 31 & 32. SEM external views of the valve face. Fig. 33. SEM internal view of a complete valve. Fig. 34. SEM internal detail of the carinoportulae showing the slits between the carinoportulae and the small papillae around each carinoportula. Fig. 35. SEM view of the valvocopula. Scale bars = 10 µm except for Fig. 34 where scale bar = 1 µm.



Figs 36–47. *Orthoseira roesena* (Rabenhorst) Pfitzer originally identified as *O. spinosa* W.Smith. LM and SEM pictures taken from the Cave Royat sample (Auvergne, France, William Smith collection in **BR**!). Figs 36–37. Frustules in girdle view connected to each other via plate-like linking structures. Figs 38–40. LM pictures of valves in valve face view showing the variability in number of carinoportulae. Fig. 41. LM picture of the central area. Fig. 44. SEM view of two valves connected to each other by linking plates. Fig. 45. SEM external view of the valve face. Fig. 46. SEM external detail of the carinoportulae after removal of the siliceous plates. Fig. 47. SEM internal detail of the carinoportulae showing the slits between the carinoportulae. Scale bars = 10 µm except for Figs 46 & 47 where scale bars = 1 µm.