

***Humidophila* (Bacillariophyceae) in subtropical mountain springs and subaerial habitats, Paraná state, Brazil, with the description of *H. piraquarae* sp. nov.**Paula Carolina FERREIRA<sup>1\*</sup>, Vanessa Merlo KAVA<sup>2</sup> & Thelma Alvim Veiga LUDWIG<sup>1</sup><sup>1</sup>Universidade Federal do Paraná, Centro Politécnico, Setor de Ciências Biológicas, Departamento de Botânica, Laboratório de Ficologia, Jardim das Américas, CEP 81531-980, Curitiba, Paraná, Brasil;  
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**Abstract:** The genus *Humidophila* is mainly characterized by small cells, striae often composed by one elongated areola occluded by thin hymenes. Species are distributed globally and are common in moist sub-aerial habitats. We identified five *Humidophila* species found in the well-conserved freshwater bodies and subaerial wet habitats from the mountain region “Mananciais da Serra”. Four previously known species (*Humidophila brekkaensis*, *H. contenta*, *H. nienta*, *H. paracontenta*) and one new species (*Humidophila piraquarae* sp. nov.) were described and illustrated based on light and scanning electron microscopy observations. *Humidophila piraquarae* sp. nov. is similar to *H. nienta* (Carter) Lowe, Kociolek, Johansen, Van de Vijver, Lange–Bertalot et Kopalová and *H. undulata* Lowe, Kociolek, Johansen, Van de Vijver, Lange–Bertalot et Kopalová, but is best distinguished by their prominent constriction, apices broadly rostrate–rounded, striae density, raised siliceous thickening in the apical region of the axial area, and raphe proximal endings laterally expanded in anchor-shaped depressions. Additionally, a list of *Humidophila* taxa already registered in Brazil is presented.

**Keywords:** diatom, *Humidophila*, morphology, new taxon, subaerial habitats, taxonomy

**INTRODUCTION**

The genus *Humidophila* comprises diatoms with small valves, usually less than 20 µm. The valves are linear, linear–elliptic, or elliptic; the apices are broadly rounded, and proximal raphe endings are straight, T-shaped or slightly anchor-shaped. The striae on the valve face are composed of one elongated areola and the valve mantle is ornate by a row of usually elongated areolae (LOWE et al. 2014). Species of the genus are distinguished by valve dimensions, number of striae in 10 µm, valve outline, striae length and striation pattern, shape of proximal and distal raphe endings, mantle areolae row interrupted or not around the apices, shape and size of the central area (LOWE et al. 2014; KOPALOVÁ et al. 2015; KOCHMAN–Kędziora et al. 2016; TAYLOR & COCQUYT 2016).

Most species of *Humidophila* prefer moist, humid and aerophilous habitats (LOWE et al. 2014). Representatives of the genus are found in

acidic oligotrophic waters, commonly in damp mosses and rocks (TAYLOR & COCQUYT 2016). Species of *Humidophila* are globally distributed, but several taxa are confined to hotspots, such as the Antarctic and Subantarctic regions (KOPALOVÁ et al. 2015). Taxonomic studies registering *Humidophila* species were developed in the Antarctic Region (VAN DE VIJVER et al. 2002; KOPALOVÁ et al. 2015), Asia (LOWE et al. 2017), Europe (LANGE–BERTALOT & GENKAL 1999; LANGE–BERTALOT & WERUM 2001; WERUM & LANGE–BERTALOT 2004; NIKOLIC et al. 2020), South America (RUMRICH et al. 2000; METZELTIN et al. 2005; VOUILLOUD et al. 2022), and New Caledonia (MOSER et al. 1998).

In Brazil, the occurrences of *Humidophila* are registered in floristic surveys, totaling 12 species citations until 2021. TREMARIN et al. (2009) catalog seven species of *Humidophila* to the state of Paraná. No exclusive study focusing only the genus *Humidophila* was developed.

We provided descriptions and illustrations of five *Humidophila* species of the pristine freshwater bodies and subaerial wet habitats from the mountain region in the east part of Paraná state. One new species of *Humidophila* is described, illustrated with LM and SEM, and compared to similar taxa. A list of *Humidophila* taxa already registered in Brazil is presented.

## MATERIALS AND METHODS

The present study was carried out in an Environmental Protection area (APA) of Piraquara city, Paraná state, Brazil. The region is inserted in the Marumbi Park at the coordinates 25°29' S, 48°58' W and covers the mountain springs, located west of the Serra do Mar, in the Atlantic Forest (REGINATO & GOLDENBERG 2007).

The samples were collected in subaerial environments and water bodies inserted in a mountainous conservation area with very low anthropic interference. Subtropical mountain springs are, often, characterized by clear and crystalline water, low temperature, low concentration of nutrients, and acidic pH (TUNDISI & MATSUMURA–TUNDISI 2008).

Samplings were carried out in 2013 and 2014, in streams and small water collecting cement reservoirs with very low anthropogenic impact, thus constituting a well-conserved environment. Eight periphytic, samples were obtained by collecting mosses (epiphytic) and submerged sand (epipsamic) and rocks (epilithic), as well as wet sub aerial substrates. The samples were fixed with the Transeau solution in a proportion of 1:1 (BICUDO & MENEZES 2006). pH (pH-meter PHTEK model pH-100), conductivity ( $\mu\text{S}^{-1}\cdot\text{cm}$ ) and water temperature ( $^{\circ}\text{C}$ ) were measured (Table 1).

The samples were cleaned according to SIMONSEN (1974) modified by MOREIRA-FILHO & VALENTE-MOREIRA (1981). Permanent slides were mounted with Naphrax® resin (I.R.: 1.74) for analysis of the specimens under light microscopy (LM). Illustrations of the species were obtained using an Olympus DP71 capture camera attached to Olympus BX40 light microscope. For the SEM observations, the oxidized samples were deposited on aluminum stubs, metalized with gold in a Balzers Union SCD 030 apparatus, and analyzed in the JEOL JSM 6360 scanning electron microscope, at the Electron Microscopy Center of the Federal University of Paraná (UFPR).

Samples and their respective permanent slides were deposited in the Herbarium of the Botany Department of the UFPR (UPCB) (Table 1). Diatom terminology follows ROUND et al. (1990) and LOWE et al. (2014).

Based on literature data, a list of *Humidophila* taxa already registered in Brazil is presented on Table S1, including geographic distribution records and proposals for correcting some misidentifications. The new species *Humidophila piraquarae* P.C.Ferreira et T.V.Ludwig is compared with similar species on Table S2.

## RESULTS

Five species were described and illustrated, including one new species, *Humidophila piraquarae* sp. nov. We

registered *H. contenta* and *H. paracontenta* in all studied samples.

***Humidophila* (Lange–Bertalot et Werum) Lowe, Kociolek, Johansen, Van de Vijver, Lange–Bertalot et Kopalová 2014**

***Humidophila brekkaensis* (Petersen) Lowe, Kociolek, Johansen, Van de Vijver, Lange–Bertalot et Kopalová 2014 (Figs. 1–26)**

Lowe et al. (2014): Diatom Research, v. 29, n. 4, p. 351–360.

### Description (n=30)

**LM (Figs 1–21):** Valves linear, slightly gibbous in the middle, length 7.3–15.7  $\mu\text{m}$ , width 2.6–3.4  $\mu\text{m}$ , with rounded apices. Axial area linear. Central area elliptical.

**SEM (Figs 22–26):** Externally, raphe filiform with external central and distal ends straight. Axial area linear. Central area elliptical, large, delimited by shorter central striae, not reaching the valve margins. Striae parallel, 25–40 in 10  $\mu\text{m}$ , formed by a single transapically elongated areola. Mantle areolae positioned at valve face, rounded, 30–35 in 10  $\mu\text{m}$ , interrupted around the apices. Copulae perforated by elongated slits. Internally, axial area linear. Central area elliptical.

**Samples occurrence:** UPCB–76004, 76005, 76059, 76066, 76074.

**Literature:** as *Navicula brekkaensis*: KRAMMER & LANGE–BERTALOT (1986); as *Diademes brekkaensis*: WERUM & LANGE–BERTALOT (2004), HOFMANN et al. (2013), WOJTAL (2013); as *Humidophila brekkaensis*: LOWE et al. (2014).

**Remarks:** *Humidophila brekkaensis* have a typical gibbous central valve, and the mantle areolae are situated on the valve face (WERUM & LANGE–BERTALOT 2004).

There are three citations of *H. brekkaensis* in Brazil. OLIVEIRA et al. (2002), in the state of Rio Grande do Sul, registered only one specimen illustrated under light microscopy, showing very slight gibbosity. However, without observing the presence of typical mantle areolae by electron microscopy, it is not possible to confirm the identity of the species. SANTOS et al. (2011) present a population that in our opinion truly belongs to *H. brekkaensis*, but has been misidentified as *Diademes pseudolangebertalotii* Metzeltin, Lange–Bertalot et Garcia–Rodriguez (description on p. 307) and *Diademes langebertalotii* Van de Vijver et Le Cohu (Figs 27–29). Finally, MARQUARDT et al. (2018) in reservoirs of the state of São Paulo illustrate specimens of *H. brekkaensis*, by SEM (Figs 111–112), but the LM images (Figs 65–69), without central gibbosity, appear to belong to *H. biscutella* (Figs 65–69, 107).

*Humidophila brekkaensis* was found in periphytic assemblages associated to *Potamogeton polygonus* Chamisso et Schlechtendal (SANTOS et al. 2011), in moss samples, almost exclusively taken from dry

mosses like *Campylopus* Spach or from liverworts (CHATTOVÁ et al. 2018).

***Humidophila contenta* (Grunow) Lowe, Kociolek, Johansen, Van de Vijver, Lange–Bertalot et Kopalová 2014 (Figs 27–54)**

Lowe et al. (2014): Diatom Research, v. 29, n. 4, p. 351–360.

**Description (n=36)**

**LM (Figs 27–49):** Valves linear, slightly concave or convex in the middle, length 6.7–10.8 µm, width 2.6–3.3 µm, with widely rounded apices. Axial area linear and central area expanded in fascia, extending from the central nodule to the valve margins.

**SEM (Figs 50–54):** Externally, raphe filiform with central and distal ends T-shaped. Axial area linear and central area expanded in fascia, extending from the central nodule to the valve margins. Striae parallel, 30–40 in 10 µm, formed by a single transapically elongated areola. Mantle areolae elongated, 40 in 10 µm, uninterrupted around the valve apices. Copulae perforated by elongated slits. Internally, axial area linear and central area expanded in fascia, extending from the central nodule to the valve margins.

**Samples occurrence:** UPGB–76004, 76005, 76056, 76059, 76066, 76074, 78123, 78127, 78128.

**Literature:** as *Navicula contenta*: KRAMMER & LANGE–BERTALOT (1986); as *Diadesmis contenta*: MOSER et al. (1998), WERUM et al. (2004), HOFMANN et al. (2013); as *Humidophila contenta*: LOWE et al. (2014).

**Remarks:** *Humidophila contenta* was characterized by SCHOEMAN & ARCHIBALD (1978) as an extremely

polymorphic taxon. KRAMMER & LANGE–BERTALOT (1986) have followed this opinion. However, MOSER et al. (1998) came to a contrary result after SEM–investigations, concluding that various populations which seemed to belong to *H. contenta* at a first glance, in reality belong to other species. *Humidophila contenta* is a very distinct cosmopolitan species that does not change its pattern of fine structures significantly (LANGE–BERTALOT & WERUM 2001).

VAN DAM et al. (1994) and VAN DE VIJVER et al. (2002) stated that *H. contenta* was often found in alkaline, oligotrophic to eutrophic freshwater, mainly occurring on wet and moist or temporarily dry places, semi–wet to dry environments, and terrestrial mosses.

In Brazil, *H. contenta* is the most frequently registered species of *Humidophila*, being cited in almost all works that include species of the genus.

***Humidophila nienta* (Carter) Lowe, Kociolek, Johansen, Van de Vijver, Lange–Bertalot et Kopalová 2014 (Figs 55–85)**

Lowe et al. (2014): Diatom Research, v. 29, n. 4, p. 351–360.

**Description (n=32)**

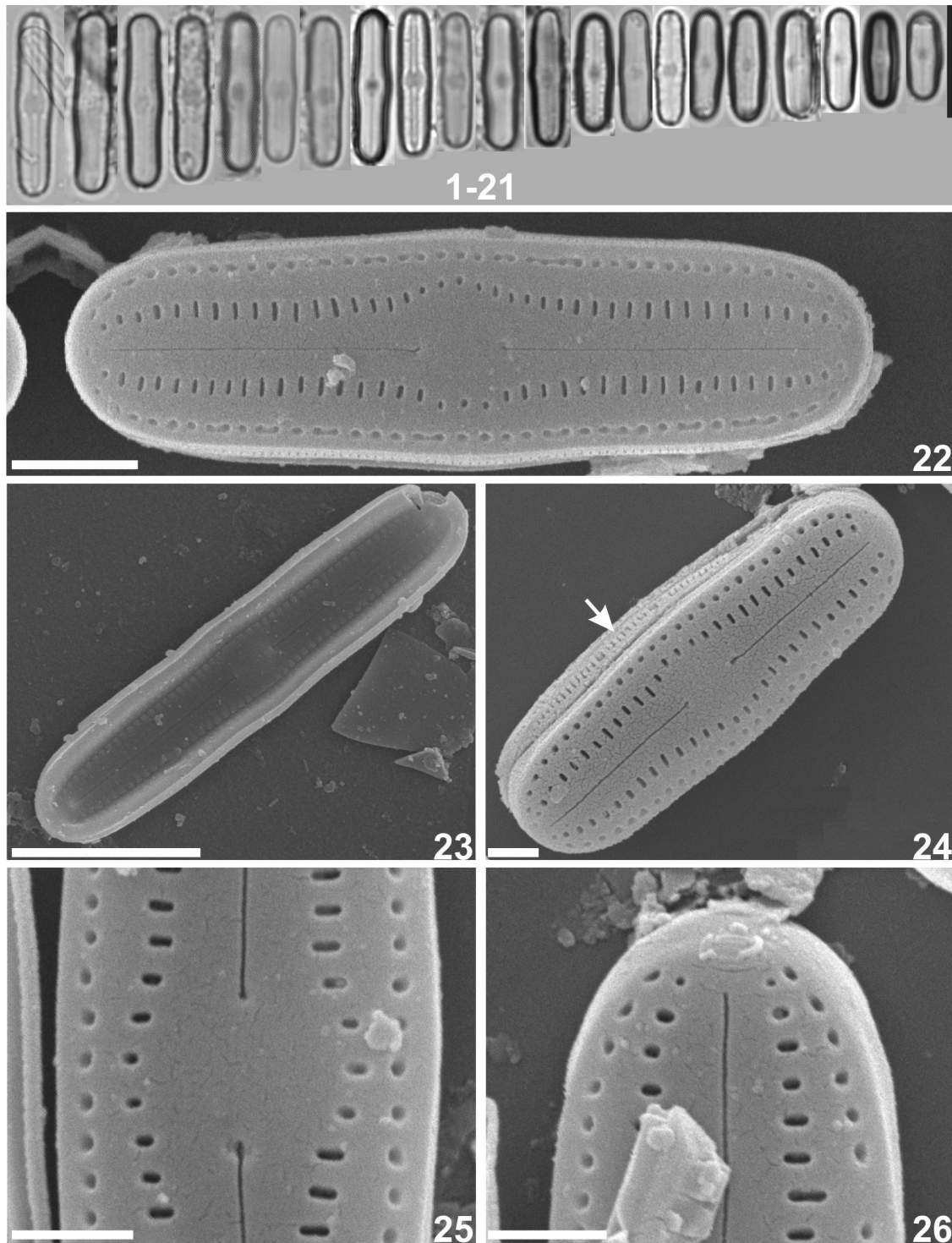
**LM (Figs 55–80):** Valves linear, with undulate margins, slightly constricted in the center, length 9.2–16.3 µm, width 2.6–3.1 µm, with subcapitate apices. Axial area linear to weakly lanceolate. Central area circular.

**SEM (Figs 81–85):** Externally, raphe filiform with central and distal ends T-shaped. Axial area linear to weakly lanceolate. Central area circular, enlarged due to shortening of the central striae. Striae parallel, 35–40 in 10 µm, formed by a single transapically elongated

Table 1: Data of samples in which *Humidophila* species was found.

UPCB	Sampling date	pH	Conductivity (µS.cm <sup>-1</sup> )	Temperature (°C)	Altitude (m)	Sample	Habitat	Coordinates
76004	09/29/2013	7.2	14.5	13.5	998	Epiphytic	Bryophyte	25°29'47" S 48°58'53" O
76005	09/29/2013	7.2	13.4	11.6	1055	Epiphytic	Pebble	25°29'37" S 48°58'41" W
76056	04/30/2014	6.7	14.3	14.8	895	Epiphytic	Moss	25°29'36.8" S 8°58'41.4" W
76059	04/30/2014	6.5	14.3	14.8	1001	Epiphytic	Moss	25°30'19" S 48°58'57" W
76066	04/30/2014	7.1	17.4	15.8	–	Epiphytic	Sand	25°29'13" S 48°59'34" W
76074	04/30/2014	6.3	–	–	–	Epiphytic	Moss	25°29'42" S 48°59'08" W
78123	10/24/2014	6.8	18.3	16.2	1065	Phyto-plankton	Phytoplankton	25°29'41.7" S 8°58'54.3" W
78127	10/24/2014	6.7	20.65	16.1	1050	Epiphytic	Bryophyte	25°29'41.7" S 8°58'54.3" W
78128	10/24/2014	6.7	20.65	16.1	1050	Epiphytic	wet walls	25°29'41.7" S 8°58'54.3" W





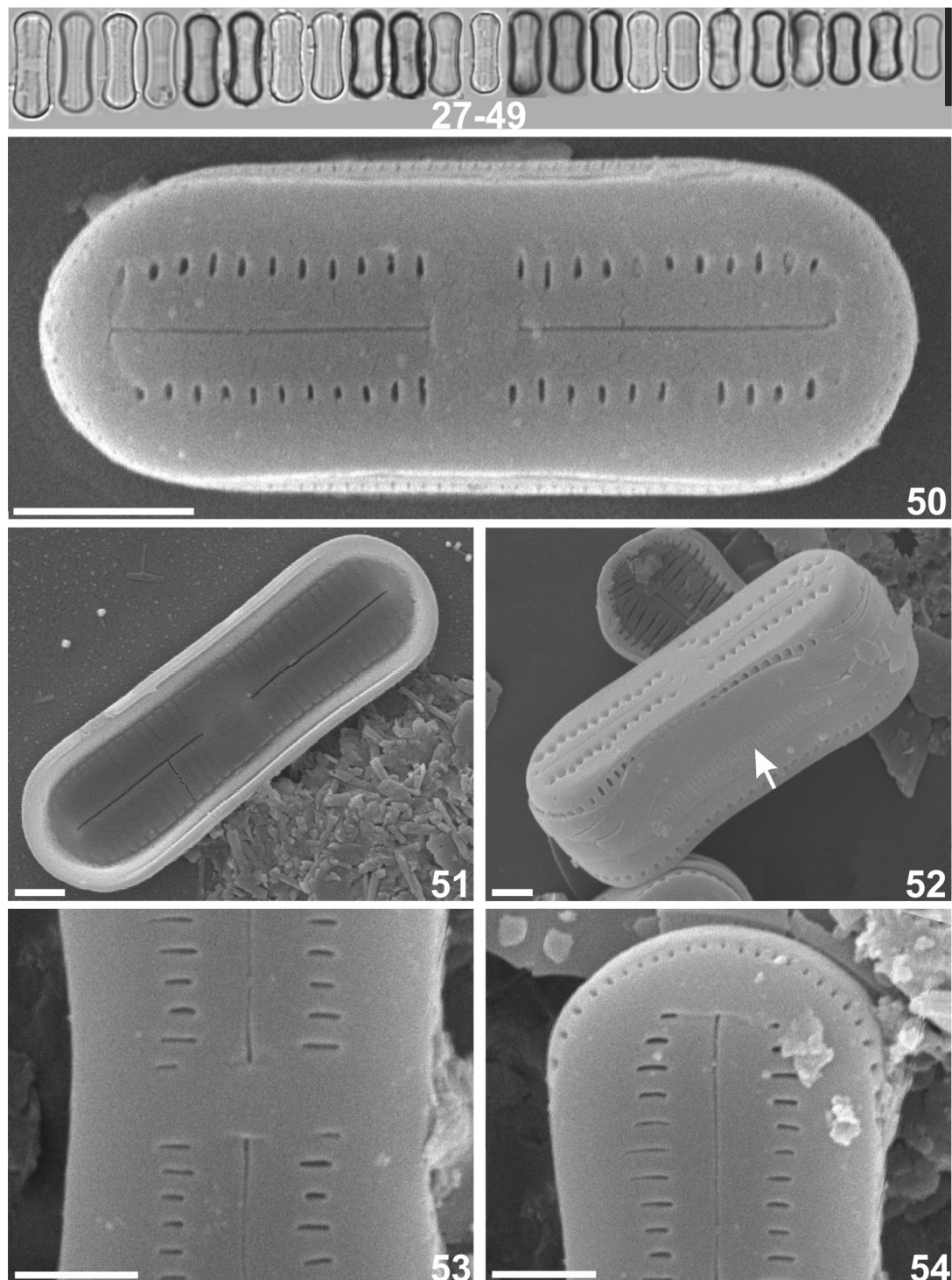
Figs 1–26. *Humidophila brekkaensis*. (1–21) LM valves view, showing morphological variability. (22–26) SEM: (22) external view of the valve; (23) internal view of the valve; (24) lateral valve view showing the row of elongated mantle areolae, interrupted around the apices, and copulae perforated by elongated slits (arrow); (25) external valve detail of the central region, raphe ends straight; (26) external view, detail of the apice, raphe ends straight. Scale bars 10  $\mu\text{m}$  (1–21), 2  $\mu\text{m}$  (22), 5  $\mu\text{m}$  (23) and 1  $\mu\text{m}$  (24–26).

areola. Mantle areolae rounded, 40 in 10  $\mu\text{m}$ , uninterrupted around the valve apices. Copulae perforated by elongated slits. Internally, axial area linear to weakly lanceolate. Central area circular, enlarged due to shortening of the central striae. Presence of pseudoseptum at both apices.

**Samples occurrence:** UPCB–78123, 78127, 78128.

**Literature:** as *Diadesmis langebertalotii*: LE COHU & VAN DE VIJVER (2002); as *Diadesmis pseudolangebertalotii*: METZELTIN et al. (2005); as *Humidophila nienta*: LOWE et al. (2014).

**Remarks:** *Humidophila nienta* was described

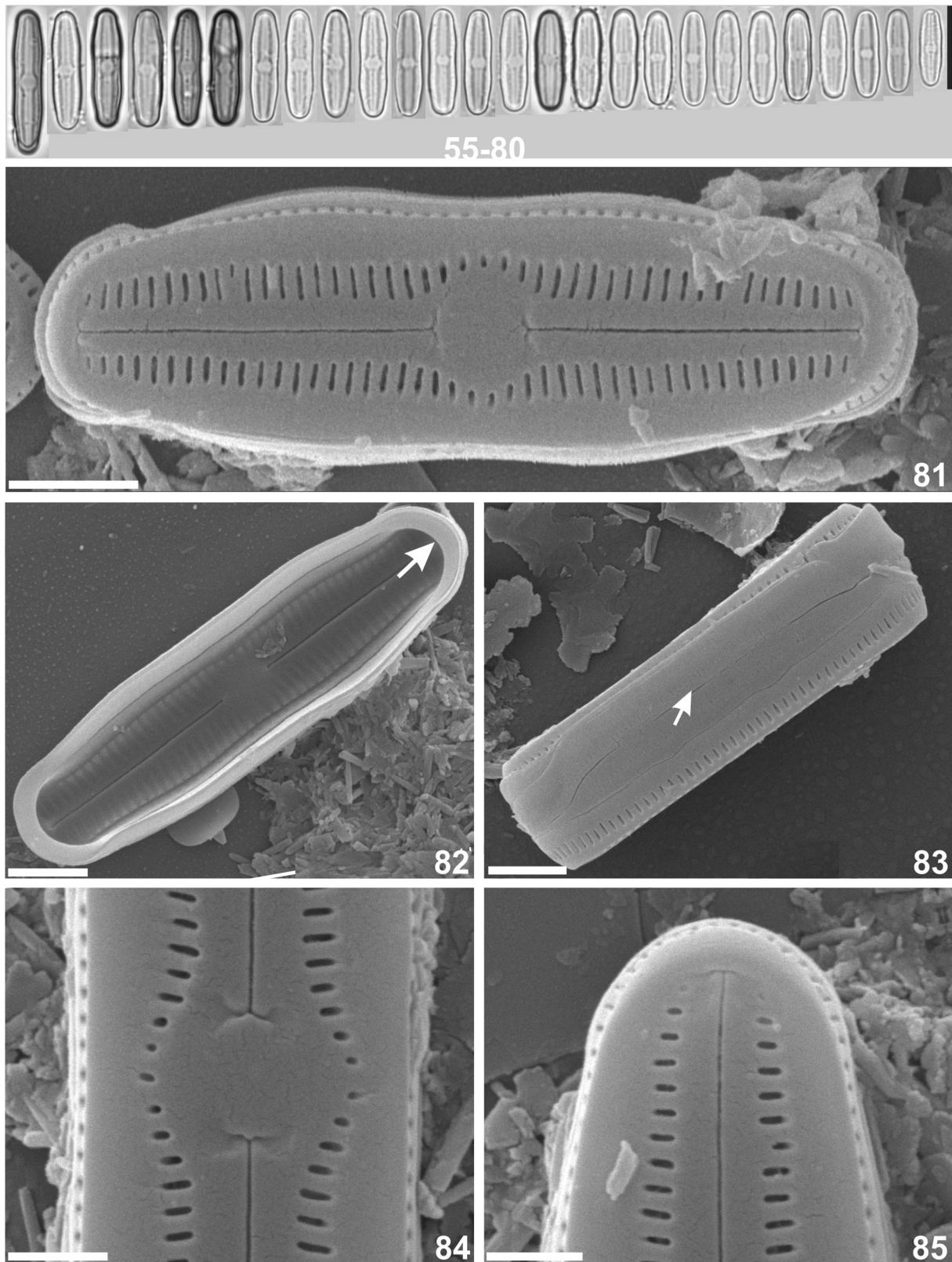


Figs 27–54. *Humidophila contenta*. (27–49) LM valves view, showing morphological variability. (50–54) SEM: (50) external view of the valve; (51) internal view of the valve; (52) lateral valve view showing the row of elongated mantle areolae, uninterrupted around the apices, and copulae perforated by delicate elongated slits (arrow); (53) external valve detail of the central region, raphe ends laterally expanded in T-shaped; (54) external view, detail of the apice, raphe ends laterally expanded in T-shaped. Scale bars 10µm (27–49), 2µm (50) and 1µm (51–54).

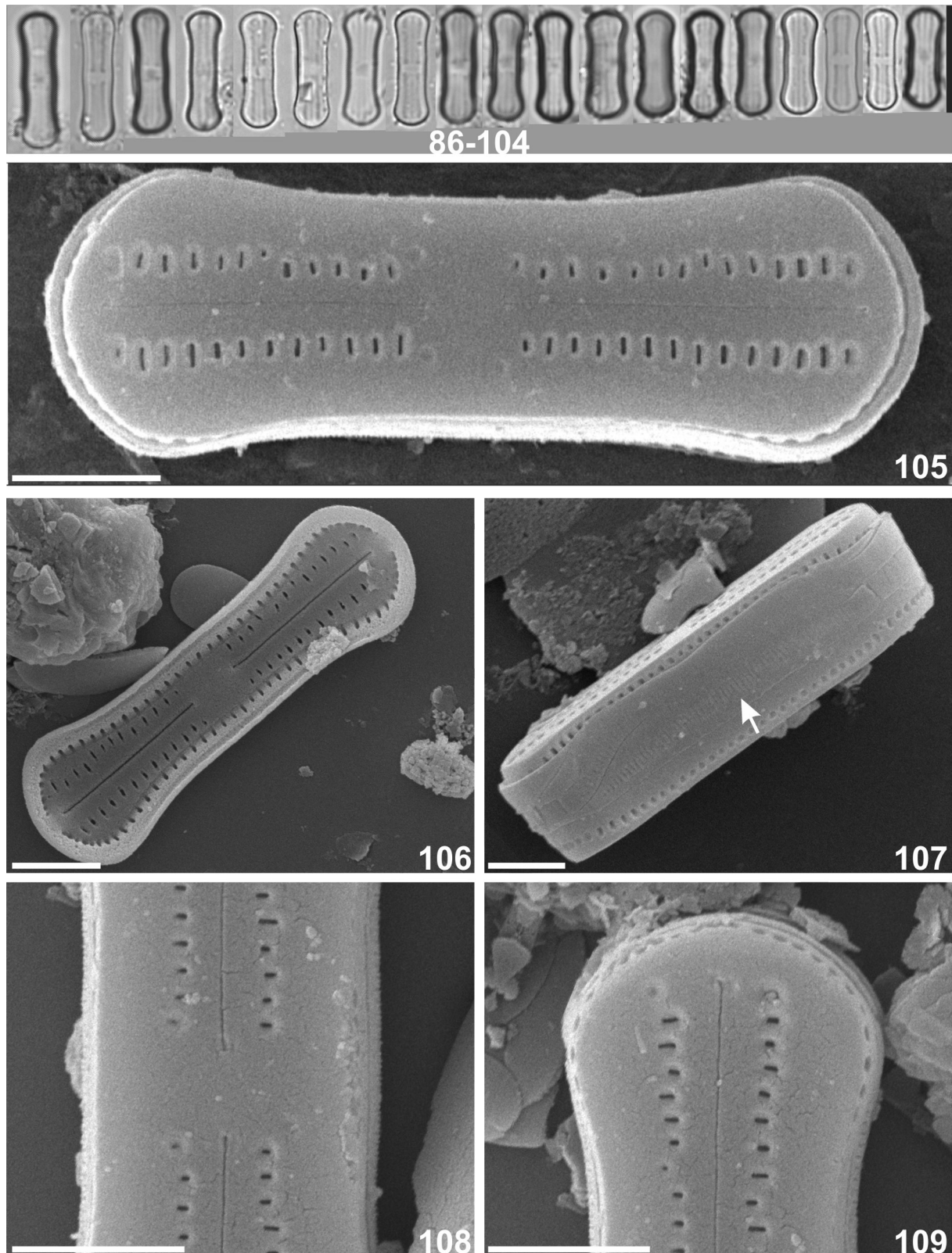
originally by CARTER (1966) as *Navicula nienta*, to the archipelago of Tristan da Cunha. LOWE et al. (2014) analyzed the ultrastructure of specimens of *H. nienta* in samples from the Kerguelen Islands, in Hawaii. The authors discussed the similarity with *D. langebertalotii* and with *D. pseudolangebertalotii* and synonymized both with *H. nienta*.

CARTER (1966) and LOWE et al. (2014) stated that *H. nienta* was often found on rocky substrates in streams, walls of dimly lit caves and lava tubes. In fact, there were no records of *H. nienta* in Brazil. However, COSTA (1995) illustrated a heterogeneous population of *Diadesmis contenta*, and we believe that the individual of fig. 184 corresponds to *H. nienta*. SANTOS et al. (2011)



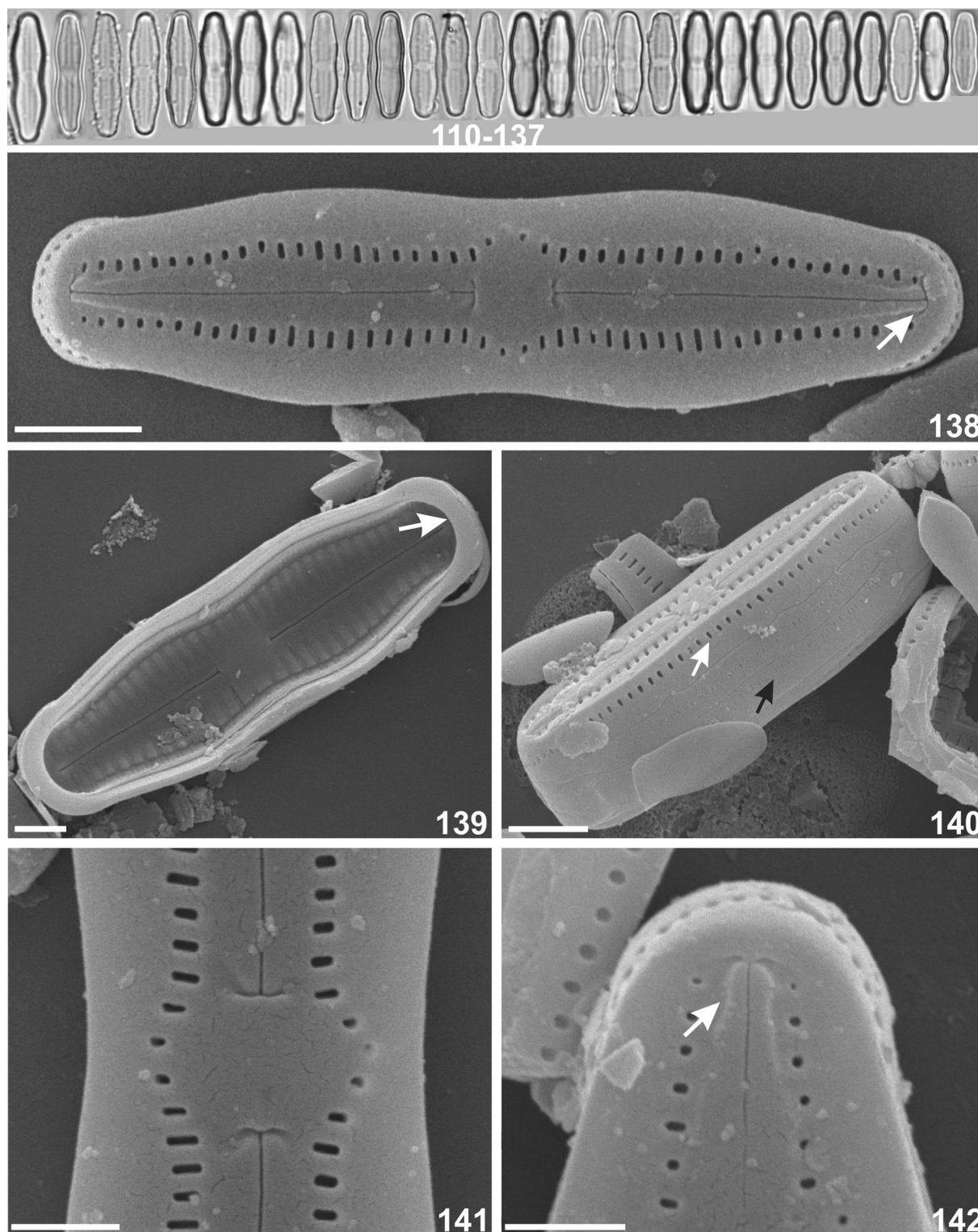


Figs 55–85. *Humidophila nienta*. (55–80) LM valves view, showing morphological variability. (81–85) SEM: (81) external view of the valve; (82) internal view of the valve, pseudoseptum (arrow); (83) lateral valve view showing the row of elongated mantle areolae, uninterrupted around the apices, and copulae perforated by delicate elongated slits (arrow); (84) external valve detail of the central region, raphe ends laterally expanded in anchor-shaped depressions; (85) external view, detail of the apice, raphe ends laterally expanded in anchor-shaped depressions. Scale bars 10µm (55–80), 2µm (81–83, 85) and 1µm (84).



Figs 86–109. *Humidophila paracontenta*. (86–104) LM valves view, showing morphological variability. (105–109) SEM: (105) external view of the valve; (106) internal view of the valve; (107) lateral valve view showing the row of elongated mantle areolae, uninterrupted around the apices, and copulae perforated by elongated slits (arrow); (108) external valve detail of the central region, raphe ends straight; (109) external view, detail of the apex, raphe ends in small pore-shaped. Scale bars 10  $\mu$ m (86–104) and 2  $\mu$ m (105–109).





Figs 110–142. *Humidophila piraquarae* P.C. Ferreira et T.V. Ludwig sp. nov., (110–137) LM valves view, showing morphological variability: (110) holotype specimen. (138–142) SEM: (138) external view of the valve, indicating raised longitudinal siliceous thickening (arrow); (139) internal valve view, pseudoseptum (arrow); (140) lateral valve view, row of elongated areolae in the mantle (white arrow) uninterrupted around the apices, copulae perforated by delicate slits (black arrow); (141) external view, detail of the central region, raphe endings laterally expanded in anchor-shaped depressions; (142) external view, apical raised longitudinal siliceous thickening (arrow). Scale bars 10  $\mu$ m (109–136), 2  $\mu$ m (137, 139) and 1  $\mu$ m (138, 140–142).



documented a species misidentified as *Diademsis im-plicata* Moser, Lange–Bertalot et Metzeltin, that, in our opinion, actually corresponds to *H. nienta*.

***Humidophila paracontenta* (Lange–Bertalot et Werum) Lowe, Kociolek, Johansen, Van de Vijver, Lange–Bertalot et Kopalová 2014 (Figs 86–109)**

Lowe et al. (2014): Diatom Research, v. 29, n. 4, p. 351–360.

**Description (n=30)**

**LM (Figs 86–104):** Valves linear, slightly gibbous in the center, length 9.2–12.7 µm, width 2.8–3.1 µm, with widely rounded apices. Axial area linear. Central area expanded in fascia, extending from the central nodule to the valve margins.

**SEM (Figs 105–109):** Externally, raphe filiform with external central ends straight and distal ends small pore-shaped. Axial area linear. Central area expanded in fascia, extending from the central nodule to the valve margins. Striae parallel, 30–40 in 10 µm, formed by a single areola. Areolae transapically shortened near the central area. Mantle areolae elongated to rounded, 35 in 10 µm, uninterrupted around the valve apices. Copulae perforated by elongated slits. Internally, axial area linear. Central area expanded in fascia, extending from the central nodule to the valve margins.

**Samples occurrence:** UPCB–76004, 76005, 76056, 76059, 76066, 76074, 78123, 78127, 78128.

**Literature:** as *Diademsis paracontenta*: WERUM et al. (2004); HOFMANN et al. (2013); as *Humidophila paracontenta*: LOWE et al. (2014).

**Remarks:** The present population resembles *Humidophila undulocontenta* (Lange–Bertalot et Werum) Lowe, Kociolek, Johansen, Van de Vijver, Lange–Bertalot et Kopalová. However *H. undulocontenta* has denser striae (40–42 in 10 µm), but lacks the consistent phantom striae around the central area and the marked depressions in the raphe distal end (LOWE et al. 2017).

FUREY et al. (2020) stated that *H. paracontenta* was often found in euaerial or pseudoaerial environments.

In Brazil, there were no records of *H. paracontenta*. However, COSTA (1995) illustrated a heterogeneous population of *Diademsis contenta*, and, in our opinion, the individual of fig. 185 corresponds to *H. paracontenta*. SILVA et al. (2010) illustrate a specimen (fig. 126) identified as *H. contenta* that we believe, corresponds meristically and morphologically to *H. paracontenta*.

***Humidophila piraquarae* P.C. Ferreira et T.V. Ludwig sp. nov. (Figs 110–142)**

**Description (n=30)**

**LM (Figs 110–137):** Valves linear, with prominent undulations, length 8.9–14.7 µm and width 2.6–3.2 µm. Apices broadly rostrate–rounded. Axial area lanceolate, tapering gradually towards the apice. Central

area transapically elliptic to almost circular.

**SEM (Figs 138–142):** Externally, central and distal endings laterally expanded in anchor-shaped depressions. Axial area lanceolate, tapering gradually towards the apice. Raised longitudinal siliceous thickening (Figs 138, 142), restricted to distal region of axial area. Central area transapically elliptic to almost circular, delimited by shorter striae. Striae slightly radiate near the rounded central area, 30–40 in 10 µm, formed by a single areola. Areolae transapically elongated, shorter and more circular near the central area and near the apices. Mantle areolae elongated, 35 in 10 µm, uninterrupted around the valve apices. Copulae perforated by delicate slits (Fig. 140). Internally, axial area lanceolate, tapering gradually towards the apice. Central area transapically elliptic to almost circular, delimited by shorter striae. Presence of pseudoseptum at both apices (Fig. 139).

**Holotype:** UPCB! Naphrax slide UPCB76074, Herbarium of Universidade Federal do Paraná (UPCB), Curitiba municipality, Brazil. Holotype specimen illustrated in Fig. 110.

**Type locality:** Brazil, State of Paraná, Piraquara municipality, Carvalho Reservoir, 25°29'42" S, 48°59'08" W, collected by R. Garbuio et al. on 04/30/2014 (Table 1).

**Habitat:** Aerophytic

**Etymology:** The specific epithet refers to the city of Piraquara, where the species was found.

**Ecology:** The new species occurred in moss found in rocky wall with low pH (6,3). In this sample there were only species of the genus *Humidophila*, in addition to the new species, there were also *H. brekkaensis*, *H. contenta* and *H. paracontenta*.

**Distribution:** The species has been found so far only in the type locality.

**Remarks:** The undulated outline is not a common feature in the genus *Humidophila*, only detected in some species, such as *H. biggiba* (Hustedt) Lowe, Kociolek, Johansen, Van de Vijver, Lange–Bertalot et Kopalová, *H. nienta* and *H. undulata* Lowe, Kociolek et Johansen. *Humidophila piraquarae* resembles *H. nienta* and *H. undulata* (Table S1). *Humidophila nienta* differs by its undulated valve outline less constricted in the middle, and distal raphe endings often with rim-shaped depressions (LOWE et al. 2014). *Humidophila undulata* differs by having a deeper constricted valve, lower striae density (29–31 areolae in 10 µm), weakly rostrate to cuneately–rounded apices, axial area linear not raised near distal raphe ends, and central raphe endings terminating in laterally expanded T-shaped depressions (LOWE et al. 2014).

We propose the new species, *Humidophila piraquarae* P.C. Ferreira et T.V. Ludwig, based on the unique features of our population comparing to similar species (Table S2): prominent constriction, broadly

rostrate–rounded, striae density, raised siliceous thickening region of axial area, and raphe central endings laterally expanded in anchor–shaped depressions. The new species was found in mosses in a well–preserved environment.

## DISCUSSION

Recent discoveries of new diatoms in aerial and subaerial environments, such as *Humidophila* (KOCHMAN–Kędziora et al. 2016; FUREY et al. 2020) and *Luticola* D.G. Mann (KALE et al. 2017; FUREY et al. 2020), highlight the great importance of taxonomic studies including collections in this type of habitat. Many *Humidophila* taxa occur in moist aerial or subaerial habitats, wet or semi–dry terrestrial habitats such as wet soils, wet walls, on tree trunks, moist mosses, damp rocks, and spray zones of waterfalls (LOWE et al. 2014; TAYLOR & COCQUYT 2016; LOWE et al. 2017; CHATTOVÁ et al. 2018). The populations studied in “Mananciais da Serra” were found in sub–aerial environments such as mosses and in aerial environments such as walls moistened by water spray, typical environments of the genus *Humidophila*. However, we also found specimens attached to submerged substrates such as pebbles and sand, and in a phytoplankton sample. Species were more frequent in wet moss samples. Also, several species of *Humidophila* often occur in oligotrophic environments (WERUM & LANGE–BERTALOT 2004; POULÍČKOVÁ & HAŠLER 2007; TAYLOR & COCQUYT 2016; RYBAK et al. 2018). Our samples were collected in subaerial environments and water bodies inserted in mountainous conservation area with very low anthropic interference. The streams have clear and crystalline water, low temperature, low concentration of nutrients, and acidic pH to be usual for this type of mountainous environment (TUNDISI & MATSUMURA–TUNDISI 2008). *Humidophila* species had been recorded in Brazilian environments with most diverse levels of trophy (SILVA et al. 2010; MARRA et al. 2016; SILVA–LEHMKUHL et al. 2019). Greater numbers of species were found in oligo–mesotrophic environments (BERTOLLI et al. 2010; MARRA et al. 2016; MARQUARDT et al. 2018; SILVA–LEHMKUHL et al. 2019). *Humidophila* taxa in environments with different levels of trophy probably occur, because cosmopolitan *Humidophila* species, such as *H. contenta*, can adapt easily to rapid changes in environmental conditions (NIKOLIC et al. 2020).

*Humidophila contenta* is frequently documented in Brazil in different habitats, mostly sub–aerial environments, and mainly attached to macrophytes or damp rocks (MARRA et al. 2016; SILVA–LEHMKUHL et al. 2019; BES et al. 2012; HEINRICH et al. 2014; SILVA et al. 2011; SILVA et al. 2017), phytoplankton samples (BARTOZEK et al. 2013), samples of submerged pebbles (MORESCO & RODRIGUES 2016) or artificial substrates (OLIVEIRA et al. 2002; SILVA–LEHMKUHL et al. 2019).

Although 65 have been flagged as taxonomically accepted (GUIRY & GUIRY 2021), only 12 species of *Humidophila* were registered in Brazil. Some are identified incorrectly. Partly due to the use of traditional European or North American identification guides (TYLER 1996; CHATTOVÁ et al. 2018), to erroneous identifications (VAN DE VIJVER & COCQUYT 2009) or absence of ultrastructure observations. In addition, some species did not have a clear delimitation, presenting very heterogeneous populations, as is the case of *H. contenta* (KRAMMER & LANGE–BERTALOT 1986). In recent years, studies focusing on the genus allowed to optimize the taxonomic delimitation of some species with heterogeneous populations, through the description of independent species based on current morphological concepts and biogeography (LOWE et al. 2014; KOPALOVÁ et al. 2015; LOWE et al. 2017; CHATTOVÁ et al. 2018).

The distribution of the genera in the country is restricted to the states of Goiás, Rio de Janeiro, Paraná, São Paulo and Rio Grande do Sul. This restricted distribution and probably the number of species are underestimated in Brazil. More extensive sampling covering uncollected regions and diverse aerophilic habitats would reveal the real diversity of the genus in Brazil.

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#### Supplementary material

The following supplementary material is available for this article:

Table S1. Occurrence of *Humidophila* in Brazil.  
Table S2. Comparison of *Humidophila piraquarae* P.C. Ferreira et T.V. Ludwig sp. nov. with similar species.

This material is available as part of the online article (<http://fottea.czechphycology.cz/contents>)