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Orthoseira gremmenii sp. nov., a new aerophilic diatom from Gough Island (southern Atlantic Ocean)

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Abstract – A new *Orthoseira* species, *Orthoseira gremmenii* sp. nov., is described from the small volcanic Gough Island, located in the southern Atlantic Ocean. Using light and scanning electron microscopy, the discriminating features of the species are revealed. *Orthoseira gremmenii* is characterized by its very large valve dimensions (total valve diameter up to 100 µm), the presence of a large hyaline central area (more than 50% of the total valve diameter), the occurrence of regularly placed bifurcated linking spines and the absence of typical features such as undulations in the valve interior (so-called caverns) and marginal porefields, commonly reported in other species.

A very small proportion of the population possesses typical internal valves, covering the entire valve interior, leaving only a few circular, window-like openings close to the valve face/mantle junction. The presence of these internal valves remains so far without explanation but may be related to the extreme environmental (i.e. aerophilic) circumstances in which the species is found.

Bacillariophyceae / *Orthoseira* / aerophilic diatoms / internal valves / Gough Island / southern Atlantic Ocean

Résumé – ***Orthoseira gremmenii* sp. nov., nouvelle diatomée de Gough Island (océan atlantique austral).** Une nouvelle espèce *d'Orthoseira, Orthoseira gremmenii* sp. nov., est décrite d'une petite île volcanique, Gough Island, située dans l'Océan atlantique austral. Ses caractéristiques discriminantes sont précisées à l'aide d'analyses microscopiques optique et à balayage. *Orthoseira gremmenii* est caractérisé par ses dimensions valvaires très grandes (diamètre total de la valve jusqu'à 100 µm), par la présence d'une aire centrale hyaline très grande (dépassant 50 % du diamètre total de la valve), par la présence des épines de jonction bifurquées, placées très régulièrement autour de la valve et par l'absence d'autres caractéristiques typiques pour certaines espèces du genre comme la présence d'ondulations dans la partie interne (= « cavernes ») et celle de champs de pores marginaux.

Une petite proportion de la population possède des valves internes typiques, couvrant entièrement la partie interne de la valve avec uniquement quelques ouvertures sous forme de fenêtres au niveau de la jonction entre la partie valvaire et le manteau. La présence de

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ces valves internes reste à présent sans explication satisfaisante mais ces valves internes servent probablement comme protection contre les conditions de vie extrêmes (i.e. aérophiles) dans lesquelles l'espèce a été trouvée.

Bacillariophyceae / *Orthoseira* / diatomées aérophiles / valves internes / Gough Island / Océan atlantique austral

INTRODUCTION

The genus *Orthoseira* Thwaites is one of the most typical aerophilic diatom genera that, together with *Luticola* Mann, *Diadesmis* Kützing and *Hantzschia* Grunow, belongs to the most reported diatom genera in terrestrial, wet to semi-wet habitats. Although the nomenclatural status of *Orthoseira* is still not entirely clear (Houk, 1993; Spaulding & Kociolek, 1998) due to the uncertainties surrounding its type species, *Melosira americana* Kützing, several new species have been described during the past few years, such as *Orthoseira biportulata* Van de Vijver & Beyens (from Ile de la Possession, sub-Antarctica: Van de Vijver *et al.*, 2002) and *O. ursula* Metzeltin & Lange-Bertalot (from Costa Rica: Metzeltin & Lange-Bertalot, 2007). Houk (1993) and Spaulding & Kociolek (1998) have discussed in detail the nomenclatural and taxonomic problems within the genus *Orthoseira*.

Following the concept of *Orthoseira* as proposed by Round *et al.* (1990) based on a set of typical features, it is possible, however, to distinguish and describe new forms (*contra* Spaulding & Kociolek, 1998).

All species in the genus *Orthoseira* described so far bear the same set of typical features, the principal character being the presence of the "carinoportulae" at the valve centre (Crawford, 1981). These are tube-like passages through the valve characterized by simple internal openings and slightly raised collars to the outside (Round *et al.*, 1990). Although the number of carinoportulae varies considerably within the same species (for instance, the two forms discussed in Spaulding & Kociolek [1998] have 2-4 or 4-5 carinoportulae respectively), their presence is a constant feature not shared with any other centric diatom. Further details on the morphology and taxonomic status of *Orthoseira* are given in Crawford (1981), Round *et al.* (1990), Houk (1993) and Spaulding & Kociolek (1998).

The present paper provides a description of an easily distinguishable new species of *Orthoseira* found during a recent survey of the terrestrial (including moss-inhabiting) diatoms from Gough Island in the southern Atlantic Ocean. Several specimens were observed with "internal valves" (Houk, 1993). Using light and scanning electron microscopy, the features of this new species are illustrated to facilitate comparison with previously described ones.

STUDY SITE

Gough Island (40°21' S, 9°53' W) is a small, uninhabited island, situated in the cool-temperate zone of the southern Atlantic Ocean. Geopolitically, it belongs to the United Kingdom Overseas Territory of Tristan da Cunha although

it is located at approximately 300 km from the other islands in this group (Tristan da Cunha, Inaccessible Island & Nightingale Island). According to Stonehouse (1982), it is part of the temperate sub-Antarctic zone, formed with Amsterdam Island, Tristan da Cunha and the Falkland Islands. The island is located approximately midway between the southern tip of Africa and South America, at a distance of ca. 3000 km from the main large continents. The island measures *ca* 6 km by 14 km resulting in a total surface of 65 km^2. Large parts of the island are quite mountainous with most of the island above 400 m. The highest peak (Edinburgh Peak) reaches 910 m a.s.l. The coastline consists of sea cliffs rising up to 300 m to 450 m in height with narrow boulder beaches lacking any sheltered harbour. The southern end of Gough is the only area below 300 m.

The island is typically oceanic having a volcanic origin. It has never been part of a continental landmass. Today, there are no signs of recent volcanic activity. The climate is cool and wet, with a mean annual temperature (at sea level) of 11°C, a mean annual precipitation in excess of 3000 mm and with frequent gale-force winds. Four main types of vegetation can be found ranging from coastal tussock to fern bushes and peat bogs (Wace, 1961; Cooper & Ryan, 1994). The island has no permanent settlements except for a weather station under South African lease.

Despite its very interesting geographical position, its oceanic origin and the number of interesting habitats, the non-marine diatom flora of Gough Island is poorly known. The only paper dealing with diatoms from this region is the one by Carter (1966), which reported on the diatom flora from the Tristan da Cunha Island group. Carter analysed 12 samples, of which only two were collected from Gough Island (Cave Bell Rocks and Stream Gonydale). A very diverse and highly specific diatom flora was found with many new species that – so far – have been seldom found elsewhere. Carter reported the presence of six species of *Melosira* (three described as new). One species, *Melosira roeseana* Rabenhorst, is now in *Orthoseira*, as *Orthoseira roeseana* (Rabenhorst) O'Meara (Round *et al.*, 1990). *Orthoseira roeseana* was not uncommon in the samples from Gough Island. Careful observations of specimens on Carter's slides (BM 77595, 77599, 77600) confirmed that the specimens are, indeed, *Orthoseira roeseana*.

MATERIAL AND METHODS

Some 75 samples from Gough Island were investigated and *Orthoseira gremmenii* was found in only one (Gough2000-297, collection date 17/06/2000, coast opposite the Admirals 40° 20' 45'' S/9° 52' 41'' W, 25 m a.s.l.: bryophyte mats in a seepage area situated on a vertical rock face).

Diatom samples were prepared following the method of Van der Werff (1955). Small portions of the samples were cleaned by adding 37% H_2O_2 and heating to 80°C for about 1h. The reaction was completed by addition of $KMnO_4$. Following digestion and centrifugation, the cleaned material was diluted with distilled water to avoid excessive concentrations of diatom valves that may hinder reliable observations. Cleaned diatom valves were mounted in Naphrax®.

Light microscope observations were conducted using an Olympus BX51 microscope equipped with Nomarski optics. For scanning electron microscopy (SEM), part of the suspension was filtered through polycarbonate membrane

filters with a pore diameter of 3 µm, pieces of which were fixed on aluminium stubs after air-drying. The stubs were sputter-coated with 50 nm of gold and studied in a JEOL-5800LV at 20 kV.

Comparisons are based mainly on information in Crawford (1981), Krammer & Lange-Bertalot (1991), Houk (1993), Spaulding & Kociolek (1998) and Van de Vijver *et al.* (2002). Terminology follows Round *et al.* (1990).

Samples and slides are stored at the National Botanic Garden of Belgium (BR), Department of Bryophytes and Thallophytes.

RESULTS

Orthoseira gremmenii sp. nov.

Valvae cylindricae catenas longas formantes. Frustula aspectu cinguli 40-62 µm longa. Diameter discorum 29-100 µm. Limbus valvarum circiter 15 µm altus. Area centralis formans aream hyalinam magnam (36-79% diametri toti valvae). Spinae marginales bifurcatae ordinate positae in iunctura faciei limbique. Striae radiantes distincte areolatae, 14-17 in 10 µm, ab marginibus usque ad tertium radii (nec longiores usque ad carinoportulas. Carinoportulae 2-4, pro parte maxima 3. Satis differt a speciebus aliis generis Orthoseirae sensu Round et al. *1990.*

Holotypus: BR-4116 (National Botanical Garden, Meise).

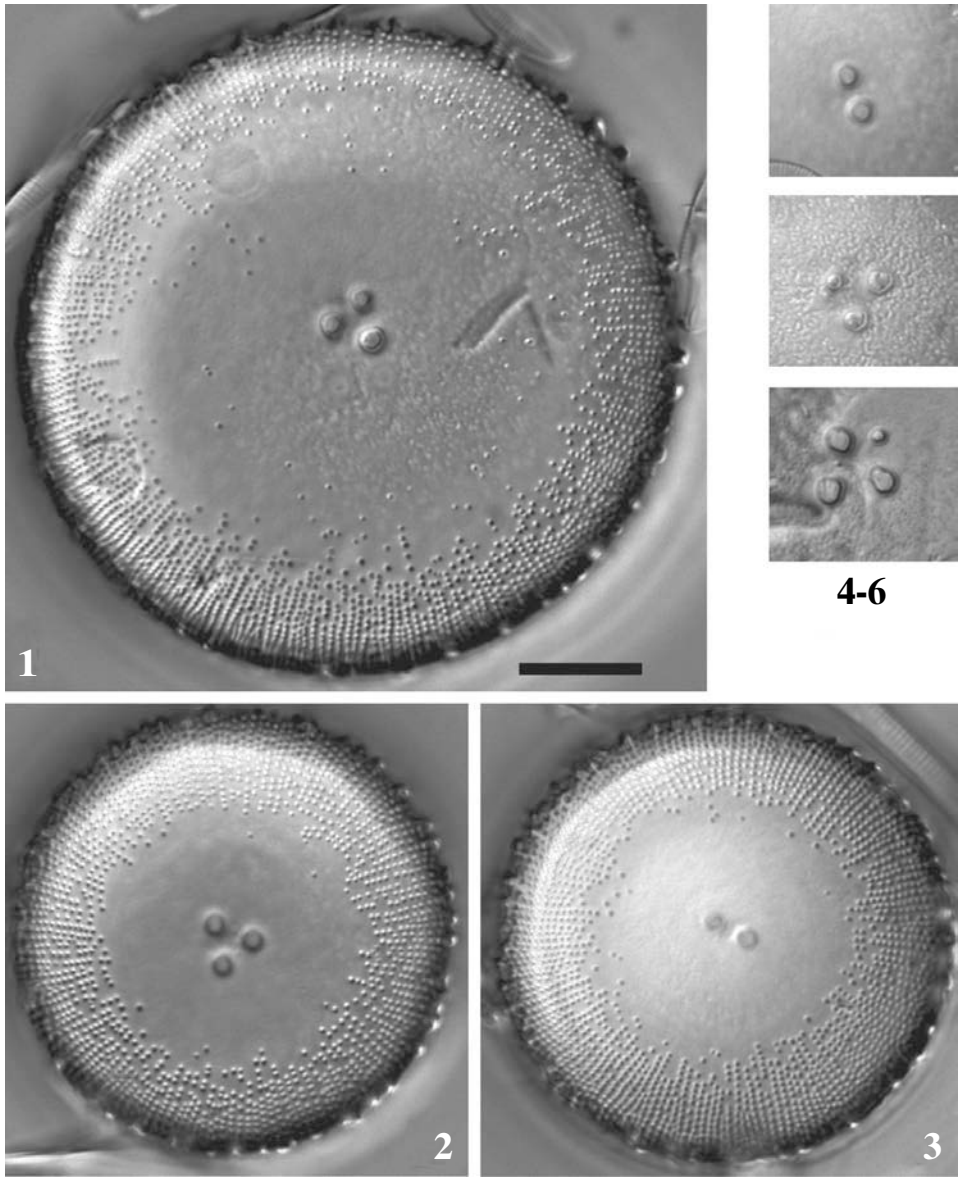
Isotypi: PLP-104 (Universiteit Antwerpen), BRM Zu 6/50 (Friedrich Hustedt Collection, Bremerhaven).

Type locality: Gough Island (southern Atlantic Ocean), the Admirals, Gough2000-297 (coll. date 17/06/00)

Etymology: The specific epithet *gremmenii* refers to our colleague, Dr. Niek Gremmen, who not only collected the sample but whose long-lasting friendship and collaboration is very highly appreciated.

Description (light microscopy: Figs 2-10; 24-26)

Cells cylindrical in girdle view (Figs 8-10), attached with linking spines (Fig. 9), occasionally forming large chains, up to 10 valves. Cells 40-62 µm, mantle height ca. 15 µm. Valves disc-shaped (Figs 1-3), diameter 29-100 µm (mean = 54 ± 16 µm, median = 50 µm, see Fig. 7A for the size frequency/valve diameter; most valves between 35 and 60 µm). Valve surface flat, forming a right angle with valve mantle. Striae composed of uniseriate areolae, 14-17 in 10 µm; valve face areolae small, radiate, occupying a small portion of the total valve face surface, remaining surface a large, central hyaline area of diameter 36-79% of total diameter (mean% = 59 ± 8%, median % = 59%; see Fig. 7B for the relationship between total valve diameter and diameter of hyaline area, larger valves with larger hyaline area. Towards the valve centre, areolae become more scattered, with the organized pattern becoming lost, 16 and 20 areolae in 10 µm, irrespective of valve size. Central area with 2-4 carinoportulae (for 100 valves, most (80%) with 3; only two valves with 4; 18% with 2). No relationship noted between number of carinoportulae and valve diameter; smaller valves tend towards 2. Distinct ring of marginal linking spines at the valve face/mantle junction, with regular pattern detected for position. No caverns or internal



Figs 1-6. *Orthoseira gremmenii* sp. nov. Light Microscopical views. **1-3.** Valve views showing different numbers of carinoportulae. Note the presence of the large hyaline central area. **4-6.** Three possible combinations of carinoportulae. Scale bar = 10 µm.

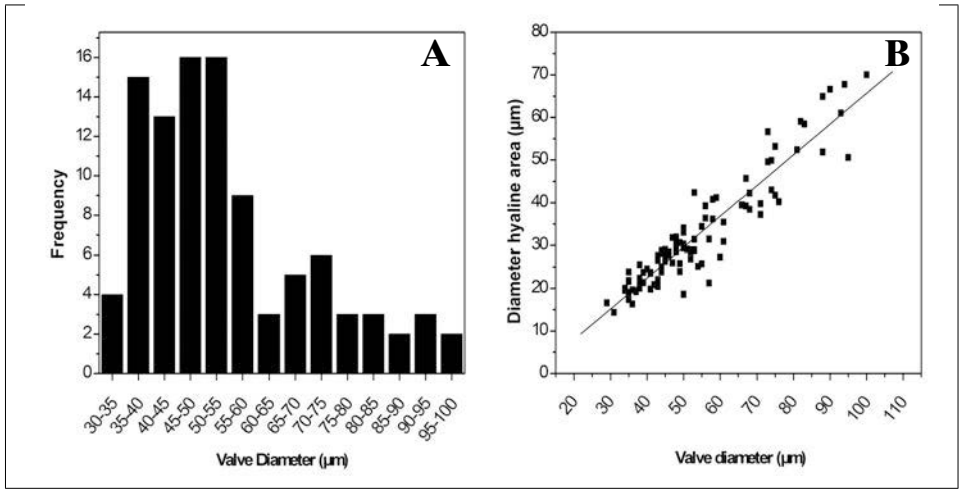
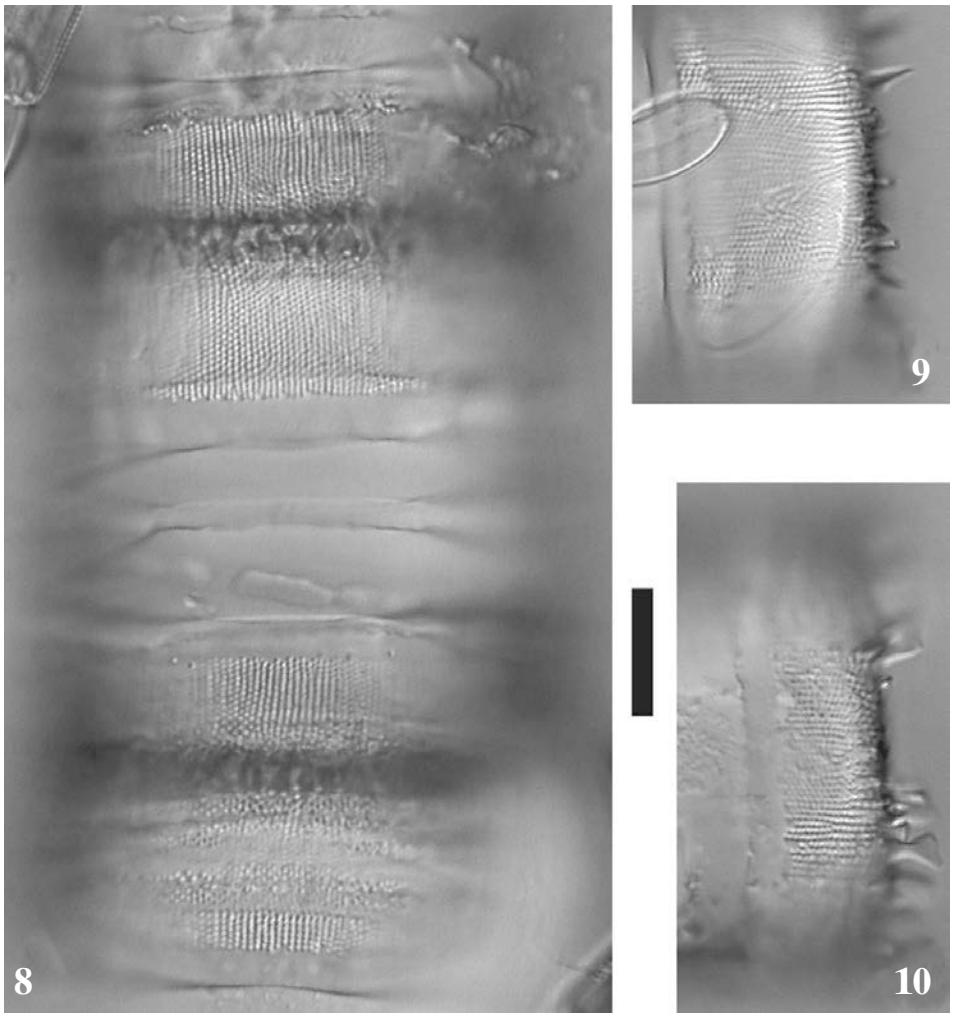


Fig. 7. Morphometric data for *Orthoseira gremmenii* sp. nov. **A**. Frequency in number of valves versus valve diameter (5 µm size classes). **B**. Diameter of the hyaline area versus total valve diameter. The plotted line represents the linear trend line (hyaline area diameter = 0.72 × total valve diameter – 6.39; n = 100; $R^2 = 0.87$; $p < 0.001$).

undulations present. Several valves produce 'internal valves'; plate-like structure inside valve covering almost entire valve diameter. In girdle view, 'internal valves' form regular pattern of open "windows", on valve mantle. Copulae number variable, between 5-7, maximum of 10 (Fig. 8).

Description (scanning electron microscopy: Figs 11-23, 27-29)

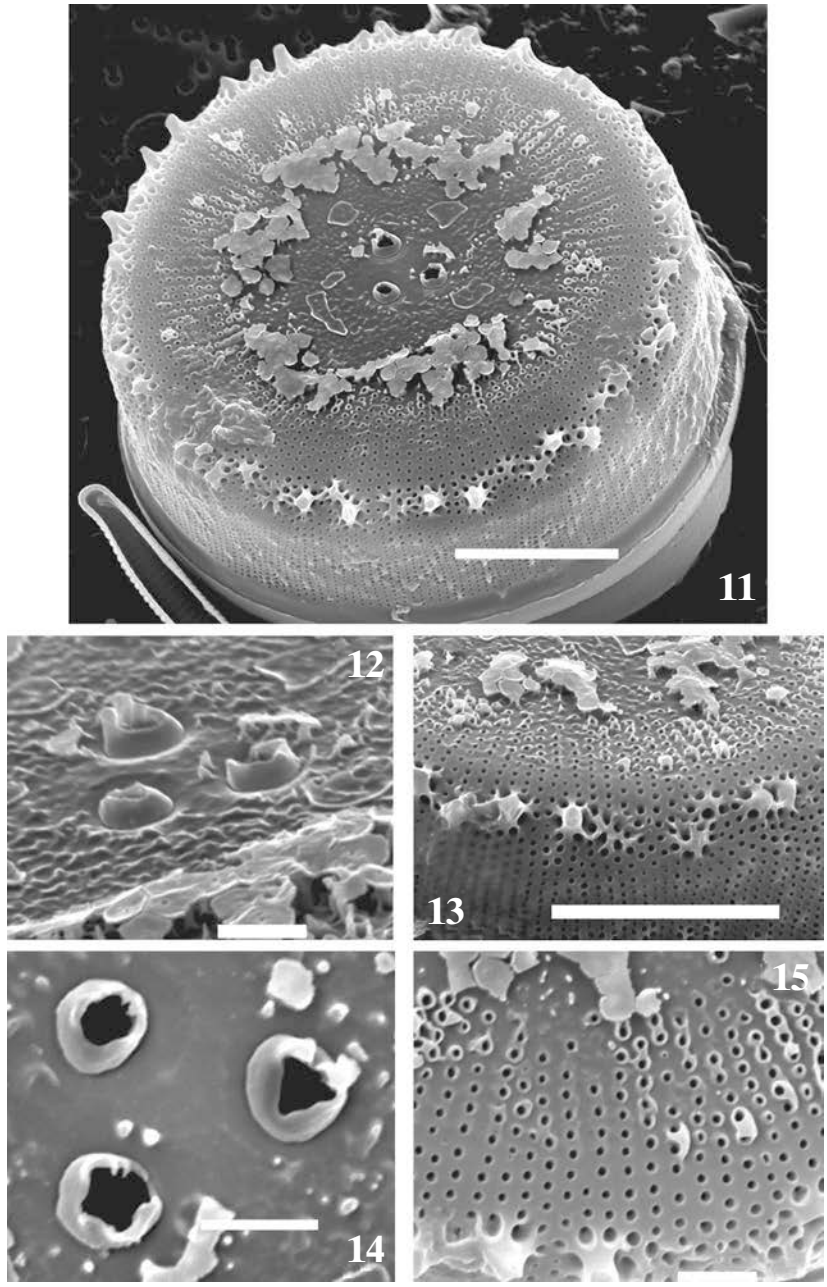
Valve face flat, often covered by a siliceous layer obscuring underlying structures (removal reveals external structure, Fig. 11). Hyaline central area smooth but with small siliceous thickenings; small spines scattered on valve face; areolae radiate and poroid (Figs 13, 15); some possessing slightly raised rim (Figs 13, 15); spinules not present. Areolae with rim occur near hyaline area; areolae near face/mantle junction usually simple (Fig. 15). No difference in areolae structure between mantle and valve face; areolae near spines increase in size (Fig. 13); no external marginal pore fields between groups of spines (Fig. 13). Valve face/mantle junction abrupt, with smoothly rounded edge (Figs 11, 13), with ring of linking spines (Figs 11, 13, 15, 22-23), regularly scattered along entire valve edge (Fig. 11), appearing as stellate structures or (more rarely) simple siliceous thickenings; often broken in unlinked valves (Fig. 13). Linked spines appear plate-like, bifurcated or with irregular shape (Figs 22-23). Carinoportulae are well distinguishable in the central area, each possessing pronounced (almost conical) silica collar (Figs 12, 14). Copulae open, with poroids scattered or organized into rows parallel to the perivalvar axis (Figs 21, 23); some copulae unornamented. Outer mantle areolae covered by siliceous thickenings (Fig. 23). Internally, valve face flat and very smooth (Fig. 16). Areolae as small rounded poroids (Figs 16, 19), internal opening with or without velum (Figs 16, 19). Longitudinal rows of areolae sometimes dichotomise towards valve mantle, with occasional short series



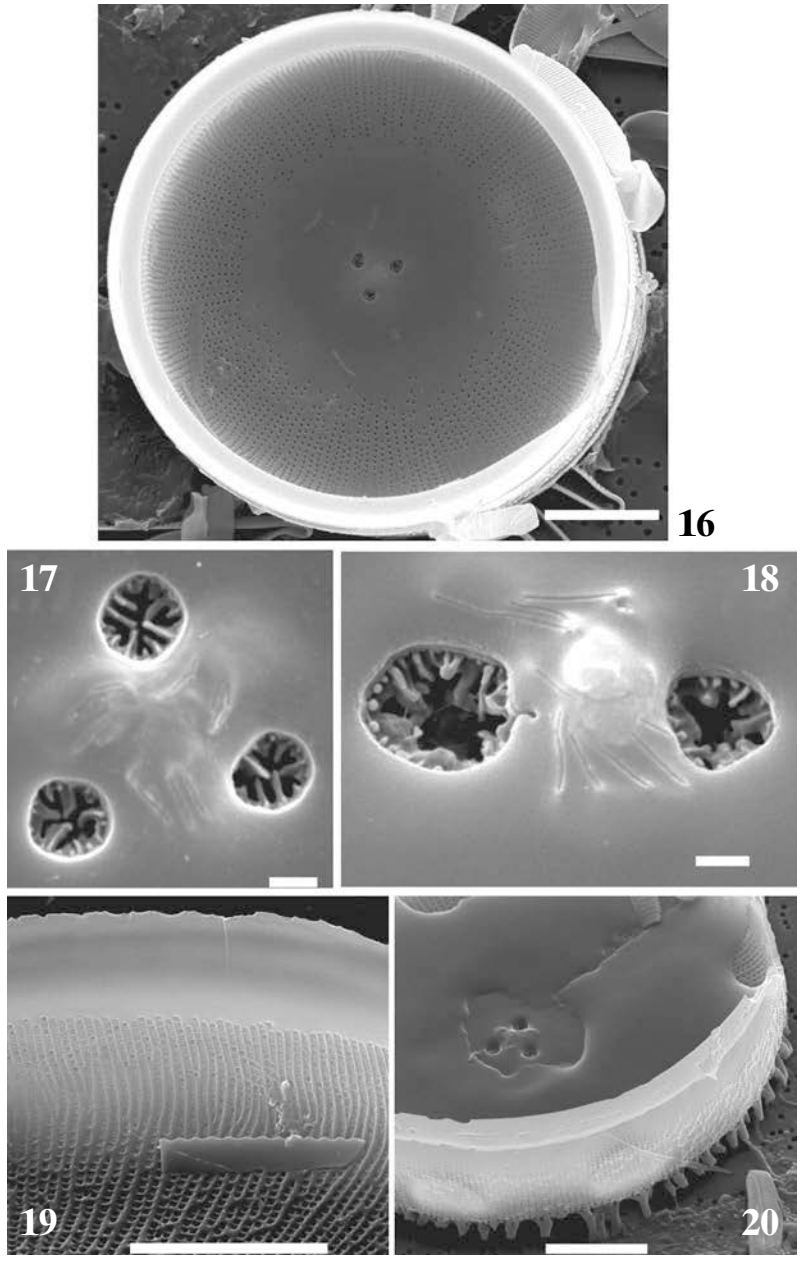
Figs 8-10. *Orthoseira gremmenii* sp. nov. Light Microscopical views. **8.** Entire frustule linked with two other cells in a long chain. **9-10.** Details of the mantle view with focus on the linking spines. Note the bifurcated spines in Fig. 10. Scale bar = 10 µm.

of areolae inserted between usual rows (Fig. 19). Lower part of valve mantle, towards cingulum, without areolae but hyaline (Fig. 19). Carinoportulae unoccluded, with well-defined, simple pits. Inside with several irregular silica outgrowths and small granules, arranged in stellate pattern; several irregular shallow slits at centre (Figs 17, 18), either single (Fig. 18) or paired (Fig. 17).

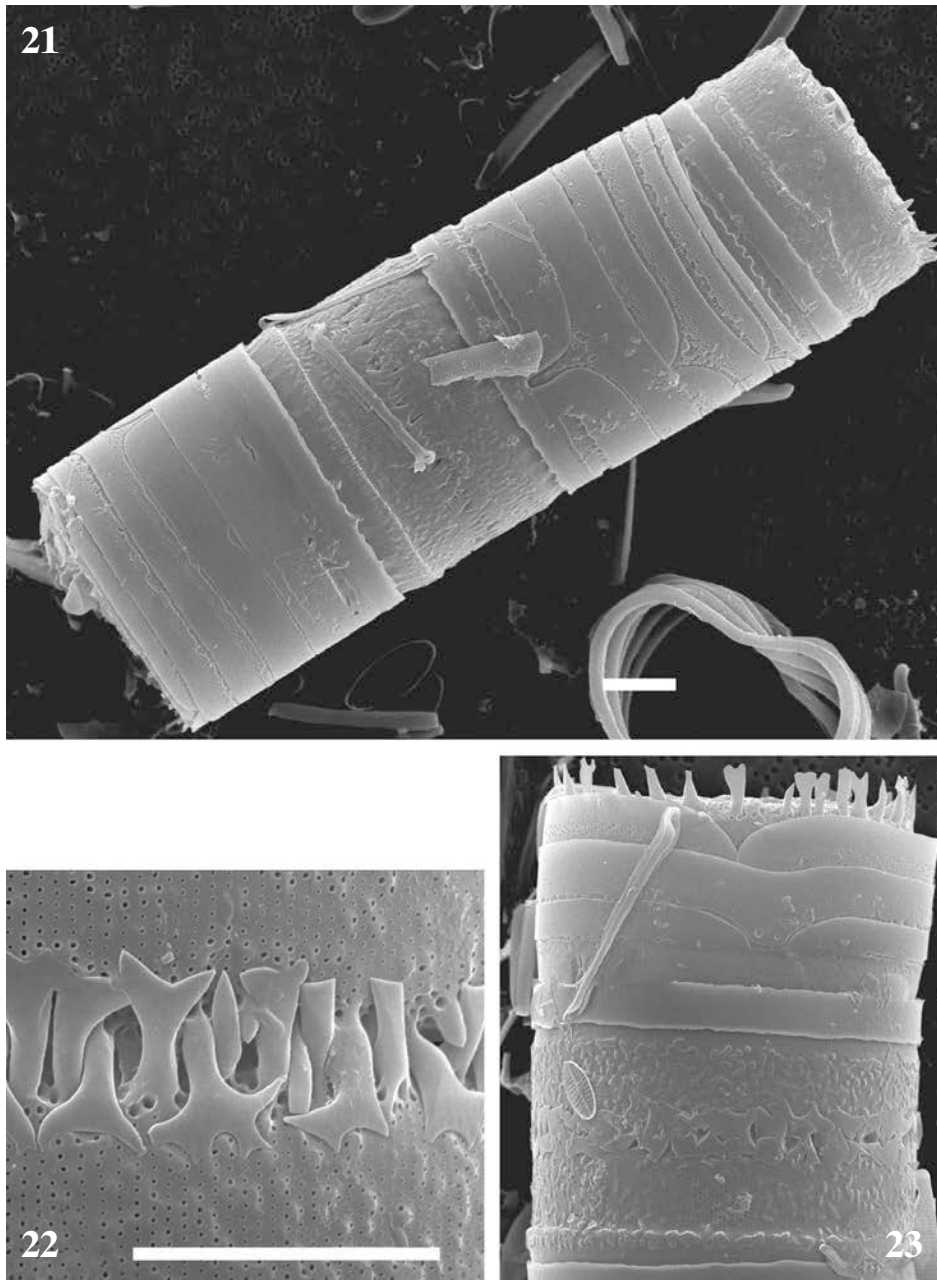
Internal valves: externally, occur as bright coloured areas on valve mantle (Fig. 20); internally, form plate-like, hyaline structure covering (almost) entire inside of valve (Fig. 27), extending towards mantle edge (Fig. 29). Several "window-like" openings present, revealing underlying valve structure, near valve face/mantle junction (Figs 27-29). Carinoportulae not covered by internal valve.



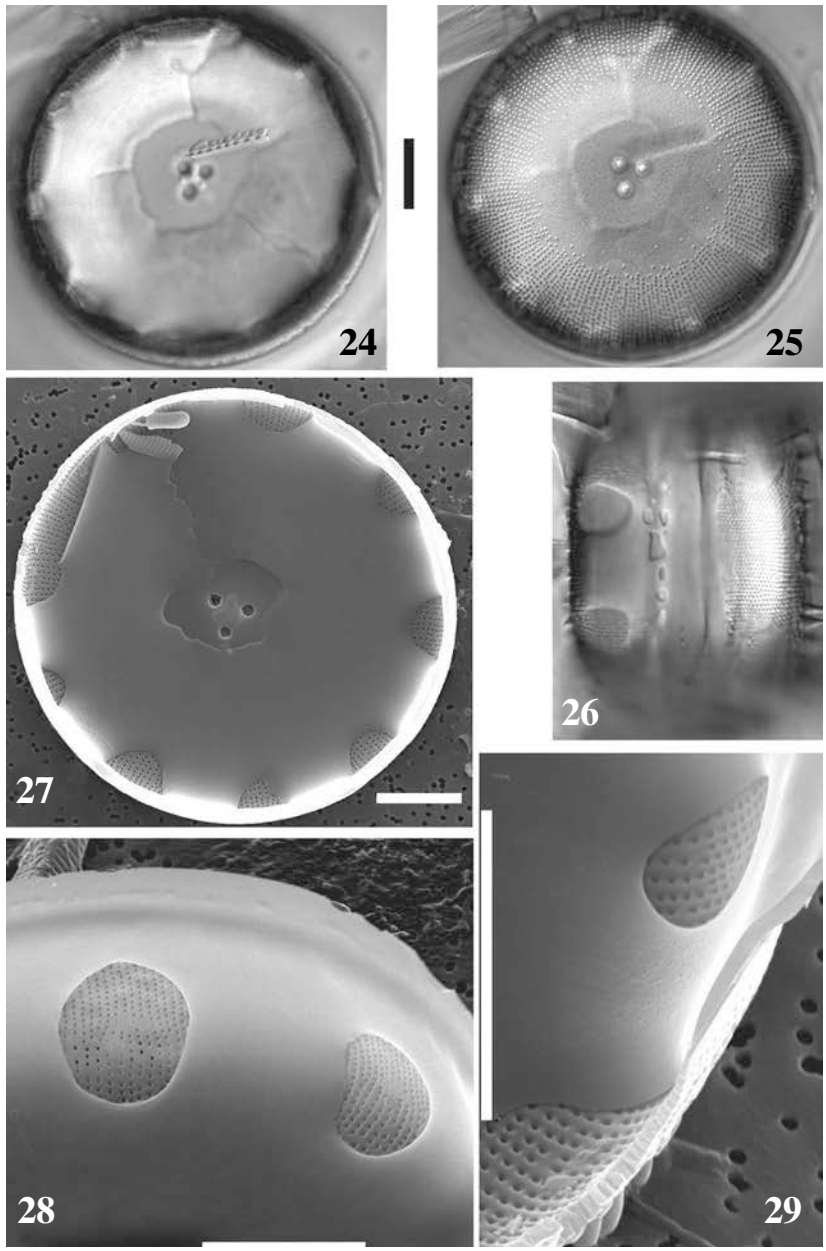
Figs 11-15. *Orthoseira gremmenii* sp. nov. Scanning Electron Micrographs. **11.** External view of an entire valve showing the remains of a silica cover, three carinoportulae, the structure of the hyaline central area and the rows of areolae and the regularly placed linking spines. **12.** Detail of carinoportulae showing the pronounced collar. Angle of tilt = 45°. **13.** Detail of the valve face/mantle junction with special focus on the linking spines. **14.** Detail of carinoportulae. **15.** Detail of the areolae structure. Note the presence of weakly raised rims round the areolae closer to the central area. Scale bar = 10 μm in figs 11 & 13 and 2 μm in figs 12, 14 & 15.



Figs 16-20. *Orthoseira gremmenii* sp. nov. Scanning Electron Micrographs. **16.** Internal view of an entire valve showing three carinoportulae, the areolae structure and the extent of the hyaline area. **17-18.** Detail of the carinoportulae. Note the presence of internal silica structures inside the carinoportulae and the single to paired slits in between the carinoportulae. **19.** Detail of the valve face/mantle junction with focus on the striae structure. Note the presence of intercalated shortened striae near the end of the mantle areolae. **20.** Detail of an internal valve inside the valve interior. The position of the open windows in the internal valve are well visible on the outside. Scale bar = 10 µm in figs 16, 19 & 20 and 1 µm in figs 17 & 18.



Figs 21-23. *Orthoseira gremmenii* sp. nov. Scanning Electron Micrographs. **21.** External view of an entire frustule linked in a short chain to another valve. The number of (perforated) copula is well visible. **22.** Detail of two valves linked together with their spines. Spines are clearly bifurcated or having a very irregular shape. **23.** Detail of the girdle and the valve mantle. Specimen only weakly cleaned showing siliceous coverings on the areolae. Scale bar = 10 µm.



Figs 24-29. *Orthoseira gremmenii* sp. nov. Light and Scanning Electron Micrographs. **24-25.** Light microscopy pictures of the same specimen at different focus levels showing clearly the presence of the internal valve. **26.** Girdle view in LM with the presence of the internal valve visible as small window-like openings on the mantle. **27.** SEM internal view of an internal valve. The carinoportulae are not covered and near the valve face/mantle junction several window-like openings are visible in the internal valve giving a view on the underlying areolae. **28.** SEM detail of the window-like openings in the internal valve. Specimen tilted at an angle of 45°. **29.** SEM detail of a broken internal valve. Scale bar = 10 µm.

Ecology and associated diatom flora

Since *Orthoseira gremmenii* was found in only one sample, its typical ecological preferences are not yet fully known. At the type location, the species was not rare, occurring with an abundance of almost 5%. Specimens were easily observed due to their large size and typical features. The type location is a relatively large, circular depression on the coastline opposite the Admiral Rocks. *Orthoseira gremmenii* was found in dense bryophyte mats in a seepage area on a vertical rock face near the bottom of a ca. 25 m deep vertical lava tunnel. The site is permanently moistened by water seeping down along the rocks in shaded (but not very dark) and strongly sheltered conditions.

The sample is almost exclusively dominated by three species: *Psammothidium investians* (Carter) Bukhtiyarova, *Eunotia* aff. Grunow *paludosa* var. *paludosa* and *Planothidium dubium* (Grunow) Round & Bukhtiyarova, with minor abundances of various *Planothidium* and *Pinnularia* species. The dominant species are known to prefer aerial but wet conditions (Van de Vijver *et al.*, 2002).

DISCUSSION

There is no doubt that the present species belongs in the genus *Orthoseira*, as defined in Round *et al.* (1990). The presence of carinoportulae, the structure of the areolae and the presence of the linking spines facilitating chain formation are features that indicate its assignment to *Orthoseira sensu* Round *et al.* (1990).

The presence of internal valves is not unusual for species of *Orthoseira* (Houk, 1993; Krammer & Lange-Bertalot, 1991 – although their detailed structure is not clear when observed only in LM). Metzeltin & Lange-Bertalot (2007) include several illustrations of *Orthoseira* cf. *dendrophila* (Ehrenberg) Crawford with similar structures. Their appearance may be related to the often extreme circumstances in which *Orthoseira* species live (Houk, personal comment). *Melosira dickiei* (Thwaites) Kützing and *M. robusta* Hustedt also produce similar internal valves. These species live under the same aerophilic conditions. In periods of drought, internal valves may help to protect the valves from desiccation.

The genus *Orthoseira* includes only a few species and therefore it is easy to separate *Orthoseira gremmenii* from all other known species. In the population studied, nearly all features, except valve diameter, were quite constant. The number of carinoportulae is usually three with only a small proportion of specimens having two. Spines are always present and the size of the hyaline central area in relation to the total valve diameter is also quite stable. This low degree of variability contrasts well with other species (Spaulding & Kociolek, 1998). Thus, we have no hesitation in describing these specimens as new, rather than leaving them undescribed as in Spaulding & Kociolek (1998), who encountered a much higher variability.

The stability of its features also facilitates the separation of *Orthoseira gremmenii* from other known or recently described species of *Orthoseira*. One of the most useful features for discriminating this species is the large hyaline central area. Most *Orthoseira* species only have a very small central area since the longitudinal rows of areolae reach almost entirely till the zone with the carinoportulae. In *Orthoseira roeseana*, for instance, the hyaline part of the valve

never exceeds 25% of the total valve diameter, whereas in *Orthoseira biportulata*, a species recently described from Ile de la Possession (Crozet Archipelago) in the southern Indian Ocean (Van de Vijver *et al.*, 2002), the central area has a rather constant size of 40%. Both forms from Madagascar have a rather variable central area but the areolae always extend to more than 50% of the valve radius (Spaulding & Kociolek, 1998). The only species that shows a similar hyaline area is the recently described *Orthoseira ursula*. However, that species is usually smaller, possesses plate-like structures between the striae, has biseriate striae near the valve face/mantle junction and always possesses only two carinoportulae.

Houk (2003) found an unnamed *Melosira* form from the High Tatra Mountains in Slovakia with a somewhat larger central area than encountered in species of *Orthoseira* so far. However, based on the combination of the presence of large ribs near the valve face/mantle junction and the lower valve dimensions, Houk's specimens can easily be separated from *O. gremmenii*.

A second feature that can be used to separate *Orthoseira gremmenii* from other species is its valve dimensions. The valve diameter of most *Orthoseira* species never exceeds 75 µm (e.g. *O. ursula* < 65 µm, *O. roeseana* < 70 µm, *O. biportulata* < 35 µm, *O. (Melosira) dendrophila* < 65 µm). As can be seen from Fig. 7A, almost 25% of the measured valves had a total valve diameter of more than 65 µm, making *O. gremmenii* the largest *Orthoseira* species that has been described so far.

Both *Orthoseira* forms from Madagascar possess undulations of the valve interior called 'caverns' (Spaulding & Kociolek, 1998). These structures were not observed in *Orthoseira gremmenii*, whose valves always had a flat valve interior. Since the presence of these caverns was evident in LM in both valve and girdle view, it is clear that they are a useful discriminating feature.

The combination of the rather regular position of the linking spines, the bifurcated shape of the spines themselves and the absence of marginal pore fields is another character typical for *O. gremmenii* and not observed in any other *Orthoseira* species.

Thus, *Orthoseira gremmenii* should be considered a distinct species, showing unique features that separate it from all known *Orthoseira* species.

The description of *Orthoseira gremmenii* confirms the highly specific nature of the non-marine diatom flora of Gough Island. Carter (1966) described 20 new taxa from two samples from Gough Island and (in the same paper) another 35 from nine samples from nearby Tristan da Cunha. This is not surprising when one considers that the islands are very isolated geographically and have a volcanic origin, which favours high levels of speciation. Similar results were found for other isolated islands at the same latitude. Recently, on Amsterdam Island, located in the southern Indian Ocean, three of five found *Eunotia* species were found to be new (Van de Vijver *et al.,* in press) whereas on Ile de la Possession (Crozet Archipelago) almost 25% of all recorded species were described as new (Van de Vijver *et al.,* 2002). A more thorough survey of Gough Island diatom flora is planned in the near future which may result in the description of many more new taxa.

Acknowledgements. The authors wish to thank Pierre Compère and Dr. Václav Houk for stimulating discussions and corrections of the Latin diagnosis. Ing. Marcel Verhaegen is acknowledged for his help with the Scanning Electron Microscope. Dr. Niek Gremmen collected the sample during fieldwork on Gough Island in 2000.

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