

A new *Pteroncola* species (Bacillariophyceae) from the South Shetland Islands, Antarctica

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ABSTRACT: The marine araphid diatom *Pteroncola carlinii* sp. nov. from a coastal Antarctic environment was described based on light and scanning electron microscopy observations. The new species was found as solitary cells in the phytoplankton and epiphytic on green algal filaments or diatom chains. It was distinguished from *P. inane*, the only recognized species, primarily by a well developed sternum, a hyaline mantle, apical pore fields composed of three horizontal rows of pores, a single rimoportula per valve, and a lower striae density. Two morphotypes of *P. carlinii* were distinguished based upon the ultrastructure of alveolate (chambered) striae in external view. While most specimens presented striae formed by transapically elongated slits, others showed striae composed of a row of small areolae. A revised description of *Pteroncola* was provided as well as a comparison with related genera in the Fragilariaceae, such as *Brandinia*, *Catacombas*, *Fragilaria*, *Fossula*, *Hyalosynedra*, *Staurosirella* and *Thalassioneis*.

KEY WORDS: Arcuate valves, King George Island, Potter Cove, Striae dimorphism

INTRODUCTION

The araphid diatom genus *Pteroncola* Holmes & Croll was based on examination of diatoms living on body feathers of diving sea birds from the Southern Pacific coast of the United States (Holmes & Croll 1984). A combination of morphological features including the presence of unique chambered (alveolate) striae, apical pore fields at each pole, girdle structure and band ornamentation differentiate *Pteroncola* from other araphid genera (Round *et al.* 1990). Holmes & Croll (1984) based *Pteroncola* on their new species *P. marina* Holmes & Croll, which had actually been described earlier by Giffen (1970) as *Dimeregramma inane* Giffen. Therefore, the correct name is *P. inane* (Giffen) Round in Round *et al.* (1990). Some years later, the new combination *Pteroncola hyalina* (Kützing) Guslyakov was proposed for *Diatoma hyalina* Kützing in an atlas of benthic diatoms from the Black Sea (Guslyakov *et al.* 1992). However, this is a controversial taxon that has more traditionally been considered as member of the genus *Fragilaria* Lyngbye (Hasle & Syvertsen 1981).

The relatively small size and delicate structure of *Pteroncola* frustules necessitate the use of electron microscopy for a reliable identification. Perhaps for this reason it has received only sparse attention in the recent literature. Nevertheless, *P. inane* has been observed as an epiphyte on marine algae and from sediments along European coasts

(Witkowski *et al.* 2000). By contrast, there are no records of *Pteroncola* for the Southern Ocean.

During field marine phytoplankton examinations carried out in Potter Cove, King George Island, Antarctica, small specimens closely resembling *Pteroncola inane* were observed. Detailed light (LM) and scanning electron microscopy (SEM) observations revealed morphological features that justify the proposal of a new species within *Pteroncola*.

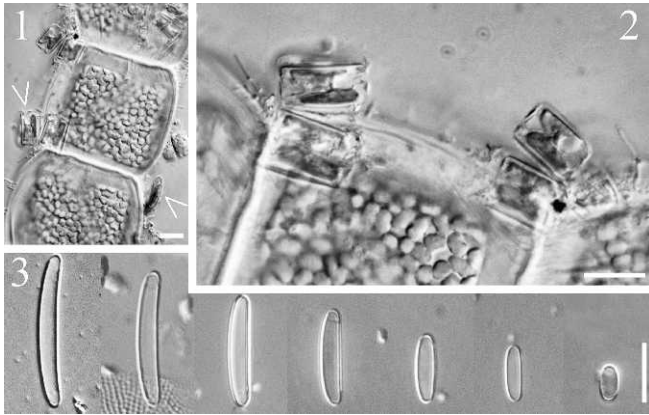
MATERIAL AND METHODS

This study was carried out at Potter Cove, a small bay located along the southwestern coast of King George Island, in the South Shetland Archipelago. Sampling was conducted close to the Argentinean Carlini Station (formerly called Jubany) as part of a long term monitoring program (see Schloss *et al.* 2012). Water was collected at two sites, the inner cove (sta. 1, 62.232°S; 58.667°W) and the outer cove (sta. 2, 62.238°S; 58.689°W). Samples were collected during the Austral summer (January and February 2010 and 2011) with 4.7 l Niskin bottles and a 20 µm mesh net. The inner cove was characterized by soft sediments and shallower depth (< 50 m) than the outer cove where the bottom was mainly rocky and depths were > 100 m. All samples were fixed with 4% acetic Lugol's solution and kept in dark conditions at room temperature until analysis. Net samples were examined using phase contrast and differential interference contrast (DIC) using two Leica DM2500 microscopes (Leica Microsystems GmbH, Wetzlar, Germany). For LM observations, organic material was removed from net subsamples using sodium hypochlorite as described in

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Figs 1–3. *Pteroncola carlinii* sp. nov. LM images. LPC 5461 and 5457. Scale bars = 10 μ m.

Figs 1–2. Epiphytic organisms on green algae.

Fig. 3. Variability of valve size and shape.

Almandoz *et al.* (2011). Clean material was then mounted on permanent slides using Naphrax mounting medium. SEM observations were made with a JEOL JSM-6360 LV (JEOL Ltd, Tokyo, Japan). Raw and cleaned materials were mounted onto glass stubs and sputter coated with Au-Pd following procedures by Ferrario *et al.* (1995). Additionally, samples were filtered onto 0.2 μ m polyamide filters.

For quantitative estimation of phytoplankton assemblage composition, cells were enumerated with a phase contrast Leica DMIL LED inverted microscope (Leica Microsystems GmbH) according to the procedures described by Utermöhl (1958). Subsamples of 50 or 100 ml were settled for 24 or 48 h, respectively, in a composite sedimentation chamber. At least 100 cells of the dominant taxa were counted in one or more strips of the chamber or within random fields at $\times 250$ or $\times 400$, depending on their concentration and size. The abundance of *Pteroncola* specimens was registered by the examination of strips at $\times 400$. Slides and aliquots of the original samples were deposited in the Diatom Collection (LPC) of the Herbarium of the División Ficología, Facultad de Ciencias Naturales y Museo, Universidad Nacional de La Plata, Argentina. Terminology follows Round *et al.* (1990).

RESULTS

Pteroncola carlinii Almandoz & Ferrario sp. nov.

Figs 1–15

DESCRIPTION: Cells rectangular in girdle view, with two plastids. Valves slightly arcuate to linear-elliptical with rounded apices, 5–24 μ m long, 2–4 μ m wide. Sternum broad, running the whole length of the valve. Striae alveolate (i.e. chambered), 36–42 in 10 μ m, opening externally via areolae or transapically oriented slits and internally via a small round foramen. Interstriae raised and usually wider than striae. An apical pore field located at each pole on the valve mantle. A single rimoportula per valve located close to the apex. Girdle composed of open bands; first copula with an irregular row of areolae, valvocopula and remaining copulae hyaline.

HOLOTYPE: Slide LPC 5461, labelled ‘Jubany 21/1/2011 sta. 2’, deposited in the Herbarium of the División Ficología, Facultad de Ciencias Naturales y Museo, La Plata, Argentina.

PARATYPE: Slide LPC 5457, labelled ‘Jubany 25/2/2010 sta. 2’, deposited in the Herbarium of the División Ficología, Facultad de Ciencias Naturales y Museo, La Plata, Argentina.

ISOTYPE: Slide ZU9/32 deposited in the Hustedt Collection, Bremerhaven, Germany.

TYPE LOCALITY: Potter Cove (62°14’S, 58°38’W), King George Island, South Shetland Archipelago, Antarctica (plankton).

ETYMOLOGY: The specific epithet has been chosen to honour the memory of Dr Alejandro R. Carlini, a researcher devoted to Antarctic science.

Intact cells typically were observed in girdle view using LM, which revealed the rectangular shape of the frustule, the relatively wide pervalar axis and the presence of two plastids (Figs 1, 2). The junction of the valve apex and valvocopula was smooth (without indentation) and there were no siliceous nodules or warts on lower valve mantle.

Valves were generally arcuate in outline with rounded apices, 5–24 μ m long, 2–4 μ m wide. The smallest valves approached a linear-elliptical outline (Fig. 3). Striae were not resolvable with the light microscope for Naphrax mounted material.

In SEM, the valve face was slightly domed and curved into a deep mantle. A conspicuous and broad sternum extended over the entire length of the valve face (Figs 4, 5). Externally, the striae were usually formed by transapically elongated slits with a single pore (Figs 6, 7). In some specimens the striae were composed of a row of small areolae (Figs 14, 15). However, in both cases the striae were alveolate (i.e. chambered) and opened internally by a single round foramen (Figs 8, 9).

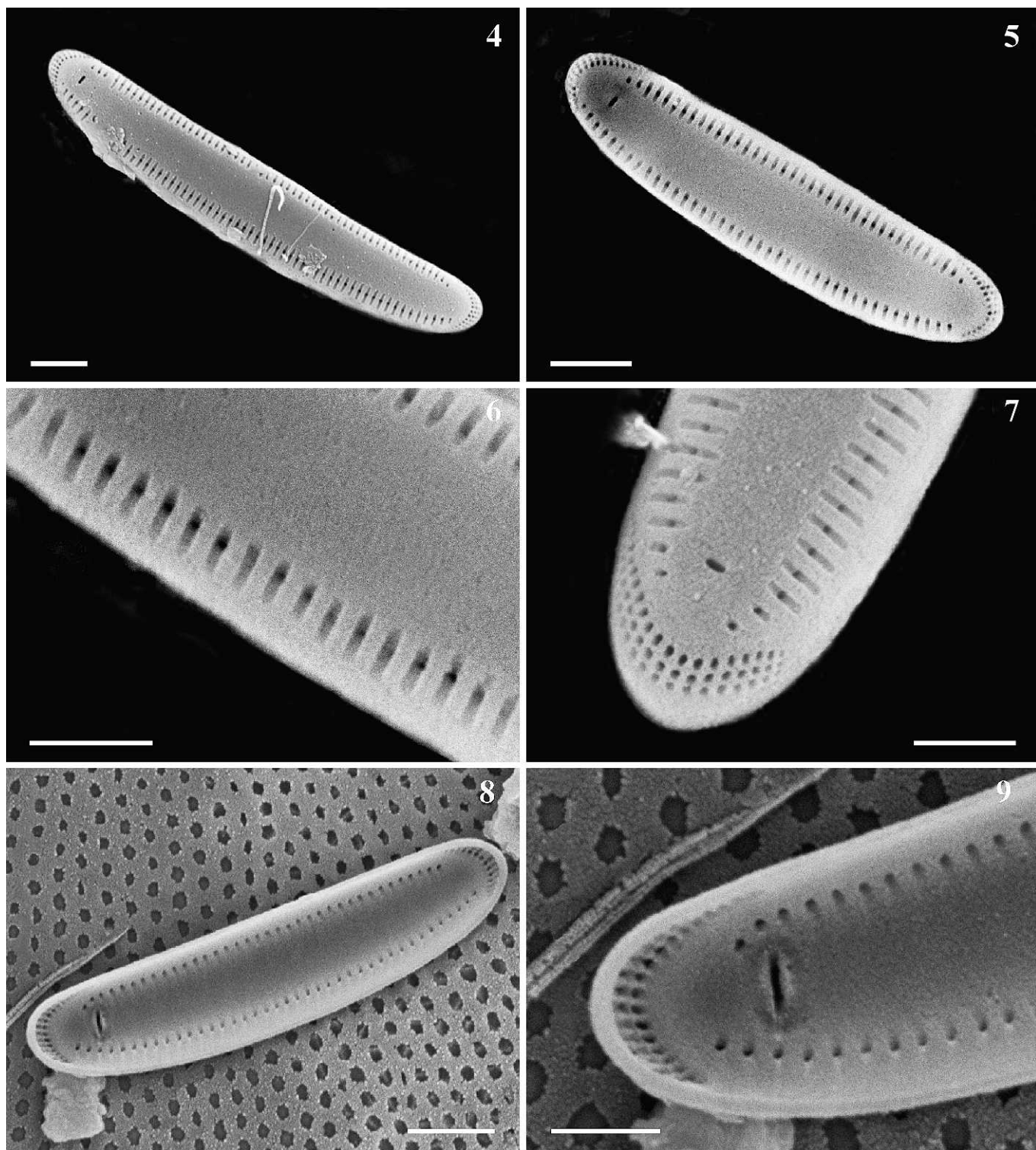
Apical pore fields were composed of three horizontal rows of pores (Figs 7, 11, 15) that also formed parallel rows vertically. The single rimoportula was orientated transapically and internally was sessile (Figs 8, 9); it opened externally via a small, narrow slit (Figs 7, 11, 14).

The valve mantle was deep and mainly hyaline (Figs 10, 11, 15). The girdle was usually composed of eight open bands (Figs 10, 13); however, a higher number of bands was observed in some dividing cells (Fig. 12). The valvocopula was plain, whereas the first adjacent copula exhibited an irregular row of small areolae; the remaining copulae were unperforated (Figs 10, 11, 15).

Pteroncola carlinii was observed only as solitary cells in the phytoplankton and epiphytic on filaments of green algae and chains of other diatoms in the genus *Thalassiosira* and more rarely on *Porosira*. The new species was found only in Potter Cove in the South Shetland Archipelago. It was observed during two consecutive summer samplings, especially at Station 2. *Pteroncola carlinii* was found at temperatures from 0.8 to 1.7°C and salinities from 33.5 to 34.1. It reached a maximum abundance of 1600 cells L⁻¹ on 31 January 2011.

DISCUSSION

Nomenclatural problems and misidentifications in the genus *Pteroncola* were pointed out by Round *et al.* (1990). Only



Figs 4–9. *Pteroncola carlinii* sp. nov. SEM images. All from LPC 5461, stubs 4–2012 (Figs 4, 7–9) and 8–2013 (Figs 5, 6). Scale bars = 2 μ m (Figs 4, 5, 8); 1 μ m (Figs 6, 7, 9).

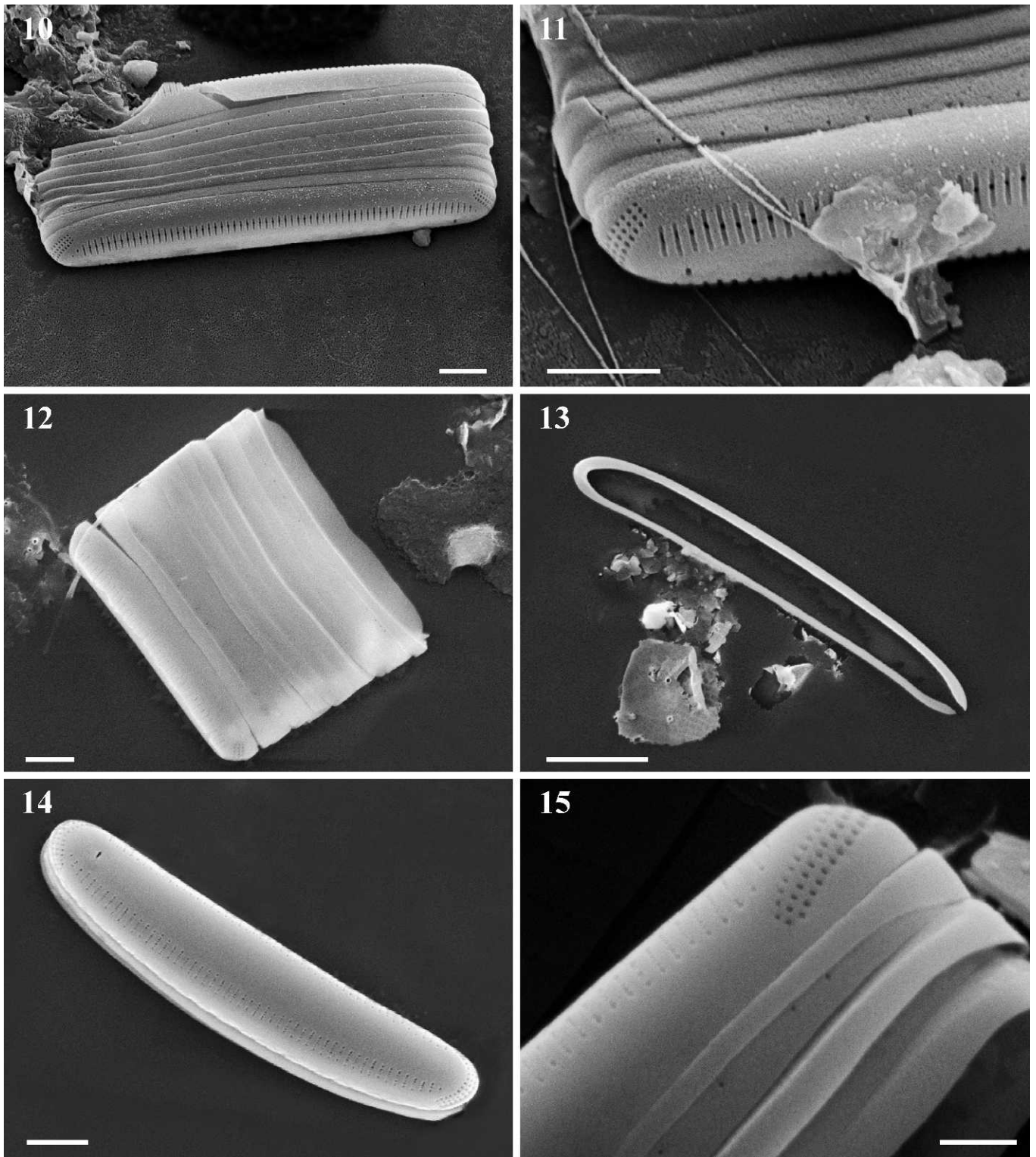
Figs 4, 5. External view showing the entire valve.

Fig. 6. Detail of striae formed by transapically elongated slits in external view.

Fig. 7. Detail of valve end showing the apical pore field, the rimoportula opening, and striae in external view.

Fig. 8. Internal view showing the entire valve.

Fig. 9. Detail of valve end showing the rimoportula, apical pore field, and striae in internal view.



Figs 10–15. *Pteroncola carlinii* sp. nov. SEM images. LPC 5461, stubs 2s/t-2012 (Figs 10, 11), 4–2012 (Fig. 13); LPC 5457, stub 2–2010 (Figs 12, 14, 15).

Figs 10–12. Girdle views showing the open bands. Note the presence of an irregular row of small areolae in the first copula. Scale bar = 2 μ m.

Fig. 13. Single band, open at one end. Scale bar = 5 μ m.

Fig. 14. External view showing the entire valve. Note the presence of striae composed of a row of small areolae. Scale bar = 2 μ m.

Fig. 15. Detail of valve end showing the apical pore field, striae and girdle bands. Scale bar = 1 μ m.

two species have been attributed to this genus by other authors: *P. inane* (Giffen) Round (basonym = *Dimeregramma inane* Giffen; synonym = *P. marina* Holmes & Croll) and *P. hyalina* (Kützing) Guslyakov. *Pteroncola marina* (Holmes & Croll 1984) was originally the generitype but Round *et al.* (1990) considered *P. marina* to be a synonym of the earlier named *Dimeregramma inane*; therefore they made a new combination based upon priority. Several subsequent works have included ultrastructural analyses of this taxon, revealing the presence of stable characters that differentiate it from other araphid genera (Round *et al.* 1990; Witkowski *et al.* 2000; Susuki & Nagumo 2004). By contrast, the new combination *Pteroncola hyalina* was proposed for *Diatoma hyalinum* Kützing by Guslyakov *et al.* (1992) but insufficient evidence supported the transference. This taxon has been generally recognized as *Fragilaria hyalina* (Kützing) Grunow (see Hasle & Syvertsen 1981). According to Guslyakov *et al.* (1992), the presence of a single rimoportula justifies the movement of this taxon out of *Fragilaria*. However, features like the kind of striae, apical pore fields and girdle structure, the presence of marginal spines and colony formation (Hasle & Syvertsen 1981) allow the differentiation of *F. hyalina* from the genus *Pteroncola*.

In this work, we have proposed the new *Pteroncola carlinii*, which is in good agreement with the original description of *Pteroncola* but presents morphological features not observed in *P. inane* that support its separation as a new species. They mainly include the presence of a well-developed sternum and a hyaline mantle, apical pore fields composed of three horizontal rows of pores, a single rimoportula per valve, and a lower striae density.

Pteroncola inane can be distinguished from *P. carlinii* in LM. An indentation of the junction of the valve apex and valvocopula and a row of irregularly spaced circular spots can be observed in girdle view of *P. inane* (Holmes & Croll 1984). The latter corresponds to thickened siliceous zones along the lower mantle placed between striae and interstriae according to Round *et al.* (1990). In contrast, in *P. carlinii* the mantle is plain (without circular spots) and there is no indentation.

The valve shape, generally arcuate in *Pteroncola carlinii* and linear-elliptical in *P. inane* can be another useful characteristic to discriminate them in valve view. However, it cannot be considered a diagnostic feature because the smaller valves of *P. carlinii* approach a linear-elliptical outline. The valve shape of *P. inane* has been described as elliptical, linear or linear-lanceolate both in Holmes & Croll (1984 as *P. marina*) and in Giffen (1970 as *Dimeregramma inane*) as well as in studies carried out by Sullivan (1979 as *Fragilaria hyalina*) and by Susuki & Nagumo (2004). By contrast, Witkowski *et al.* (2000) described the valves of *P. inane* as arcuate; however, in that report the only illustrations of the species are photographs of elliptical cells.

SEM observations reveal a number of differences between *Pteroncola carlinii* and *P. inane*. The striae density is smaller in *P. carlinii*. In *P. inane* the sternum is very narrow whereas in *P. carlinii* it is broad and runs along the entire valve length. Although the development of the sternum has been observed as a variable character in other araphid genera such as *Synedropsis* (Hasle *et al.* 1994), no intermediate forms were observed in *P. carlinii*.

The ornamentation of the valve mantle is another useful characteristic to discriminate the two species. While striae occupy nearly the entire mantle in *Pteroncola inane*, the mantle of *P. carlinii* is essentially hyaline. The presence of sessile rimoportulae, which are transapically oriented close to the valve ends and open externally through a narrow slit, is a feature shared by both species. However, two rimoportulae are typically present in *P. inane* but only a single rimoportula is present in *P. carlinii*. Apical pore fields also show a different structure. In *P. carlinii*, they are composed of three horizontal rows of pores, whereas four or five rows are present in the apical pore fields of *P. inane* (Holmes & Croll 1984; Round *et al.* 1990; Susuki & Nagumo 2004).

The girdle structure is similar in both species, showing several open bands. In the new species, the valvocopula and all copulae except the first copula are unperforated. The first copula is perforated by an irregular row of poroids close to the pars interior. In contrast, Holmes & Croll (1984) show a valvocopula with a single row of irregularly spaced small pores parallel to the valve edge. However, a plain valvocopula and an ornamented first copula can be also observed in images of *Pteroncola inane* provided by Round *et al.* (1990), Witkowski *et al.* (2000) and Susuki & Nagumo (2004).

Pteroncola inane has been recorded as an epizoic, epiphytic and epilithic species (Holmes & Croll 1984; Witkowski *et al.* 2000). On the other hand, our new species *P. carlinii* was mostly observed as free-living planktonic cells but occasionally as an epiphyte on green algae and other diatoms. Given that only bottle and net plankton samples collected from coastal areas were included in this study, it is hard to establish whether the free-living cells commonly observed during microscopic examinations are really euplanktonic or could have detached from their host during sample manipulation.

The genus *Pteroncola* was erected to accommodate specimens with an atypical kind of striae, which had never been described in other araphid genera (Holmes & Croll 1984). According to these authors, 'striae are a thin membrane depressed below the exterior surface of the interstriae and perforated by an unoccluded circular pore which is located approximately midway between the axial zone and the valve mantle'. However, Round *et al.* (1990) later introduced the concept of chambered (alveolate) striae in *Pteroncola*, opening to the outside by a row of small areolae and to the inside by a small round foramen. Curiously, they did not provide any explanation for this discrepancy with the original description of the genus.

In our new species two morphotypes could be distinguished in relation to the ultrastructure of striae in external view. While most specimens presented striae composed of longitudinal slits (Figs 6, 7, 11), others showed striae composed of a row of simple areolae (Figs 14, 15). In both cases, they open internally by a single round foramen. Moreover, the same striae density was observed in both morphotypes. Therefore, there is a dimorphism in the external ultrastructure of striae in *Pteroncola carlinii*. In the araphid freshwater genus *Staurosirella*, the striae are composed of slit-like areolae transversally separated by narrow bridges of silica. These slits may be lost during rigorous cleaning of samples causing an artificial areolae loss

Table 1. Comparison of key features among *Pteroncola* and similar genera in the Fragilariaceae.

	<i>Brandinia</i> ¹	<i>Catacombas</i> ²	<i>Fragilaria</i> ³	<i>Fossula</i> ⁴	<i>Hyalosynedra</i> ²	<i>Pteroncola</i>	<i>Staurosirella</i> ⁵	<i>Thalassioneis</i> ⁶
Chloroplasts	two	numerous	two	two	?	two	two	numerous
Cells	solitary	solitary	usually ribbon-shaped colonies	ribbon-shaped colonies	solitary	solitary	stellate and ribbon-shaped colonies	usually curved and twisted colonies
Striae	uniseriate, poroid areolae externally occluded	uni- or multiseriate, alveolate	uniseriate, poroid areolae	uniseriate, poroid areolae	mostly uniseriate, alveolate	uniseriate/slits, alveolate	uniseriate, slit-like areolae parallel to the apical axis	uniseriate, poroid areolae
Apical fields	ocellulimbus	ocellulimbus	ocellulimbus	two irregular horizontal rows of pores	ocellulimbus	three to five horizontal rows of pores	several vertical rows of pores	several vertical rows of pores
Rimoportulae	two per valve, at the valve ends	two per valve, at the valve ends	one per valve, at the valve end	one per valve, at the valve mantle	one per valve, at the valve end	one or two per valve, at the valve ends	absent	one per valve, near valve center
Bands	open, with a single row of poroids	open, plain or with a single row of poroids	open, with a single row of poroids	open, with vertical rows of poroids	open, plain	open, most plain but others with a single row of poroids	open or closed, plain	open, with 2–4 rows of poroids
Spines	not present	a few overhang the pore plate	present	not present	a few overhang the pore plate	not present	present or absent	not present
Habitat	marine, epilithic	marine, epiphytic	mainly freshwater	marine planktonic	marine, epiphytic	marine, epiphytic, epizoic, epilithic, planktonic	freshwater, epipsammic	sea ice

¹ Fernandes *et al.* (2007).² Williams & Round (1986).³ Williams & Round (1987).⁴ Hasle *et al.* (1996).⁵ Morales & Manoylov (2006).⁶ Ferrario *et al.* (2012).

(Round *et al.* 1990; Morales & Manoylov 2006). Perhaps this may explain the presence of slits instead of areolae in our observed specimens of *P. carlinii*. Nevertheless, both morphotypes were observed in cleaned and untreated material. In addition, the two morphotypes were never observed in the same sample and intermediate forms were not observed. Another explanation to this variability could be linked to the development of resting stages, which is known to induce morphological changes in Antarctic diatoms (Hasle *et al.* 1996; MacQuoid & Hobson 1996). Alternatively, the two morphotypes represent independent cryptic species.

Besides the alveolate structure of the striae, *Pteroncola* possess a combination of morphological characters that differentiate it from other similar araphid genera in the Fragilariaceae (Table 1). The apical pore fields of *Pteroncola* are composed of horizontal rows of small pores and resemble those of *Fossula*, although only two irregular rows of pores are found in the latter. In *Staurosirella* and *Thalassioneis* the rows of pores of the apical fields are arranged in vertical position, whereas in *Brandinia*, *Catacombas*, *Hyalosynedra* and *Fragilaria* an ocellulimbus is present. The apical location of the rimoportula in *Pteroncola* towards the ends of the valve face is similar to that observed in *Brandinia*,

Catacombas, *Fragilaria* and *Hyalosynedra*. However, the rimoportula is situated very near the valve centre in *Thalassioneis* and over the mantle in *Fossula*, while rimoportulae are absent in *Staurosirella*.

The structure of the girdle bands results another useful feature to discriminate *Pteroncola* from other related genera. While only a few bands possess a single row of poroids and most are hyaline in *Pteroncola* and *Catacombas*, all copulae have a row of poroids in *Brandinia* and *Fragilaria*. Multi-seriate rows of poroids are found in the bands of *Fossula* and *Thalassioneis* whereas all copulae are smooth in *Hyalosynedra* and *Staurosirella*.

Revised description of the genus *Pteroncola*

Solitary cells. Frustule rectangular in girdle view, with two plastids. Valves elliptical to arcuate in outline. Chambered (alveolate) striae, with transapically elongated slits or a row of areolae in external view, indistinguishable by light microscopy. Sternum barely perceptible to well developed, delicate and limited to the valve tips or broad and running the entire valve length. Mantle deep, varying from striate to almost hyaline. Rimoportulae either one or two in number and located at valve ends, opening externally through a narrow slit. Apical pore fields at both poles, composed of three to five horizontal

rows of pores. Girdle composed of several open bands; either with a single row of pores or hyaline.

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