



<http://dx.doi.org/10.11646/phytotaxa.239.3.9>

***Pinnularia caprichosa* sp. nov.: a diatom from a black water Brazilian Amazon system**

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Abstract

While possessing a remarkable diversity in the acidic and oligotrophic waters of the Amazon basin, the genus *Pinnularia* appears underdescribed in the region. In this study, we present light and scanning electron microscopical observations on *Pinnularia caprichosa* sp. nov. from Tupé Lake, a dendritic lake located on the floodplain of the Negro River basin. This new taxon has a large axial area and transapical striae that are slightly radiate to parallel and longer in the middle portion of the valve. The species was compared with *Pinnularia elliptica*, *P. instabilis*, *P. lacunarum*, *P. montana*, *P. permontana* and *P. subflexuosa*, all of which closely resemble *P. caprichosa* but differ from the new species in specific details of size, striae density and valve shape.

Key words: Bacillariophyta, *Pinnularia*, taxonomy, Amazonian region, South America, scanning electron microscopy

Introduction

Pinnularia Ehrenberg (1843: 45) is one of the richest genera in the Brazilian Amazon, and to date 49 species have already been discovered in a wide range of aquatic environments such as rivers, lakes and ‘igarapés’ (narrow and not very deep water streams inside the tropical forest), as recorded by Hustedt (1965), Metzeltin & Lange-Bertalot (1998, 2007) and Pereira *et al.* (2012, 2013, 2014). The current report is the third in a series of contributions to the knowledge of the genus *Pinnularia* from Tupé Lake. The first publication detailed the presence of 28 taxa, with three first reports of *Pinnularia* species or varieties to the Brazilian Amazon (Pereira *et al.* 2013). The second publication described four new *Pinnularia* species from Amazonian black water (Tupé Lake, Amazonas State, Brazil) (Pereira *et al.* 2014). The special oligotrophic and low pH conditions and, probably, the contribution of allochthonous inputs from ‘igarapés’ to Tupé Lake, are important factors influencing *Pinnularia* species richness in this system. In addition to the known diversity of the genus, there appears to be additional taxa awaiting description in the relatively unexplored regions of the Amazon. In the present study we present light (LM) and scanning electron microscopy (SEM) observations of a new species of *Pinnularia*, and compare it with other similar *Pinnularia* species.

Material & Methods

Tupé Lake ($03^{\circ} 02' 35.4''$ S– $60^{\circ} 15' 17.5''$ W) is a typical black-water aquatic system, according to the classification system of Sioli (1984). The lake is located on the left bank of the Negro River, to the west of Manaus, Amazonas State, Brazil. The lake is connected with the Negro River by a channel, and with several ‘igarapés’ whose levels vary according to the floods of the system. Four periods can be identified during an annual cycle in an amazon floodplain

lake such as Tupé Lake: rising, high, falling and low level water periods (Junk *et al.* 1989). Lake Tupé is situated on a slightly elevated area of land. Water depth ranges from 4 to 15 meters in the central region of the lake. The waters of Tupé Lake are poor in suspended material and minerals, but rich in organic compounds and humic acids (Rai & Hill 1981).

This study was based on samples taken at four stations in the lake in December 2002 and 2003 (low water period), and September 2008 (falling water period), including samples of plankton and sediments. Plankton samples were collected with a 25 µm mesh net and the surficial sediments (2 cm depth) were obtained using a sediment corer. The samples were fixed with Transeau solution (Bicudo & Menezes 2006). Aliquots of sample were concentrated and cleaned according to the method of von Stosch (1970). For light microscopy (LM), material was mounted on permanent slides using Naphrax® and examined under a Zeiss Axioplan microscope (1000×). Figures were obtained with a Zeiss AxioCam ERC5s camera. For scanning electron microscopy (SEM), cleaned material was mounted on glass on stubs, coated with gold-palladium (16 nm), and analyzed using a Jeol JSM-6060 operated at 15–20 kV. Terminology used to describe morphological features follows Barber & Haworth (1981), Round *et al.* (1990) and Krammer (2000). Some physical and chemical variables of the water were measured, such as temperature (Yellow Springs Instruments model 55), pH, electrical conductivity, dissolved oxygen (Yellow Springs Instruments model 63), water depth, and transparency using a Secchi disk.

Results

Pinnularia caprichosa A.C. Pereira & Torgan, sp. nov. (Figs 1–22)

Valves linear, elliptic to lanceolate, with subcapitate-rounded apices. Axial area around ½ of the breadth of the valve, narrowing towards the apices (Figs 1–12). Length 123.6–166.9 µm (average 141.5 µm; s.d. 12.1), breadth 30.9–36.4 µm (average 33.2 µm; s.d. 1.5) (n=26). Length/breadth ratio 3.5–4.6. Raphe lateral, weakly curved near the proximal endings. Proximal fissures drop-like central pores deflected to the primary side reaching the mantle (Figs 16, 18). Distal fissures hooked-shaped, curved towards secondary side of valves (Figs 13–15). Internally, distal fissures slightly deflected ending in prominent helictoglossae distant 2.1–2.3 µm from apices (Figs 19, 20). Internal raphe branch moderately lateral and almost straight (Fig. 21). Transapical striae slightly radiate, parallel and longer in the middle, convergent to parallel at the ends (Figs 14, 16, 21). Striae 8–9 in 10 µm (Fig. 17). Alveoli pattern with 5–7 rows of rounded areolae, each row with 25–26 areolae in 5 µm (Figs 17, 22).

Type:—BRAZIL. Amazonas: Tupé Lake, sediment at station 2, 03° 02' 17.1" S, 60° 15' 45.3" W, S. Melo, 23 September 2008 (holotype HAS! 6716, circled specimen on slide, here illustrated as fig. 2; isotype BR! 4419, circled specimen on slide).

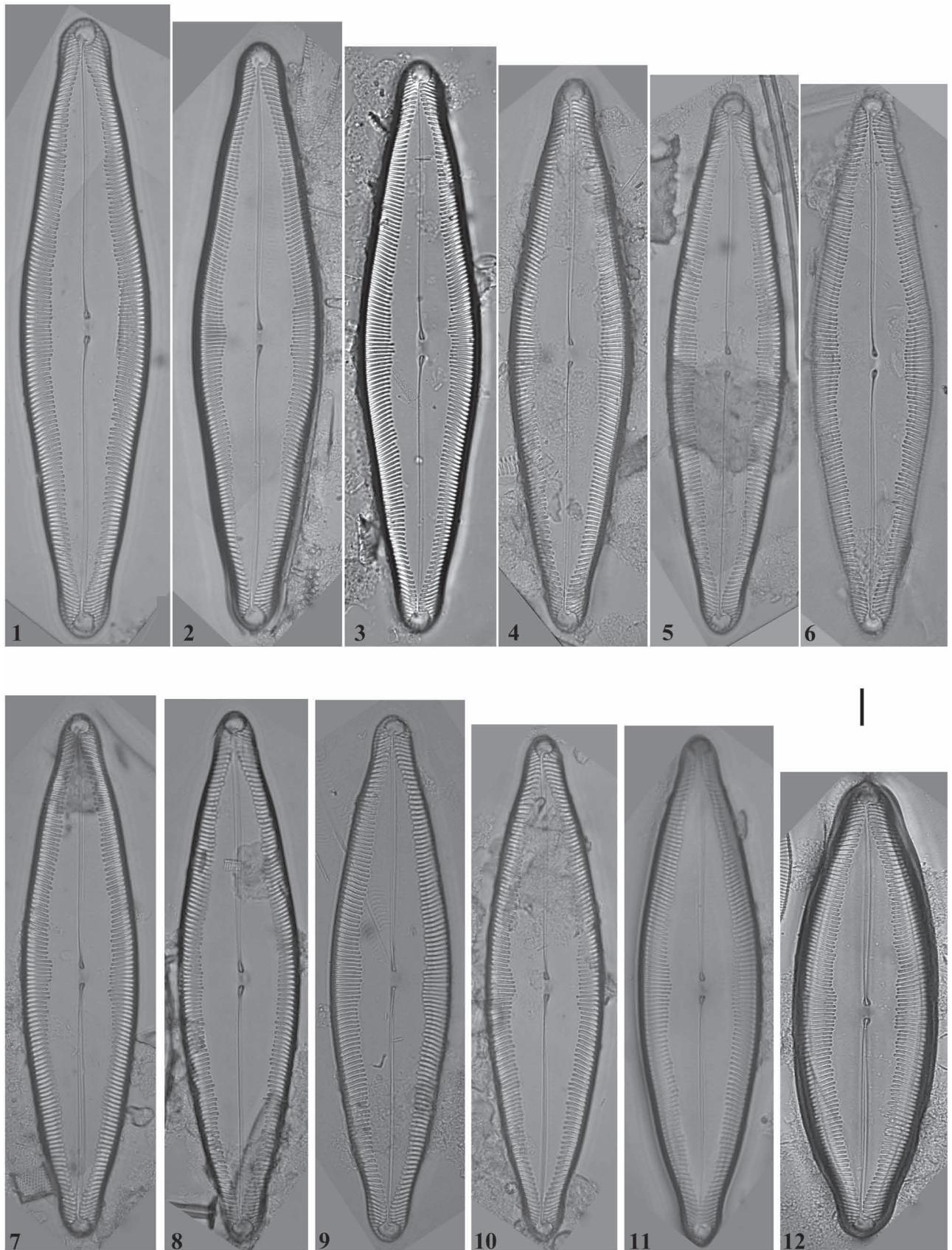
Etymology:—This taxon is dedicated to the group Caprichoso, from “Boi Bumbá” Folkloric Festival, Parintins City, Amazonas State.

Remarks:—*Pinnularia caprichosa* is similar to some other species of the genus found in the Amazon region. Comparison to other similar taxa is presented in Table 1. *Pinnularia hybrida* (Peragallo & Héribaud in Héribaud 1893: 85; pl. 4, fig. 9) Cleve-Euler (1915: 29; pl. 2, fig. 32) and *P. montana* Hustedt (1927: 155; pl. 5, fig. 2), for example, differ from *P. caprichosa* mainly by their breadth, while *P. lacunarum* Hustedt in Schmidt *et al.* (1934: pl. 389, figs 4–5) *emend.* Schoeman (1969: 63; pl. 4, figs 80–81) (Hustedt 1942: 85–86, figs 164–165) on the other hand, has smaller dimensions. *Pinnularia subflexuosa* Hustedt in Schmidt *et al.* (1934: pl. 389, figs 1–2) is larger, and also differs in striae density and valve shape. *Pinnularia instabilis* (A.W.F. Schmidt in Schmidt *et al.* 1876: pl. 43, figs 37, 39) Metzeltin in Metzeltin & Lange-Bertalot (1998: 178–179; pl. 194, figs 1–3, pl. 205, figs 4–5) differs in valve length (69–91 µm), width (21–23 µm), in striae pattern (striae parallel with above of 9 striae in 10 µm), and in the shape of the apices (obtuse not subcapitate-rounded). *Pinnularia permontana* Krammer & Metzeltin in Metzeltin & Lange-Bertalot (1998: 183–184; pl. 198, fig. 1) is also smaller (length 121 µm, breadth 23 µm), with more than 9 striae in 10 µm, and differs by having broadly rounded apices (not subcapitate-rounded apices). *Pinnularia caprichosa* resembles *P. elliptica* K.E. Lohman & G.W. Andrews (1968: E19; pl. 2, figs 1–2) (non *P. elliptica* Rabenhorst (1853: 42; pl. 6, fig. 23a–b) nec *P. elliptica* Frenguelli (1942: 146; pl. 3, fig. 27)), but this species, described from Eocene nonmarine sediments in Wyoming, U.S.A., has elliptical valves with rounded to subacute apices, as well as a slightly asymmetric axial area and some very short striae alternating with longer striae in central part of the valves. We summarize the differences between the new species and the most similar taxa in Table 1.

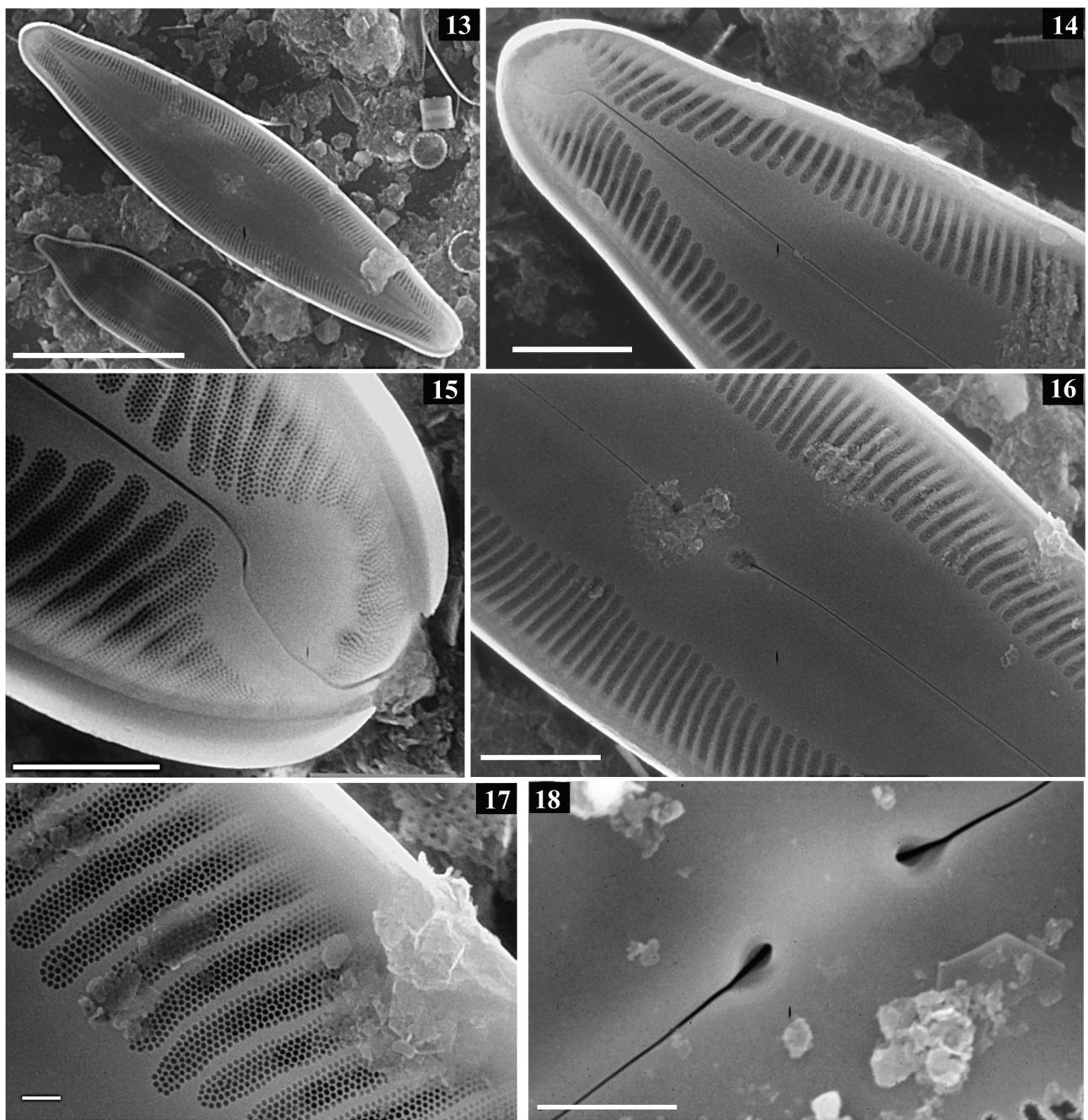
TABLE 1. Morphological and morphometric data for *Pinnularia caprichosa* sp. nov. from Tupé Lake compared to closely related *Pinnularia* species.

Taxa	<i>P. caprichosa</i>	<i>P. elliptica</i>	<i>P. hybrida</i>	<i>P. instabilis</i>	<i>P. instabilis</i> (A.W.F. Schmidt)	<i>P. lacunarium</i> (A.W.F. Schmidt)	<i>P. montana</i>	<i>P. permontana</i>	<i>P. permontana</i> Krammer & Metzelin	<i>P. permontana</i> Hustedt	<i>P. subflexuosa</i> Krammer & Metzelin	<i>P. subflexuosa</i> Hustedt
References	(this study)	Lohman & Andrews (1968)	Cleve-Euler (1915)	Metzelin & Lange-Bertalot (1998)	Pereira <i>et al.</i> (2013)	Hustedt (1942)	Hustedt (1927)	Lange-Bertalot (1998)	Pereira <i>et al.</i> (2012)	Pereira <i>et al.</i> (2013)	Schmidt <i>et al.</i> (1934)	
Valve length (μm)	123.6–166.9	90–160	75–215	69–91	61–85	110–135	120–150	121	118.5–120	105–118.5	229–293.3*	
Valve width (μm)	30.9–36.4	30–40	13–30	21–23	17–20	27	25–30	23	20–23.7	20–23.7	58.7–60*	
Valve outline	Elliptic-lanceolate	Linear-elliptical	Linear-lanceolate	Linear	**	Linear-	Elliptic-lanceolate	Linear-elliptic-lanceolate	Linear-elliptic	**	**	
Valve apices	Subcapitate-rounded	Rounded-subacute	rounded, obtuse, sometimes subacute	Attenuated-apiculate	**	Broadly rounded	Broadly rounded	Broadly rounded	Broadly rounded	Broadly rounded	**	**
Axial area	Wide	Wide	Wide	Wide	**	Wide	Wide	Wide	Wide	Wide	**	**
Raphe	Lateral	Straight	**	Filiform	**	Straight	Filiform	Latera	Latera	Lateral	**	**
Number of striae in 10 μm	8–9	7–8	10	9–10	10–12	8	7	9–10	10–11	10–11	10–11	6*

* measured in the illustrations, ** without description.



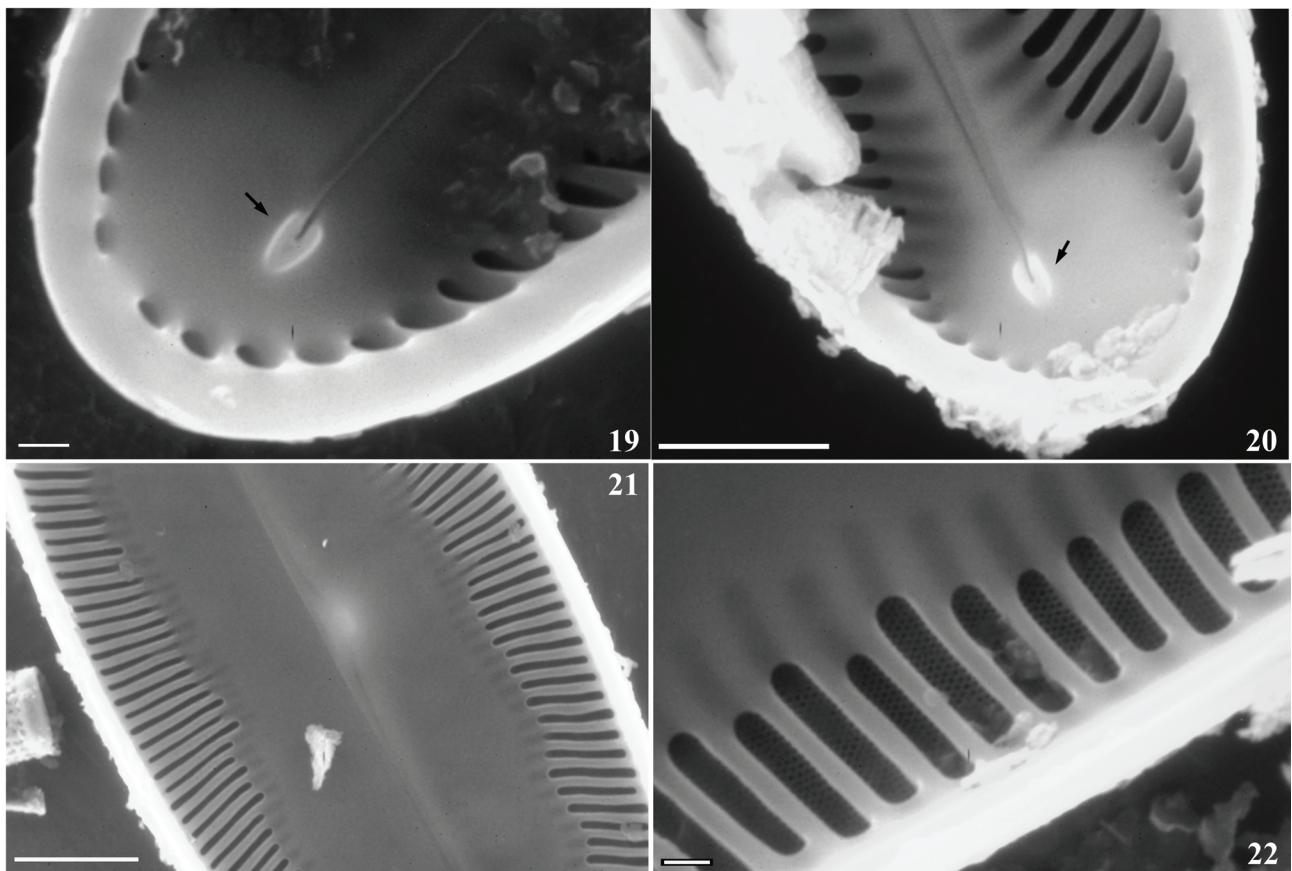
FIGURES 1–12. *Pinnularia caprichosa* A.C. Pereira & Torgan, sp. nov. LM, General valve view. Scale bar: 10 µm.



FIGURES 13–18. *Pinnularia caprichosa* A.C. Pereira & Torgan, *sp. nov.* SEM external views. 13. External valve view. 14, 15. Detail of the sickle-shaped raphe fissure endings. 16, 18. Central area showing proximal raphe fissures slightly bent to the same direction. 17. Striae and alveoli pattern rows of rounded areolae. Scale bars: Fig. 13 = 50 µm; Figs 14, 16 = 10 µm; Figs 15, 18 = 5 µm; Fig. 17 = 1 µm.

Ecological observations:—Physical and chemical conditions measured from Tupé Lake are shown in Table 2. The water samples studied by us had high water temperature (28.6–31.1°C), as expected since these Amazonian ecosystems, located in tropical latitudes, have high sunlight incidence. The slightly acidic water (pH 4.01–5.10) and very low electrical conductivity ($6.02\text{--}8.60 \mu\text{S cm}^{-1}$) values are common in Amazonian water bodies, due to the characteristics of soils in the Amazon region and adjacent regions (Sioli 1984). In general, due to the low electrical conductivity, low turbidity and low concentrations of nutrients, these ecosystems studied are considered as “black water”, according to the classification proposed by Sioli (1984) for streams in the Amazon region.

Pinnularia is a benthic genus (Round *et al.* 1990) and its presence in the plankton is unusual (Pereira *et al.* 2012). The new species has not been observed during rising and high water periods. The vertical circulation of the water column and the suspension of the bottom sediments during the falling and low waters may explain the presence of *P. caprichosa* in the plankton of Tupé Lake.



FIGURES 19–22. *Pinnularia caprichosa* A.C. Pereira & Torgan, sp. nov. SEM internal views. 19–20. Ends showing terminal raphe with prominent helictoglossae (arrow). 21–22. Alveoli pattern rows of rounded areolae. Scale bars: Figs 19, 22 = 1 µm; Figs 20, 21 = 5 µm.

TABLE 2. Physical and chemical water conditions during the falling and low waters in Tupé Lake, Amazonas where *Pinnularia caprichosa* was found.

Station	Month/Year	Depth (m)	Transparency (m)	Temperature (°C)	pH	Conductivity ($\mu\text{S cm}^{-1}$)	Dissolved oxygen (%)
1	Dec/2002	2.20	1.10	31.1	4.01	8.60	85.5
2	Sep/2008	2.40	1.00	29.8	4.50	7.30	40.9
7	Dec/2003	0.30	0.30	28.6	5.10	7.10	50.0
10	Dec/2002	5.50	1.10	31.1	4.54	6.02	65.0
10	Dec/2003	4.80	0.70	29.9	4.98	6.30	45.0

Pinnularia caprichosa was associated with several other species of *Pinnularia* including *P. angustivalva* Krammer & Metzeltin in Metzeltin & Lange-Bertalot (1998: 164), *P. confirma* Metzeltin & Krammer in Metzeltin & Lange-Bertalot (1998: 166), *P. instabilis* and *P. instabiliformis* Krammer & Metzeltin in Metzeltin & Lange-Bertalot (1998: 177), and with the exception of the latter, this diatom association is so far found only in black waters of the Amazon basin.

Acknowledgments

We thank the Universidade Federal do Oeste do Pará (UFOPA) and the Fundação Zoobotânica do Rio Grande do Sul (FZB) for logistics and laboratory facilities To CNPq (Conselho Nacional de Desenvolvimento Científico e Tecnológico) for financial support. We also extend thanks to Priscila Tremarin, Dávia Marciana Talgatti and Bart Van de Vijver for technical support. We are grateful to the anonymous reviewers for their valuable contributions.

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