

Nupela semifasciata (Bacillariophyceae), a new species from subtropical lotic environments in Western Paraná State, Brazil

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Abstract: During a survey on freshwater epilithic diatoms from subtropical lotic environments belonging to the Cascavel River microbasin, Paraná State, Brazil, we observed populations of a new *Nupela* Vyverman et Compère species. Morphological and meristic analyses were performed using light and scanning electron microscopy, resulting in the description of *Nupela semifasciata* sp. nov. This species is characterized by elliptic–lanceolate valves containing subcapitate apices in larger individuals and broadly rostrate–rounded apices in smaller ones, while the length/width ratio gradually decreases as the individuals become smaller. The central area unilaterally reaches the margin and always interrupts the row of areolae on the valve mantle. The valve mantle is externally composed of a second row of areolae near the apices. We cross-checked information with similar *Nupela* taxa, highlighting the main features that separate them. *Nupela semifasciata* was found in streams with acidic–neutral pH, low conductivity and low to high nutrient concentrations.

Key words: diatoms, morphology, new taxa, *Nupela*, periphyton, taxonomy

INTRODUCTION

One of the major obstacles in diatom taxonomy is that light microscopy is often insufficient for species level identification (POTAPOVA et al. 2003). This process is particularly accentuated in small-sized diatoms, such as *Nupela*, in which the individuals generally measure less than 20 µm (SPAULDING & EDLUND 2008). Ultrastructural analysis of frustules obtained by electron microscopy provides observation of additional features that improves accurate species diagnosis (ZIMMERMANN et al. 2014) and, thus, supports taxonomic robustness for biodiversity conservation purposes (THOMSON et al. 2018).

Nupela Vyverman et Compère was proposed as a distinct genus based on samples collected in high elevation ponds with a peaty bottom in Papua New Guinea, with *N. giluensis* as the generitype. The authors pointed out morphological similarities with *Brachysira* Kützinger and *Diadensis* Kützinger, such as striae composed of a few areolae transversally

elongated, a ridge or a hyaline area at the junction of the valve face and mantle, and simple or inconspicuous proximal raphe fissures (VYVERMAN & COMPÈRE 1991). However, recent phylogenetic investigations suggested that *Nupela* and *Brachysira* form a non-monophyletic group, with *Nupela* not being assigned to the family Brachysiraceae (KULIKOVSKIY et al. 2020). In addition, SIVER et al. (2007) expanded the circumscription of this genus to comprise cells with small size, terminal raphe ends externally curved to the secondary side, straight proximal raphe ends, Voigt faults indicating the secondary side of the valve, and a single row of elongated areolae throughout the valve mantle. The last feature appears to be well-established within the genus (VYVERMAN & COMPÈRE 1991; SIVER et al. 2007; FALASCO et al. 2015), along with the typical external openings of areolae covered by hymen that are larger than internal ones (SIVER et al. 2007; SPAULDING & EDLUND 2008). Regarding the raphe, *Nupela* possesses isovalvar forms with two fully developed raphe slits, such as in *N. giluensis* (VYVERMAN & COMPÈRE

1991) and *N. amabilis* (TREMARIN et al. 2015); however, the vast majority of species are heterovalvar with one valve containing shortened raphe slits, as in *N. pardinhoensis* (BES et al. 2012), rudimentary slits, as in *N. scissura* (SIVER et al. 2007) and *N. rumrichorum* (LANGE-BERTALOT & MOSER 1994), or absent raphe slits as in *N. praecipuoides* (TREMARIN et al. 2015) and *N. major* (YU et al. 2017).

Since it was established, several species have been transferred from other genera to *Nupela*, mainly *Achnanthes* Bory, due to its monoraphid condition, and *Navicula* Bory, due to the areolae openings shape, which are externally elongated and internally rounded (see LANGE-BERTALOT & MOSER 1994; POTAPOVA 2013; TREMARIN et al. 2015). The number of new *Nupela* taxa is still increasing, consisting of 85 species taxonomically accepted for science, according to Algaebase (GUIRY & GUIRY 2020).

The genus generally occurs in oligotrophic waters with acidic to circumneutral pH (e.g., MONNIER et al. 2003; POTAPOVA et al. 2003; WOJTAŁ 2009; KULIKOVSKIY et al. 2010), being particularly well represented in the Neotropics (METZELTIN & LANGE-BERTALOT 1998; WOJTAŁ 2009). At present, seven taxa were described as new species for science based on Brazilian samples. One of them, *N. pardinhoensis* Bes, Torgan et Ector was found in slow moving waters with high dissolved oxygen, low biochemical oxygen demand, and high total phosphate concentration in the State of Rio Grande do Sul (BES et al. 2012) and the other six, *N. amabilis* Tremarin et Ludwig, *N. difficilis* Straube, Tremarin et Ludwig, *N. kocioleckii* Straube, Tremarin et Ludwig, *N. metzeltinii* Tremarin et Ludwig, *N. praecipuoides* Tremarin et Ludwig and *N. torganiae* Tremarin et Ludwig, were generally found in rivers with high water speed, low conductivity, and neutral

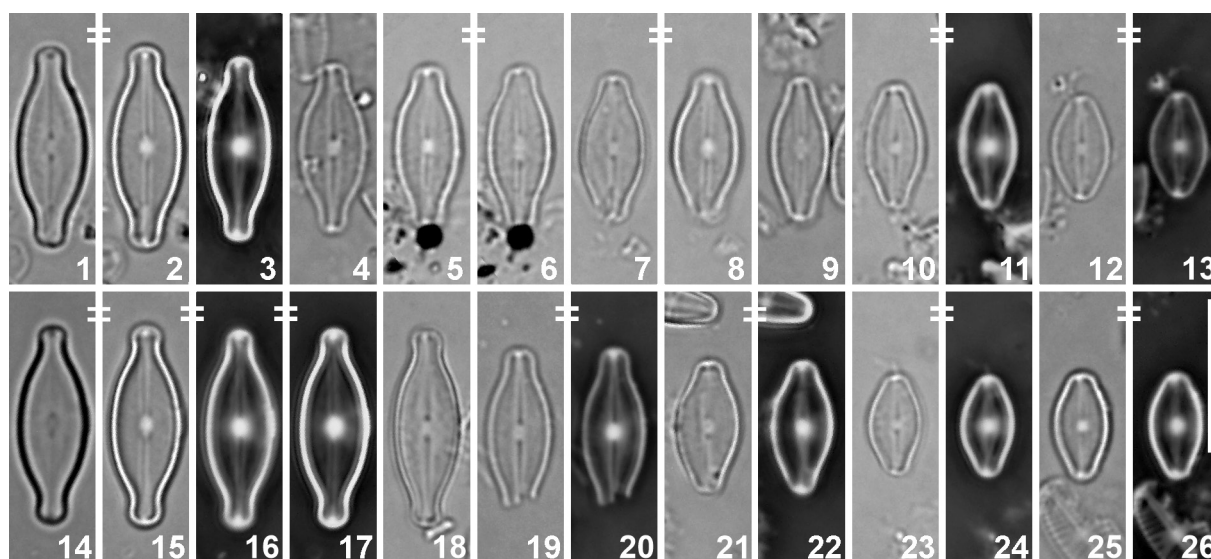
pH in Paraná State (TREMARIN et al. 2015).

The aim of this study was to describe and illustrate a new species of *Nupela* using light and scanning electron microscopy observations, and compare it with similar taxa, thus, expanding the knowledge on the genus.

MATERIAL AND METHODS

Study area. The Cascavel River (24°32' and 25°17'S; 53°05' and 53°50'W) is 17.5 km long and represents the principal source of water supply for Cascavel City, with the main tributaries within the urban perimeter (FUNDETEC 1995). The river is located in Western Paraná, a subtropical region characterized by hot and humid summers, with an average annual temperature of 22 °C (ALVARES et al. 2013).

For the study, we selected eight sampling sites (S) along the Cascavel River microbasin, as follows: S1 – urban perimeter, inside the conservation area of Paulo Gorski Ecological Park, with presence of *Hydrochoerus hydrochaeris* Linnaeus populations. Stream with visible silting up process. S2 – urban area, close to a highway, containing ciliary vegetation. S3 – urban area, close to a highway, lacking ciliary vegetation and visible silting up process. S4 – urban area, close to a garbage collection property, containing sparse ciliary vegetation and receiving water from storm drains. Presence of a bridge over the stream for pedestrian traffic. S5 – rural area, close to temporary croplands and a highway, containing ciliary vegetation. Presence of a bridge over the stream for pedestrian traffic. S6 – conservation area within a rural property, with goat farming, *Pinus* sp. plantation and temporary croplands. S7 – urban area, close to a deactivated fridge, containing ciliary vegetation. Stream with visible silting up process. S8 – rural area with temporary croplands, close to a basalt mining company using mineral deposits. Presence of a bridge over the stream for vehicle traffic.



Figs 1–26. *Nupela semifasciata* sp. nov., light microscopy images showing size diminution series and the variability of the valve outline: (1, 2) are the holotype, (3, 11, 13, 16, 17, 20, 22, 24, 26) were taken in phase contrast. The = symbol represents different valves of the same frustule. Scale bar represents 10 µm.

Table 1. Range of physical, chemical, and biological variables of the water in which *Nupela semifasciata* was found. Legend: (Temp) water temperature, (Cond) electrical conductivity, (DO) dissolved oxygen, (Turb) turbidity, (COD) chemical oxygen demand, (BOD) biochemical oxygen demand, (TKN) Kjeldahl nitrogen, (NO₃) nitrate, (N–NH₃) ammoniacal nitrogen, (TP) total phosphorus, (PO₄[–]) orthophosphate, (TS) total solids, (DS) dissolved solids, (SS) suspended solids, (CLa) chlorophyll-a, (TC) total coliforms, (*E. coli*) *Escherichia coli*.

Variables	Range (min – max)
Temp (°C)	16.67–23.70
Cond (mS.cm ^{–1})	0.001–0.091
DO (mg.l ^{–1})	6.53–14.27
pH	5.40–7.42
Turb (NTU)	0.06–41.90
COD (mg.l ^{–1})	5.10–24.32
BOD (mg.l ^{–1})	1.19–9.90
TKN (mg.l ^{–1})	0.00–0.92
NO ₃ (mg.l ^{–1})	0.32–14.00
N–NH ₃ (mg.l ^{–1})	0.003–0.448
TP (mg.l ^{–1})	0.005–0.060
PO ₄ [–] (mg.l ^{–1})	0.002–0.030
TS (mg.l ^{–1})	4.00–81.00
DS (mg.l ^{–1})	3.30–66.70
SS (mg.l ^{–1})	0.20–40.00
CLa (µg.l ^{–1})	0.000–16.382
TC (NMP 100 ml ^{–1})	1–606000
<i>E. coli</i> (NMP 100 ml ^{–1})	1–606000

Field and laboratory procedures. Samplings were performed seasonally from 2016 to 2018, comprising eight samplings. We collected periphytic substrates (stones) in triplicates at each sampling site, totaling 204 qualitative samples. Water samples for physical, chemical, and biological analysis were collected one per sampling site.

The physical and chemical variables, such as water temperature, conductivity, dissolved oxygen, pH, and turbidity were measured in situ with a HORIBA U–5000 multiparameter probe. Additionally, water samples were collected using polyethylene bottles immersed in the surface of the water column, being adequately cooled, and kept in the dark, from which the environmental variables function of chemical oxidation and organic material, concentrations of total Kjeldahl nitrogen, nitrate, ammoniacal nitrogen, total phosphorus, orthophosphate, and total, dissolved, and suspended solids were measured. The biological variables chlorophyll-a, total coliforms, and *Escherichia coli* were also analyzed. All analyses were realized following the Standard Methods (APHA 2012). The flow (m³.s^{–1}) and depth (m) were measured using a ruler, metric tape, and polystyrene floating object, in a transect previously delimited for each stream. The flow was

calculated by multiplying the length of the transect and the average speed resulting from the object's displacement.

Diatoms were scraped off the stones and fixed in Transeau solution 1:1 according to BICUDO & MENEZES (2017). Subsamples of 10 ml were oxidized following the SIMONSEN (1974) technique, modified by MOREIRA-FILHO & VALENTE-MOREIRA (1981). Permanent slides were mounted from the cleaned diatom material using Naphrax® for light microscopy (LM) observations, using an Olympus BX60 microscope with a DP 71 capture camera attached, at 1000×. Cleaned samples were also placed in aluminum stubs, sputter-coated with gold in Balzers Union SCD 030, and examined with JEOL JSM 6360LV scanning electron microscope, operated at 15 kV and 9–10 mm of working distance. Species description follows the terminology of specialized literature.

RESULTS

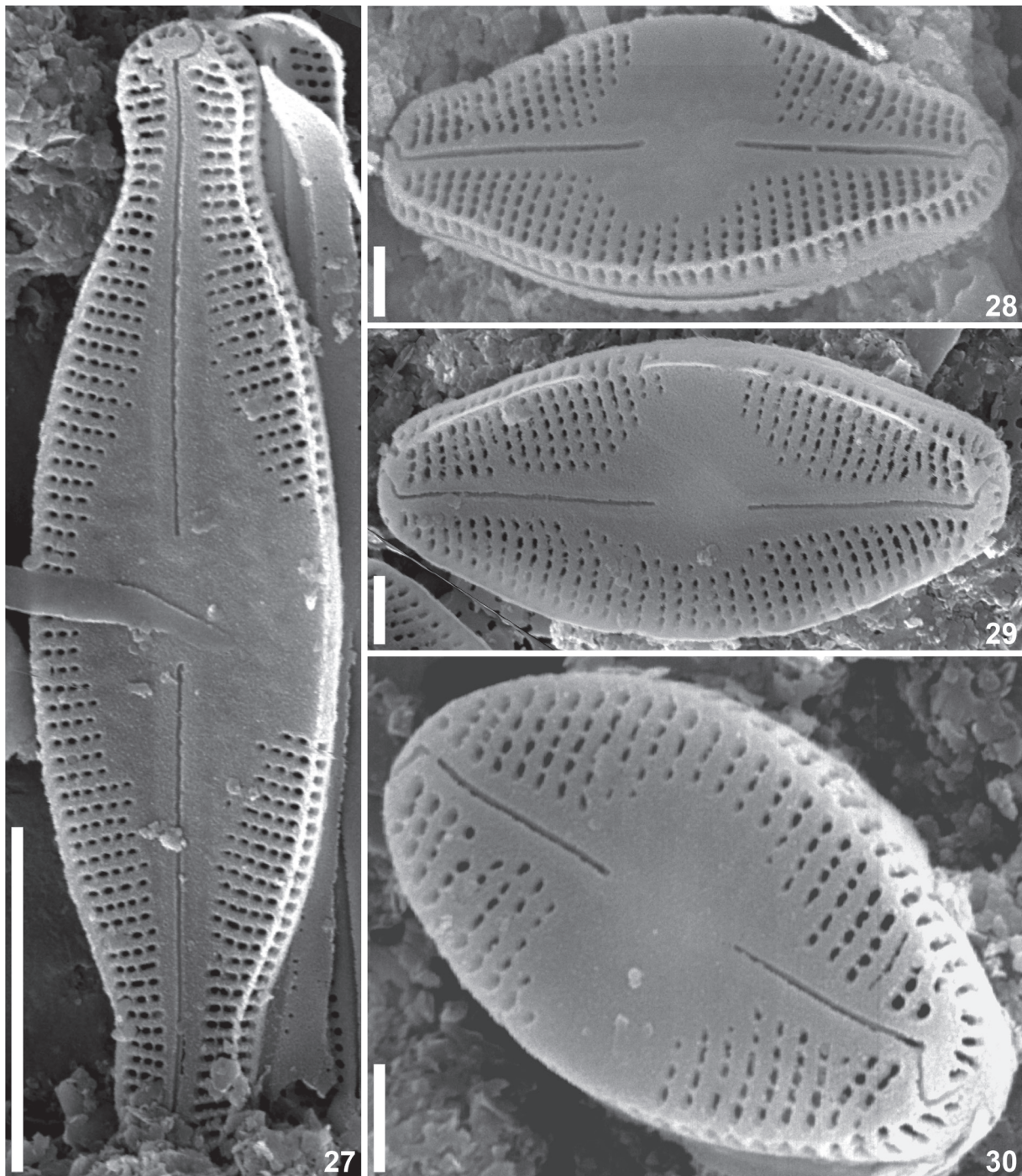
Nupela semifasciata Amaral, T.Ludwig et Bueno sp. nov. (Figs 1–50)

Description

Light microscopy (Figs 1–26): frustules heterovalvar regarding the raphe development, sometimes asymmetrical about apical and transapical planes (Figs 1–26). Length/width ratio gradually decreases as the valves become smaller, while the valve outline is modified. Valves elliptic–lanceolate containing subcapitate apices in larger individuals (Figs 1–3, 14–18) and rostrate in middle ones (Figs 4–11, 19–22), slightly drawn-out, becoming broadly rostrate–rounded in smaller individuals (Figs 12, 13, 23–26). Axial area narrowly lanceolate, slightly broadening toward the central area. Central area transversely expanded, asymmetric, unilaterally reaching the valve margin, discernible only in phase contrast images (Figs 3, 11, 13, 16, 17, 20, 22, 24, 26). One valve with long raphe slits and the other valve with slightly shorter raphe slits (compare Figs 1 to 2, 5 to 6, 7 to 8, 14 to 15).

Measurements (n= 89, occurring in 46 diatom samples): 6.7–16.4 µm long; 3.2–4.8 µm wide; 1.7–3.7 ratio length/width.

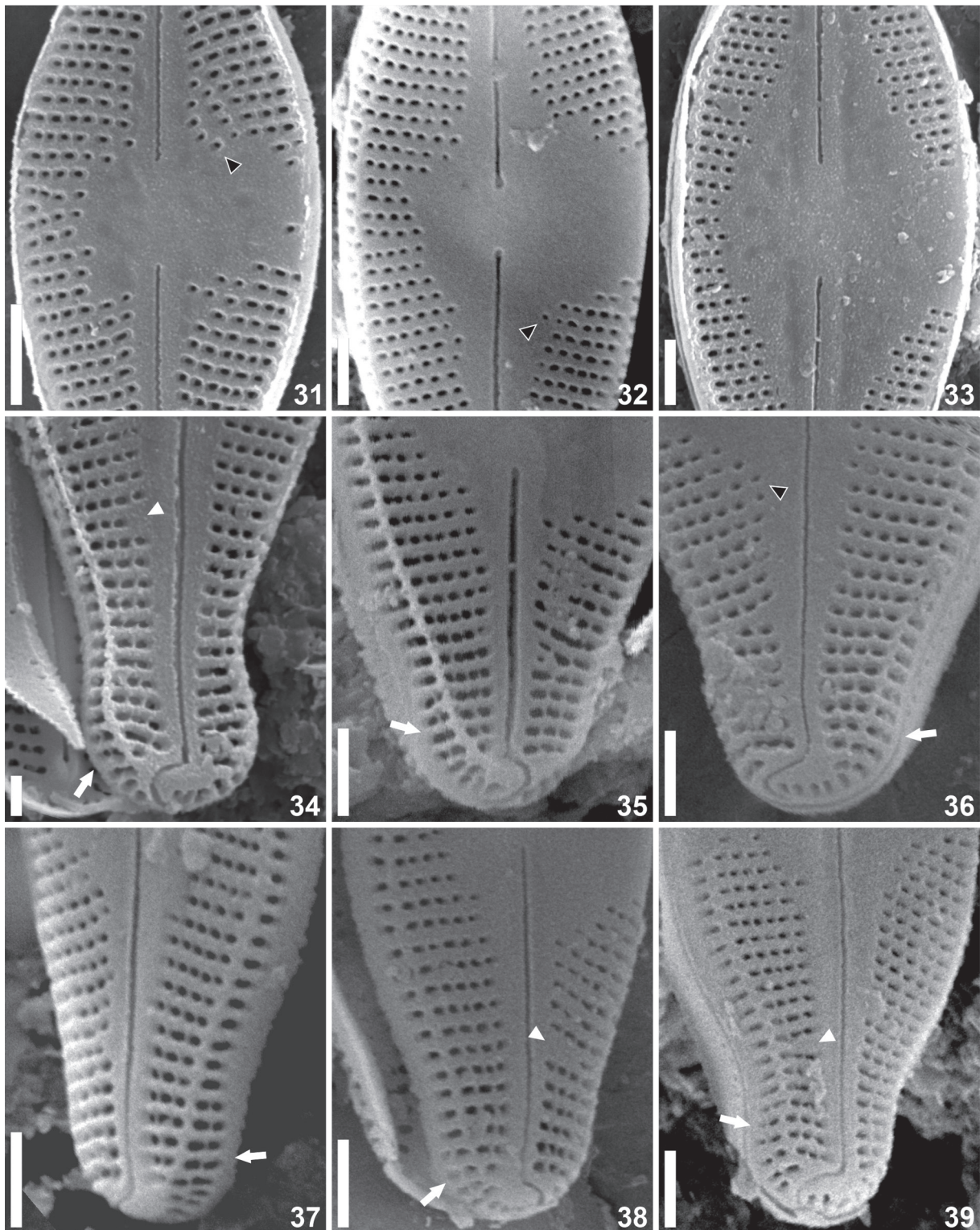
Scanning electron microscopy (Figs 27–39, 48–50 external views, Figs 41–47 internal views): axial area appears narrowly lanceolate (Figs 28–30, 41) or broadly lanceolate (Fig 27), sometimes with external siliceous thickenings surrounding the raphe and delicate depressions along the apical axis (Figs 27, 28, 31, 33, 49, 50). Central area broadly asymmetric, limited by irregular shortened marginal striae, ranging from rectangular (Fig 31) to rounded (Figs 28–30, 32) or rhombic (Figs 27, 33) shape, unilaterally reaching the secondary margin of the valve. The fascia is variable in shape, large or reduced, and always extends onto the valve mantle (Figs 27, 29, 31–33). Sometimes, a single areola appears in the fascia (Figs 31, 41). Valve mantle ornamented with one row of transversely elongated areolae along the valve (Fig 28) and at the end of



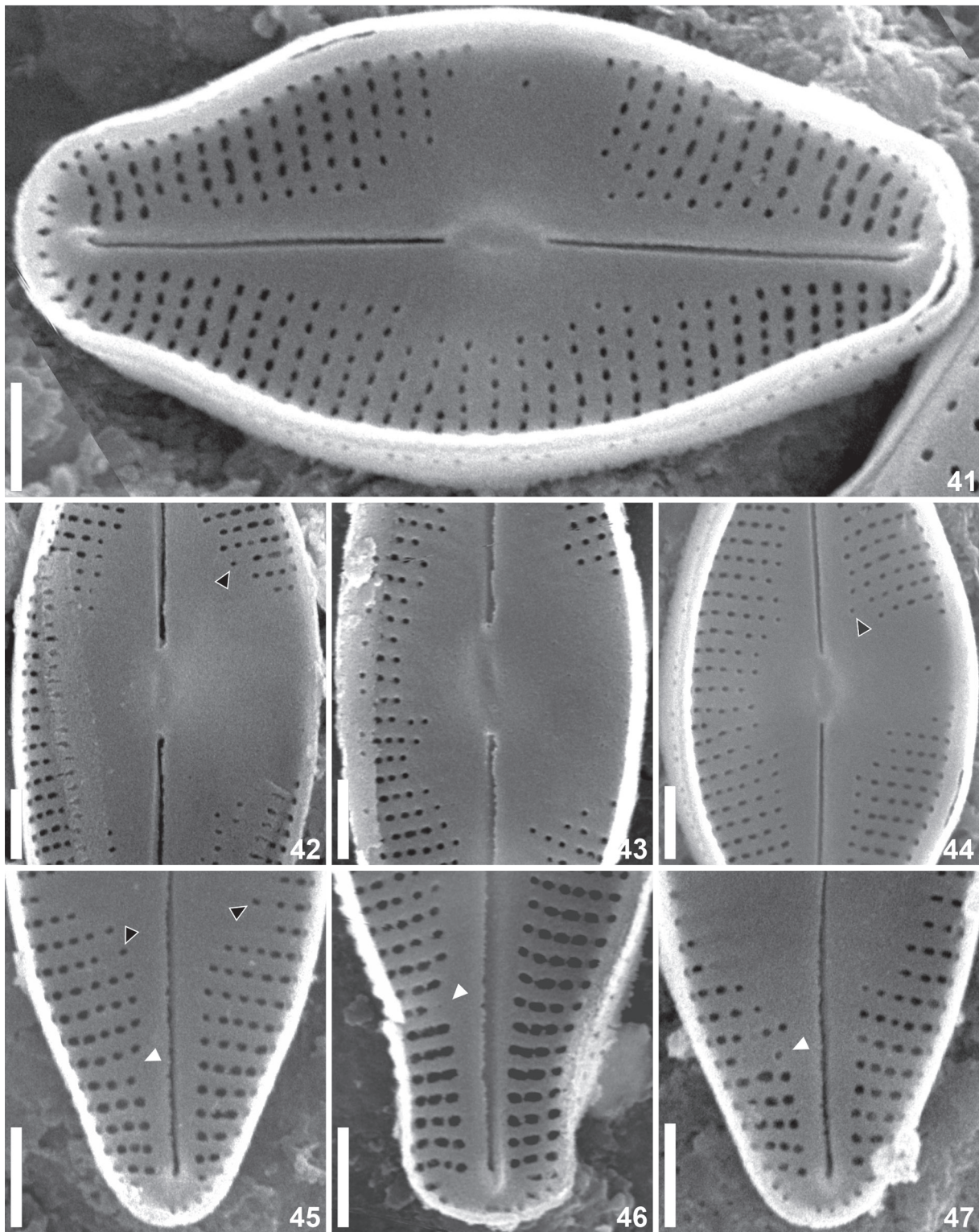
Figs 27–30. *Nupela semifasciata* sp. nov., SEM external whole valve views: (27) entire valve with shorter raphe slits showing the siliceous thickenings surrounding the raphe, delicate depressions along the apical axis, and the rhombic-shaped central area. Also note the ridge along the valve face–mantle junction and the interrupted mantle. (28, 29) entire valves with long raphe slits showing the mantle composed of one row of transversely elongated areolae along the whole valve, becoming two rows near the apices (28), and interrupted by the unilateral fascia (29). Note the asymmetrically rounded central area. (30) entire valve of a smaller individual with shorter raphe slits. Detail of the asymmetrically rounded central area and the hook-shaped terminal raphe fissure extending onto the valve mantle, interrupting the row of elongated areolae. Note the Voigt fault (27–30) on the secondary side of the valve, marked as a change in the striae pattern from radiate to convergent. Scale bars represent 5 μm (27), 1 μm (28–30).

the apex (Figs 30, 36), however, externally becoming two rows of elliptic areolae near the apices (white arrows in figs 34–39). Internally, the mantle appears to be ornamented with only one row of areolae (Figs 41, 47). The mantle is interrupted by the central area on

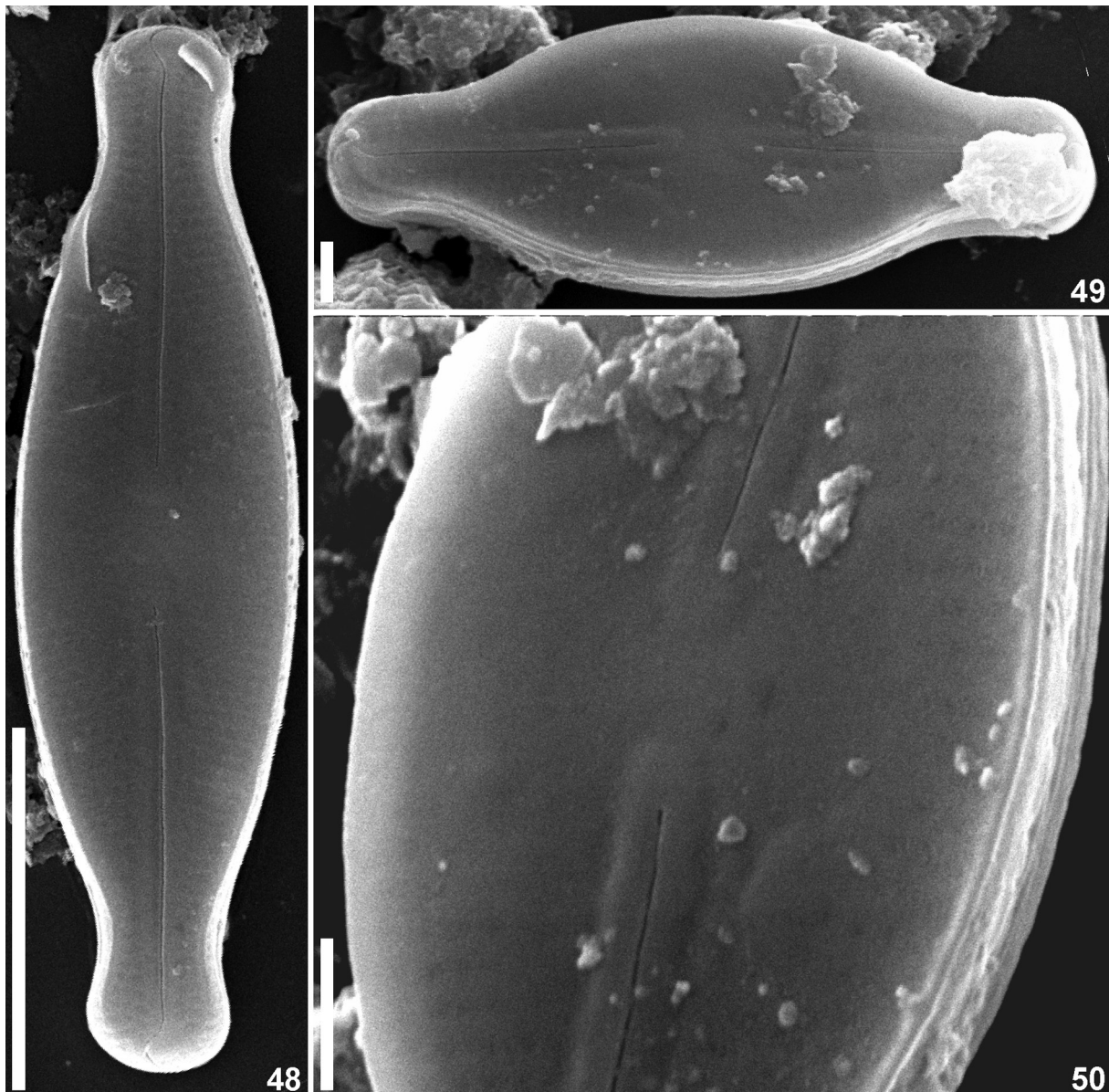
the secondary margin (Figs 27, 29) and by the terminal raphe fissures at the apices (Figs 30, 34–36, 38, 39). In some cases, a ridge is evident on the valve face–mantle junction (Figs 27–29). Raphe filiform, almost straight (Fig 28, 41, 49) or slightly sinuous (Figs 27,



Figs 31–39. *Nupela semifasciata* sp. nov., SEM external views: (31–33) asymmetrically shaped central area, rectangular (31), rounded (32), or rhombic (33). Note the single areola in the middle of the fascia (31). Raphe with long slits (32) or slightly shorter (31, 33). Proximal raphe endings simple (31, 33) or pore-like (32), straight (31) or slightly curved to the secondary side of the valve (32, 33). Note the siliceous thickenings surrounding the raphe and delicate depressions along the apical axis (31, 33). (34–39) valve apices. Second row of areolae on the valve mantle (white arrows in 34–39). White arrowheads indicate Voigt fault as a missing areola (34) or as a marked change in striae pattern (39). Black arrows indicate an isolated areola at the end of striae (31, 32, 36). Scale bars represent 1 μm .



Figs 41–47. *Nupela semifasciata* sp. nov., SEM internal views: (41) whole valve. Axial area narrowly lanceolate, raphe almost straight, and small inner areolae openings. Note the single areola in the middle of the fascia. (42–44) central area. Proximal raphe endings straight (42, 43) or bent (44), simple (44) or pore-like (42, 43). Note the mantle interrupted by the unilaterally expanded fascia. (45–47) valve apices. Terminal raphe ends finishing in small helictoglossa, straight (46) or slightly curved (45, 47). Note the Voigt fault as a marked change in striae pattern (white arrowhead in 45, 47) or like a missing areola (white arrowhead in 46). Also note isolated areola in (42, 44, 45) (black arrowheads). Scale bars represent 1 μ m.



Figs 48–50. *Nupela semifasciata* sp. nov., SEM external views showing areolae occluded by hymenes: (48–49) whole valve views. Raphe filiform, almost straight (49) or slightly sinuous (48), with simple proximal ends, slightly curved to one side, and terminal ends hook-shaped, reaching the mantle. (50) detail of central region of valve. Raphe with simple proximal raphe ends, curved to the same side, and surrounded by siliceous thickenings. Scale bars represent 5 μ m (48), 1 μ m (49, 50).

29, 30, 48). Proximal raphe ends are simple (Figs 31, 33, 44, 48, 49) or pore-like (Figs 32, 42, 43), straight (Figs 31, 42, 43) or somewhat bent to the primary side of the valve (Figs 32, 33, 44, 48, 49). Terminal raphe ends are externally hook-shaped, curved to the secondary side of the valve, reaching the valve mantle (Figs 34–39, 48, 49) and internally finishing in small helicoglossa, straight (Fig 46) or slightly curved (Figs 45, 47). Voigt fault occurs as a markedly changing striae pattern (white arrowheads in figs 38, 39, 45, 47) or seems like an areola is missing (white arrowhead in figs 34, 46). Striae radiate throughout the valve, becoming slightly (Figs 36–39, 45–47) to strongly (Figs 30, 34, 35) convergent near the apices, 40–48/10 μ m. Striae are composed by a variable number of areolae

(commonly 4–6), arranged in continuous lines of areolae, although an isolate areola can be present at the end of some striae (black arrowheads in Figs 31, 32, 36, 42, 44, 45). Areolae small, rounded to transversally elongated in elliptic or rectangular shape (Figs 27–39), internally smaller in diameter relative to outer openings (Fig 41–47), 39–59/10 μ m, commonly ca. 50. Outer openings of areolae are occluded by hymenes (Figs 48–50).

Holotype: slide no. 5364, sample site 4, designated here in figs 1 and 2, deposited in Norma C. Bueno collection at the Herbarium of Western Paraná State University (UNOPA), Cascavel municipality, Brazil.

Type locality: Brazil, State of Paraná, Cascavel municipality, Cascavel River, 24°32' and 25°17'S; 53°05' and

53°50'W, epilithic samples, collected by G. Medeiros et al. on 03/08/2018.

Etymology: the specific epithet refers to the wide central area unilaterally reaching the margin, always interrupting the single row of mantle areolae.

Ecology: *Nupela semifasciata* was found in 46 of 204 epilithic samples analyzed, from which the average values of ecological parameters were calculated, considering the eight samplings performed. The new species occurred in streams with low flow (average of 0.24 m³.s⁻¹), low depth (average of 0.06 m), low conductivity (average of 0.05 mS.cm⁻¹), low to high nutrient concentrations (e.g., NO₃: 2.00–14.00 mg.l⁻¹), and low pH (average of 6.28) (Table 1). Other diatom taxa co-occurred in the samples, such as *Achnantheidium minutissimum* (Kützing) Czarnecki, *Humidophila contenta* (Grunow) Lowe, Kociolek, Johansen, Van de Vijver, Lange-Bertalot et Kopalová, *Sellaphora saugerresii* (Desmazières) Wetzel et Mann, *Sellaphora nigri* (De Notaris) Wetzel et Ector and some other unidentified species of *Eunotia* Ehrenberg and *Nupela* Vyverman et Compère.

DISCUSSION

Nupela semifasciata is characterized by elliptic-lanceolate valves containing subcapitate apices in larger individuals and broadly rostrate-rounded apices in smaller ones, while the length/width ratio gradually decreases as the individuals become smaller. The central area unilaterally reaches the margin and always interrupts the row of areolae on the valve mantle. The valve mantle is externally composed of a second row of areolae near the apices.

The new species shares morphological and morphometric similarities with some currently known species of the genus, but some features can be used to differentiate them (see Table 2). *N. deformis* Lange-Bertalot has deeper depressions along the apical axis, clearly visible in LM images, one valve with rudimentary raphe, and higher striae density (ca. 60 vs 40–48/10 µm) (LANGE-BERTALOT & MOSER 1994). *N. lesothensis* (Schoeman) Lange-Bertalot contains a considerably higher number of areolae in 10 µm (50–68 in SCHOEMAN 1973; 50–74 in SALA et al. 2014; 50–70 in KULIKOVSKIY et al. 2020 vs 39–59 in our study) and one valve with rudimentary raphe slits (SCHOEMAN 1973; SALA et al. 2014). Additionally, the central area does not always reach the valve margin (see SALA et al. 2014). The main features that differentiate *N. paludigena* (Scherer) Lange-Bertalot are the wider and often capitate apices, smaller central area, and proximal raphe ends, externally teardrop-shaped and internally t-shaped (LANGE-BERTALOT & MOSER 1994; SIVER et al. 2007). *N. tenuicephala* (Hustedt) Lange-Bertalot slightly resembles *N. semifasciata*, however,

it has dorsiventral valves with strongly capitate apices, the internal proximal raphe ends are hook-shaped, and the striae are generally higher in number (50–60 vs 40–48/10 µm), composed of wider areolae (LANGE-BERTALOT 1993). Lastly, *N. neotropica* Lange-Bertalot and *N. subpallavicinii* Metzeltin et Lange-Bertalot share the fascia that unilaterally reaches the valve margin, but in both taxa, the internal proximal raphe endings are hook-shaped (LANGE-BERTALOT & MOSER 1994; SALA et al. 2014, respectively). In addition, *N. subpallavicinii* has a radiate striae pattern along the whole valve (METZELTIN & LANGE-BERTALOT 1998), and *N. neotropica* has wider areolae (LANGE-BERTALOT & MOSER 1994).

Nupela wellneri (Lange-Bertalot) Lange-Bertalot, the most similar taxon, was first described in Germany as *Navicula wellneri* by LANGE-BERTALOT & KRAMMER (1987) and was later transferred to *Nupela* based on samples collected in the Andean region by RUMRICH et al. (2000). *Nupela semifasciata* and *Nupela wellneri* possess lanceolate valve outlines, one valve containing long raphe slits and the other valve containing shorter raphe slits, as well as a broad central area, expanded more pronouncedly to one side of the valve. However, in comparison to the protologue, the individuals found in our samples have broader variation in size (length: 6.7–16.4 vs 12–14 µm; width: 3.2–4.8 vs 4–4.8 µm), striae (40–48/10 µm vs ca. 45/10 µm) and areolae densities (39–59/10 µm vs ca. 50/10 µm). Previous studies registered *N. wellneri* with a larger range in size (8.5–15 µm long and 3–4.5 µm wide in POTAPOVA et al. 2003; 11.8–16.6 µm long and 3.7–4.4 µm wide in TREMARIN et al. 2015), striae (38–42/10 µm in POTAPOVA et al. 2003; ca. 42/10 µm in TREMARIN et al. 2015), and areolae (40–50/10 µm in TREMARIN et al. 2015). However, this species was not described and illustrated showing gradual length/width diminution associated with valve outline modification, as we observed in *N. semifasciata*. While in *N. semifasciata* the axial area varies from narrow to wide and the central area always extends onto the valve margin, LANGE-BERTALOT & KRAMMER (1987) originally described *N. wellneri* with broadly expanded axial and central areas, limited by very short striae. POTAPOVA et al. (2003), POTAPOVA (2010), and TREMARIN et al. (2015) illustrated individuals of *N. wellneri* similar to the protologue, but exemplars with a narrower axial area (Figs 76, 77; internal view; 137, 139, respectively) and the central area reaching the margin (Figs 78, 79 in POTAPOVA et al. 2003 and internal view in POTAPOVA 2010) are included, suggesting the close relationship of these specimens with *N. semifasciata*.

Considering the central area, its symmetry, shape, size, and extent are important morphological features within *Nupela* (see SIVER et al. 2007; WOJTAŁ 2009; BAHLS 2011; YU et al. 2017; GENKAL & YARUSHINA 2018; RYBAK et al. 2020). The central area unilaterally reaching the margin, consistently observed in *N.*

Table 2. Morphological information of *Nipela semifasciata* sp. nov. compared to similar taxa.

Species	<i>N. semifasciata</i>	<i>N. deformis</i>	<i>N. lesothenis</i>	<i>N. neotropica</i>	<i>N. paludigena</i>	<i>N. subpullavicini</i>	<i>N. tenuicephala</i>	<i>N. wellneri</i>
Reference	Our study	LANGÉ-BERTALOT & MOSER (1994)	SCHOEMAN (1973) ¹ SALA et al. (2014) ²	LANGÉ-BERTALOT & MOSER (1994)	SIVER et al. (2007)	METZELTIN & LANGÉ-BERTALOT (1998) ¹ SALA et al. (2014) ²	HUSTEDT (1942) ¹ LANGÉ-BERTALOT (1993) ²	LANGÉ-BERTALOT & KRAMMER (1987)
Length (µm)	6.7–16.4	11–18	5.3–13.5 ¹	12–18	10–20	11–16 ¹	11–15 ¹	12–14
Width (µm)	3.2–4.8	4–5.5	2.6–3.2 ¹	3–4	3–5	3.3–4.7 ¹	2.5–3 ¹	4–4.8
Length/width	1.7–3.7	n.d.	3–3.6 ²	n.d.	n.d.	n.d.	n.d.	n.d.
Striae in 10 µm	40–48	ca. 60	42–46 ¹	50–55	38–45	40–45 ¹	n.d.	45
Areolae in 10 µm	39–59	n.d.	50–68 ¹	n.d.	n.d.	28–37 ²	n.d.	50
Valve outline	Elliptical–lanceolate, with subcapitate to broadly rostrate–rounded apices	Elliptical–lanceolate, with rostrate apices	Elliptical–lanceolate, with capitate to broadly rounded apices ¹	Elliptical–lanceolate, with abruptly rostrate–protracted apices	Elliptical–lanceolate to linear–lanceolate, with rostrate to capitate apices	Elliptical, with capitate apices ¹	Linear, with small and capitate apices ¹	Lanceolate, with subcapitate–rounded protracted apices
Central area	Asymmetric, broadly rhombic to rounded, unilaterally reaching the mantle	Asymmetric, lanceolate, not reaching the mantle	Asymmetric, indistinct, unilaterally reaching the mantle ¹	Asymmetric*, unilaterally reaching the mantle	Asymmetric*, elliptical to rectangular, not reaching the mantle	Asymmetric*, broadly triangular, unilaterally reaching the mantle ¹	Symmetric*, indistinctly linear, unilaterally reaching the mantle** ²	Asymmetric, broadly lanceolate, not reaching the mantle*
Valve mantle	Single row of areolae, interrupted at center, becoming two rows near the apices	n.d.	Single row of areolae, interrupted near the apices ²	Single row of areolae, interrupted by the central area*	Single row of areolae, not interrupted	Single row of areolae, interrupted at the apices and at the center ²	Single row of areolae, interrupted by the central area** ²	n.d.
Raphe development	One valve with long raphe slits and the other valve with slightly shorter raphe slits	One valve with long raphe slits and the other valve with rudimentary raphe slits	One valve with long raphe slits and the other valve with rudimentary raphe slits ¹	Both valves with long raphe slits*	Both valves with long raphe slits	At least one valve with long raphe slits*	One valve with long raphe slits and the other valve with short raphe slits** with “One valve with long raphe slits and the other valve with short raphe slits” ^{1,2}	Both valves with long raphe slits*
External/internal proximal raphe ends	Simple or pore-like	Inconspicuous / n.d.	Slightly expanded ^{1,2}	Pore-like / n.d.	Teardrop-shaped / T-shaped	Pore-like ¹ / Hook-shaped ²	Pore-like ² / Hook-shaped ²	Pore-like / Simple*

*Personal observations based on cited literature. n.d.: no data.

semifasciata, is shared by some other *Nupela* taxa (see LANGE–BERTALOT 1993, 1999; LANGE–BERTALOT & MOSER 1994; METZELTIN & LANGE–BERTALOT 1998; SIVER & HAMILTON 2005; SIVER et al. 2007; POTAPOVA 2011; KULIKOVSKIY et al. 2015; TREMARIN et al. 2015; BAHLS 2017), indicating a well-established feature that should be considered for species level identification within this genus.

The mantle ornamentation of *N. semifasciata* highlights interesting morphological details. Firstly, the mantle partially lacks the row of transversely elongated areolae due to the central area that reaches the valve margin and extends toward the mantle. In fact, *Nupela* species with a central area expanded onto the margin are observed in *N. amabilis*, *N. torganiae* (see TREMARIN et al. 2015, fig 9 on page 81, and figs 124, 127 on page 89), *N. potapovae* (see BAHLS 2011, fig 14 on page 170 and figs 19, 21 on page 171), *N. elongata* and *N. vasta* (see KULIKOVSKIY et al. 2015, fig 10 on page 267, and fig 23 on page 271), but the row of areolae on the mantle is continuous, not interrupted as in *N. semifasciata*.

Further, the mantle is composed of a single row of elongated areolae throughout the whole valve, and a second row can be visualized near the apices. Taxa sharing similar morphological aspects were recently described by KULIKOVSKIY et al. (2015) based on samples from Lake Baikal, in south-eastern Siberia, Russia. *Nupela gomphosphenioides* Kulikovskiy et Lange–Bertalot possesses one or two areolae at the junction with the valve mantle and the second row appearing near the apices (Fig 24 on page 257). *N. neogracillima* subsp. *baicalensis* Kulikovskiy et Lange–Bertalot contains one or two areolae at the edge of the valve face and mantle, with the second row more evident in the median region of the valve (Figs 1, 2 on page 279). Despite that, those taxa appear to have areolae only at the junction between face and valve mantles, differing from *N. semifasciata*, in which the two areolae are always positioned in the mantle. Moreover, *N. potapovae* Bahls seems to have a second row of areolae near the apices on the valve mantle (BAHLS 2011, fig 16 on page 170), although the author has not pointed this out. Both of these aspects were not highlighted in the descriptions of *N. wellneri* provided by LANGE–BERTALOT & KRAMMER (1987), POTAPOVA et al. (2003), and TREMARIN et al. (2015), however, they represent important features to distinguish *Nupela* species.

Considering the striae, *N. wellneri* in LANGE–BERTALOT & KRAMMER (1987) has a radiate pattern throughout the valve, almost parallel in the apices, while POTAPOVA et al. (2003, figs 77–79 on page 304), POTAPOVA (2010, internal view), and TREMARIN et al. (2015, fig 139 on page 90) depicted some specimens with slightly convergent striae patterns near the apices. In *N. semifasciata*, the convergent pattern near the apices was quite common due to the occurrence of well-marked Voigt fault.

The proposition of the new species is supported by observations in our populations compared to descriptions of other *Nupela* taxa provided in the literature. Particularly, *Navicula wellneri* (= *Nupela wellneri*), which shares the closest morphological features, differs by the broadly expanded axial and central area limited by shortened striae, not reaching the valve margin, and by the radiate striae pattern along the valve. This contributes to support the idea that individuals sharing these same aspects, described as *Nupela wellneri* by POTAPOVA et al. (2003), POTAPOVA (2011), and TREMARIN et al. (2015), might be considered *Nupela semifasciata*. Furthermore, it is possible that the second row of areolae at the junction of the valve face and mantle, or even the second areolae positioned in the mantle, instead of a single row, appeared several times in *Nupela*, but further studies, especially those using molecular and phylogenetic techniques, are needed in order to clarify whether this feature represents a homoplasy. Considering that the number of *Nupela* taxa continues to increase, we suggest the central area unilaterally reaching the valve mantle, interrupting the row of areolae, and the second row of areolae on the valve mantle as morphological criteria that must be considered for species level identification within the genus *Nupela*.

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