Denticula jamesrossensis, a new freshwater diatom (Bacillariophyta) species from the Maritime Antarctic Region

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Abstract: During a survey of the freshwater diatom flora of James Ross Island (northern Weddell Sea) in the Maritime Antarctic Region, an unknown *Denticula* taxon was found in some lakes of Clearwater Mesa. The taxon showed some similarities with *D. rainierensis* Sovereign and *D. subtilis* Grunow but based on detailed light (LM) and scanning electron microscopy (SEM) the species showed sufficient morphological differences to separate it as a new species: *D. jamesrossensis* sp. nov. The new taxon is characterized, and distinguished from other species of the genus, by a typical constriction in the middle of the valve and a very high number of striae (up to 60 in $10 \mu m$). So far, the new taxon has only been found on James Ross Island where it occurs in alkaline lakes with high specific conductance values.

Key words: Denticula, Maritime Antarctic Region, morphology, new species

Introduction

During the past few years taxonomic research in the Antarctic Region has significantly intensified resulting in the description of a large number of new and most likely endemic taxa from a wide variety of genera such as Muelleria (Frenguelli) Frenguelli (Spauld-ING et al. 1999; VAN DE VIJVER et al. 2010, 2014), Luticola D.G.Mann (Kopalová et al. 2011; Zidarova et al. 2014), Navicula Bory (VAN DE VIJVER et al. 2011), Pinnularia Ehrenb. (Zidarova et al. 2012) and Stauroneis Ehrenb. (Zidarova et al. 2014). Compared to this high number of endemic Antarctic taxa, the number of new Bacillariales taxa for the Antarctic Region is very low and restricted to some new taxa in the genera Nitzschia Hassall and Hantzschia Grunow. The first Antarctic freshwater nitzschioid diatom to be described was Fragilaria tenuicollis var. antarctica W. et G.S.West in 1911 (later transferred to Nitzschia westii D.E.Kellogg et al. in 1980 and corrected to Nitszchia westiorum in Van de Vijver et al. 2012). Bourrelly & Manguin (1954) published three new diatoms from the sub-Antarctic Îles Kerguelen including one Denticula Kütz. taxon: Denticula elegans var. robusta Manguin. Almost fifty years later, Van de Vijver et al. (2002)

described *Nitzschia chardezii* Van de Vijver et Beyens and *Hantzschia possessionensis* Van de Vijver et Beyens. The largest contribution to new nitzschioid diatoms in the region so far was Zidarova et al. (2010), who described five new taxa from the genus *Hantzschia* from Livingston Island. Until now however, no new freshwater taxa of *Denticula* have been described from the Maritime Antarctic Region.

Although originally only freshwater taxa were included within the genus Denticula, nowadays, the genus comprises a lot of marine and brackish taxa, several of which were transferred to the fossil genera Crucidenticula (AKIBA & YANAGISAWA 1985) and Neodenticula (AKIBA & YANAGISAWA 1985) and the extant genera Denticulopsis (SIMONSEN 1979), Nagumoea WITKOWSKI et KOCIOLEK (in WITKOWSKI et al. 2011) and Tetralunata (Hamsher, Graeff, Stepanek & Kociolek 2014). Hamsher et al. (2014) reviewed the taxonomic history of the genus Denticula indicating a total number worldwide for this genus to be around 100 taxa based on the list in Fourtanier & Kociolek (2011). The number of freshwater taxa of *Denticula*, however, is limited. Krammer & Lange-Bertalot (1988) list seven, that number now considered too low as the broad species definition they adopted, placed many validly

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described taxa in synonymy. Since then, new taxa have been described, such as *Denticula thermaloides* (VAN DE VIIVER & COCQUYT 2009), but compared to the vast increase in genera such as *Pinnularia*, *Luticola* or *Amphora* EHRENB. (LEVKOV 2009; LANGE—BERTALOT et al. 2011; ZIDAROVA et al. 2012; LEVKOV et al. 2013), the total species number in this genus remains low.

Some of the freshwater taxa, such as *Denticula tenuis* Kütz. and *D. kuetzingii* Grunow, are widespread in the Arctic Region (Greenland, Spitsbergen, Ellesmere Island) forming often large populations in lakes and wet moss vegetation (Douglas & Smol 1993; Van De Vijver et al. 1999, 2003; Antoniades et al. 2008). On the contrary, in the Antarctic Region, *Denticula* is almost completely absent and so far only reported in very low abundances from some sub—Antarctic islands in the southern Indian Ocean (Van de Vijver et al. 2002). Kellogg & Kellogg (2002) list only one species present in a handful of papers published on Antarctic localities.

During a survey of the lacustrine diatom flora of James Ross Island, located in the northern Weddell Sea, several very large *Denticula* populations were discovered. In some investigated lakes, more than 90% of all counted valves belonged to this taxon. Using the currently available literature, it was however impossible to identify these populations and after careful comparison with similar taxa, these populations are described here as *Denticula jamesrossensis* sp. nov. and its morphological variability is documented using light– (LM) and scanning electron (SEM) microscope images.

MATERIAL AND METHODS

During a survey of non-marine diatoms from James Ross Island, a series of samples collected on Clearwater Mesa, have been investigated. Clearwater Mesa is a volcanic mesa (= tableland) composed of single hyaloclastite breccia unit capped with subaerial basalt of the James Ross Island Volcanic Group (Nelson 1975; Košler et al. 2009). The volcanic rocks of the Clearwater Mesa delta have not been dated directly, but the age of the adjoining Förster Cliffs main delta (2.50±0.07 Ma; Smellie et al. 2008) shows on the earliest Pleistocene age of this subglacial volcanic phase and could be correlated with the Clearwater Mesa volcanic delta (SMELLIE et al. 1996). On the flat mesa surface (mostly 250-300 m a.s.l.) tens of lakes evolved after the glacier retreat during the early Holocene (Nedbalová et al. 2013). These lakes are generally shallow with a maximum depth of a few meters and could be regarded as permanent and old with a persistence of thousands of years. Their littoral zone is covered by microbial mats dominated by filamentous cyanobacteria.

Diatom samples for LM observation were prepared following the method described in VAN DER WERFF (1955). Small parts of the samples were cleaned by adding $37\% \, H_2O_2$ and heating to $80\,^{\circ}\text{C}$ for about 1 h. The reaction was completed by addition of KMnO₄. Following digestion and centrifugation (three times 10 minutes at $3700\times \, \text{g}$), cleaned material

was diluted with distilled water to avoid excessive concentrations of diatom valves on the slides. Cleaned diatom material was mounted in Naphrax®. The slides were analysed using an Olympus BX53 microscope, equipped with Differential Interference Contrast (Nomarski) and the Colorview I Soft Imaging System. Samples and slides are stored at the BR-collection, property of the Belgian federal government and given in permanent loan to the Botanic Garden Meise (Belgium). For SEM, parts of the oxidized suspensions were filtered through polycarbonate membrane filters with a pore diameter of 1 µm, pieces of which were fixed on aluminium stubs after air-drying. The stubs were sputter-coated with a Gold-Palladium layer of 20 nm and studied in a ZEISS ULTRA SEM microscope at 3 kV (Natural History Museum (BM), London, UK). Morphological terminology follows ROUND et al. (1990). The morphology of the new species has been compared to the ultrastructure of several described taxa (SOVEREIGN 1963; ARCHIBALD 1982; KRAMMER & LANGE-BER-TALOT 1988; LANGE-BERTALOT & KRAMMER 1993; JOHANSEN et al. 1994; Hamsher et al. 2014).

RESULTS

Division Bacillariophyta

Class Bacillariophyceae Haeckel emend. Medlin & Kaczmarska 2004

Subclass Bacillariophycidae D.G.Mann in Round et al. 1990

Order Bacillariales Hendey 1937 Family Bacillariaceae Ehrenb. 1831 Genus *Denticula* Kütz. 1844

Denticula jamesrossensis sp. nov. (Figs 1-30)

Light microscopy (Figs 1-22): Frustules rectangular in girdle view (Figs 1, 2). Costae weakly or not capitate in girdle view (Figs 1, 2). Valves strictly linear (= never linear to linear-lanceolate) with parallel margins and non-protracted, bluntly rounded to wedge-shaped apices. Most valves with a clear constriction near the valve middle. Valve dimensions (n = 20): length 13.0-19.5 µm, width 2.8-3.3 µm, length/width ratio 4.0–7.4. Raphe near the margin, not discernible in LM. Costae, 9-10 in 10 µm, running parallel to each other throughout the entire valve, slightly more radiate near the apices. Striae not visible in LM. SEM (Figs 23–30): Girdle composed of open copulae bearing one row of small, rounded poroids (Fig. 30). Valvocopula showing an undulating pars interior with undulations covering the onset near the margins of the transapical costae (Fig. 30). Edge of valve mantle covered by a continuous series of small siliceous wraths giving the edge a dentate appearance, well visible on internal views (Fig. 24). Striae, ca. 60 in 10 µm, grouped externally in series of 3-6 rows of areolae, alternating with parallel hyaline zones formed by neighbouring costae (Fig. 23). Striae composed of a series of small, rounded areolae, the outer striae in each group showing the largest areolae (Fig. 23). Near the axial area bearing the raphe,

a continuous longitudinal series of enlarged, rounded areolae present, running irregularly from apex to apex bordering that way the axial area (Figs 23, 25). Striae interrupted near the axial area bearing the raphe and continuing on the mantle as small porefields with up to 6–8 rounded areolae arranged in small descending triangles (Fig. 25). On the other valve margin, continuous longitudinal series of areolae lacking, replaced by a very narrow hyaline zone. Striae continuing on the valve mantle as small porefields (Fig. 23). Costae externally usually not or only slightly narrowing near the margin opposite the raphe. Internally costae clearly developed, thickened near both margins, always more on the raphe-bearing margin than on the opposite site (Figs 27, 29). Coastal thickenings bridging over the raphe canal (Fig. 29) on the raphe-bearing margin. Occasionally, costal thickenings on the raphe-bearing margin joined by shallow thickenings (Fig. 29). Height of the costae lower in the middle than near the margins (Fig. 27). Raphe always in an eccentric position, located on the edge of the valve face (Fig. 23). Raphe branches straight with straight, almost not expanded proximal raphe endings, separated ca. 0.1 µm from each other (Fig. 25). Distal raphe fissures clearly deflected, expanded near the ends (Fig. 26). Internally, raphe running between two very weakly raised ridges (Figs 28, 29). Proximal endings separated by small central nodule (Fig. 29). Distal raphe endings terminating on small helictoglossae (Fig. 28).

Etymology: The species is named after James Ross Island where the new species was described from. Up to now, the new species has only been found on this island

Type locality: Antarctica, James Ross Island, Clearwater Mesa, sample D62 (64°01'58"S, 57°43'41"W), leg. L. Nedbalová, coll. date 09/02/2009.

Holotype (designated here): BR–4346 (Botanic Garden Meise, Belgium).

Isotypes (designated here): PLP–240 (University of Antwerp, Belgium).

Ecology and Distribution: *Denticula jamesrossensis* has been found only on James Ross Island. The largest population was observed on the Clearwater Mesa. Its distribution on other Maritime or Continental Antarctic

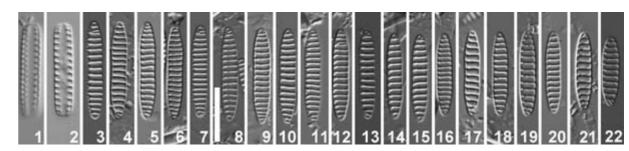
localities is at present unknown. The largest population of *Denticula jamesrossensis* was found in two samples (epilithic and epipelic) from the littoral zone of a stable shallow lake (see Nedbalová et al. 2013 for more details on lake types on James Ross Island), altitude 250 m a.s.l., max. depth 0.5 m, with an alkaline pH of 8.6, a very high specific conductance value of 4000 μS.cm⁻¹ and low nutrient values (Total Phosphorus = 17.9 μg.l⁻¹). The samples were dominated by *Denticula jamesrossensis* reaching abundance values to almost 90%. Other taxa in the sample include *Craticula antarctica* Van de Vijver et Sabbe, *Pinnularia australomicrostauron* Zidarova et al. and *Halamphora oligotraphenta* (Lange–Bert.) Levkov.

DISCUSSION

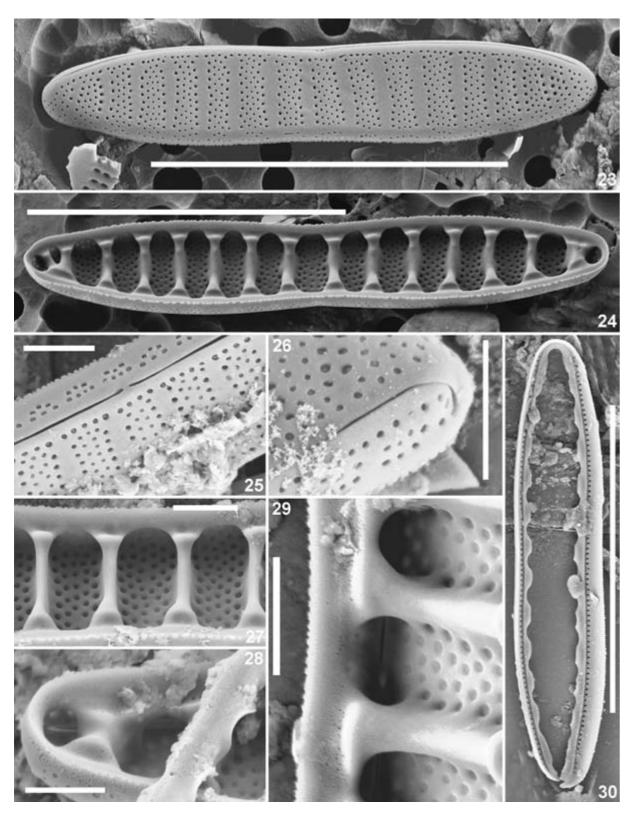
There are only a few small-celled, finely striated freshwater or brackish Denticula taxa known worldwide. A total of 5 species, including D. jamesrossensis, can all be included in a subgroup within the genus *Denticula*, as recognized in 1993 by Lange-Bertalot & Krammer who suggested this subgroup is a 'taxonomic bridge' between the families Bacillariaceae and Epithemiaceae. All species in this subgroup show an eccentric raphe, a feature usually found in the genus Nitzschia, and have distinct costae and an adaptation of the valvocopula relative to the costae, a feature typical for the genus Denticula. Whether they should be separated from Denticula and Nitzschia as a separate genus is at present not clear and thorough molecular or cladistic research will be necessary to reveal the correct taxonomic position of these taxa.

The group of species related to *D. tenuis*, another smaller freshwater *Denticula* species, can be separated by their different position of the raphe canal, being situated on a distinct raphe keel on the valve face instead of being entirely eccentric and the presence of typical capitate costae in girdle view.

Although all taxa from this subgroup show some similarity with *D. jamesrossensis*, they can be separated based on a combination of morphological differences. The most similar taxon is *D. rainierensis*, described in 1963 by Sovereign from Ohanapecosh



Figs 1–22. LM: Denticula jamesrossensis, all images from holotype slide (BR–4346), (1–2) Entire frustules in girdle view, (3–22) Valve face views showing variation in size and valve outline. LM scale bar 10 μm.



Figs 23–30. SEM: *Denticula jamesrossensis*, all pictures taken from holotype material, (23) External view of entire valve showing raphe and stria structure, (24) Internal view of entire valve showing arrangement of costae and striae. Note silica dentation on mantle edge, (25) External detail of central area with straight proximal raphe endings, (26) External detail of valve apex with deflected distal raphe fissure, (27) Internal view showing shape of costae and areolae, (28) SEM internal detail of valve apex with helictoglossa, (29) Internal detail of central area with central nodule and proximal raphe endings, (30) Entire valvocopula showing undulated pars interior and single row of small pores. SEM scale bar 1 μm (25–29), 10 μm (23, 24, 30).

Hot Springs, USA. Initially, Krammer & Lange-Ber-TALOT (1988) considered it to be a synonym of D. subtilis but after a detailed study of its ultrastructure by JOHANSEN et al. (1994), both taxa were separated again. Denticula jamesrossensis can be distinguished from D. rainierensis in having slight larger valves (8–16.5 vs. 13-19 µm), a larger length/width ratio giving the valves a more slender, elongate outlook, a strictly linear valve outline compared to the more linear-lanceolate to elliptic-lanceolate valve outline of D. rainierensis, the presence of a clear constriction in the valve middle of D. jamesrossensis and a higher number of striae (ca. 60 vs. 45-52 in 10 µm) (see also Hamsher et al. 2014). Denticula rainierensis shows a comparable morphology to D. creticola (ØSTRUP) LANGE-BERTALOT et Krammer. Lange-Bertalot & Krammer (1993) suggested a possible conspecificity between these two species but refrained from taking a clear decision. Based on the LM images taken from the holotype of D. creticola presented in their paper, it is clear that the latter shows some similarities with D. jamesrossensis but can be separated based on the following features: the valves of D. creticola have a more elliptic-lanceolate valve outline with more rounded apices, always lacking a constriction in the valve middle; the valves are usually smaller (< 15 µm). ØSTRUP (1910) mentions only 10 striae in 10 µm but most likely this refers to the number of costae. Based on figure 36 in Lange-Berta-LOT & KRAMMER (1993) from a valve found in Ukraine, the striae are grouped with a maximum of 6 striae per group, similar to D. jamesrossensis. Based on the same figure, the proximal raphe endings are apparently also much less separated than in D. jamesrossensis. Two other small-celled freshwater Denticula taxa are D. subtilis and D. sundaysensis Archibald. Both taxa have a clearly lanceolate to linear-lanceolate valve outline with a lower number of striae (26-30 in 10 µm) which are discernible in LM and is almost half the number of striae in D. jamesrossensis. Denticula sundaysensis has typically only two rows of areolae between the costae contrary to the 3-6 rows in *D. jamesrossensis*. On the sub-Antarctic Ile de la Possession (Crozet Archipelago) in the southern Indian Ocean, VAN DE VIJVER et al. (2002) report the presence of a small population of D. sundaysensis but the population is not conspecific with D. jamesrossensis being much smaller with only two rows of areolae between the costae.

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Additionally, ecological differences can be noted between the different species mentioned above. *Denticula rainierensis* was originally described from a hot spring in the state of Washington (western USA). The species was also found, as was *D. creticola*, on wet walls (Johansen et al. 1994) although Lange-Bertalot & Krammer (1993) also reported their presence in brackish waters. *Denticula jamesrossensis* was typically found in alkaline lakes with very high specific conductance values and higher chloride values (up to 181 mg.l⁻¹). The two other taxa, *D. subtilis* and *D. sun-*

daysensis are typical brackish taxa.

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