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Two new species of *Nitzschia* (Bacillariophyta) from shallow wetlands of Peninsular India

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Abstract

The majority of species belonging to the genus *Nitzschia* are distinguished by minute taxonomic features that are difficult to observe and document. Currently, geographical distributions for many species are recognized as cosmopolitan; in contrast endemic species are poorly documented and studied. Our study describes two new species of *Nitzschia* from shallow wetlands across the Bangalore urban district of peninsular India, *Nitzschia taylorii*, *sp. nov.* and *Nitzschia williamsi*, *sp. nov.* Morphological analyses of these new species were performed with light and scanning electron microscopy, and the ecology of inhabited wetlands are discussed briefly. New species records from urban polluted wetlands provide evidence for broadening taxonomic and ecological investigations of cosmopolitan genera like *Nitzschia* in the Southern Hemisphere.

Key words: Diatoms, Bangalore, Tropical wetlands, Nitzschiaceae, new species

Introduction

The freshwater diatom flora of the Indian subcontinent has been studied since the 18th century (Ehrenberg 1845, Skvortzow 1935, Gandhi 1966, 1998, Foged 1976, Sarode & Kamat 1980). Recently there have appeared a few reports of new species and new combinations from India, particularly from biodiversity hotspots like the Himalayas and Western Ghats. Some of these recently described species belong to the genera *Achnanthidium* Kützing (1844: 75; Wojtal *et al.* 2010, Jüttner *et al.* 2011), *Cymbopleura* (Krammer 1982: 20) Krammer (1999: 292; Van de Vijver *et al.* 2011), *Gomphonema* Ehrenberg (Jüttner *et al.* 2004, Karthick *et al.* 2011), *Oricymba* Jüttner, Krammer, Cox, Van de Vijver & Tuji (Jüttner *et al.* 2010: 408), *Pleurosigma* W.Smith (1852: 2; Karthick & Kociolek 2012), *Surirella* Turpin (1828: 362; Karthick *et al.* 2012) and *Pleurosira* (G. Meneghini) V.B.A. Trevisan di San Leon (1848: 96) and *Spicaticribra* J.R.Johansson, Kociolek & R.L.Lowe (2008: 368; Karthick & Kociolek 2011). Most of these studies report new taxa and combinations from samples collected in conservation reserves. In contrast, studies on the diatom floral diversity of wetlands in human dominated landscapes (anthropogenically impacted) are scarce.

In India taxa belonging to the genera *Nitzschia* Hassall (1845: 435), *Navicula* Bory de Saint-Vincent (1822: 128), *Cyclotella* (Kützing 1833: 535) de Brébisson (1838: 19) and *Diadesmis* Kützing (1844: 109) are recorded as cosmopolitan, dominating impacted water bodies and thus are pollution tolerant taxa (Alakananda *et al.* 2011). The genus *Nitzschia* is less understood because of the complexity in identification and its wide ecological preferences. Members of the genus *Nitzschia* are generally recognized as cosmopolitan in distribution and many have wide species tolerances to ionic concentrations and nutrient enriched aquatic

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ecosystems. Studies from tropics particularly in India (Humane *et al.* 2010, Sharma *et al.* 2011) have focused on diatom distributions from various environments; but inevitably taxon identifications remain at the genus level. Examining the taxonomy and ecology of species of *Nitzschia* in wetlands near human habitation within the continent of India will provide more insights into documenting dominant species and their application in biomonitoring.

Bangalore, with an urban dominated ecosystem in the southern regions of India, harbors many historical wetlands which are unexplored for diatom distribution and diversity. These wetlands maintain a semi-arid climate containing numerous interconnected shallow aquatic systems that helps sustain water residence in the neighborhood. Most of the wetlands in peninsular India are less than 2000 years old, created at the time of Kings Era to meet water requirements during the water scarce summer months (Buchanan-Hamilton 1870, Kamath 1980). Forty-eight of these historically constructed wetlands were examined in an ongoing Aquatic Ecosystems Research program in the region. During this study, two new species of *Nitzschia* were observed from Thalghattapura and Begur wetlands within the region of Bangalore and presented with autecological information.

Materials and Methods

Study Area

The wetlands of urban and rural districts of Bangalore, South India were surveyed for diatoms and water quality assessment during February–March 2009. This communication reports samples from two wetlands:

Thalghattapura wetland (Lat. 12°52'1.00" N, Long. 77°31'52.66" E) is located at the south part of Bangalore. This wetland is 2.5 m deep with a catchment of 217.5 ha including water body, open fields, forest and residential areas. This wetland also receives domestic sewage from nearby residential area.

Begur wetland (Lat. 12° 52′ 20″ N, Long. 77° 37′ 58″ E) is situated in the southeast sector of Bangalore with an area of 49.8 ha and a deep of 3 m. Agricultural fields and residential area surround the immediate vicinity of the wetland. This wetland is considered sacred due to the presence of a 1300-year-old Dravidian temple from the Chola Dynasty. This wetland also receives domestic sewage from nearby residential area.

Sample collection and treatment

Epiphytic samples were collected from aquatic plants near the wetland inlet region by crushing the submerged roots and stems. The resulting suspension was transferred into a plastic storage container. Epilithic samples were collected by vigorously scrubbing 3–5 stones from the substratum with a toothbrush and the resulting suspension transferred into plastic storage containers. Both samples were preserved in 70% ethanol.

A portion of the sample was cleaned by removing the organic matter using a hot HCl and $KMnO_4$ treatment (Taylor *et al.* 2007a, Karthick *et al.* 2010). Subsamples of the cleaned material were mounted onto glass slides with Naphrax® as the mounting medium and subsequently observed with an Olympus BX-51 light microscope equipped with DIC optics and a Jenoptic digital camera for image capture. Scanning electron microscopy was performed using cleaned specimens air dried on small squares of aluminium foil, and mounted on aluminium stubs using double-sided carbon tape. Stubs were sputter coated with gold and subsequently examined with a FEI XL30 ESEM (Canadian Museum of Nature Ottawa) using accelerating voltages of 5–30 kV and a working distance of 7–12 mm. Alternately cleaned specimens were air-dried onto cover glasses, attached to aluminium stubs, sputter-coated with 10 nm of Au-Pd, and examined in high vacuum mode with FEI Quanta 200 ESEM at the Nanoscience Initiative Facility, Indian Institute of Science. Water chemistry analyses were carried out according to standard methods (APHA 2005, Trivedy & Goel 1986).

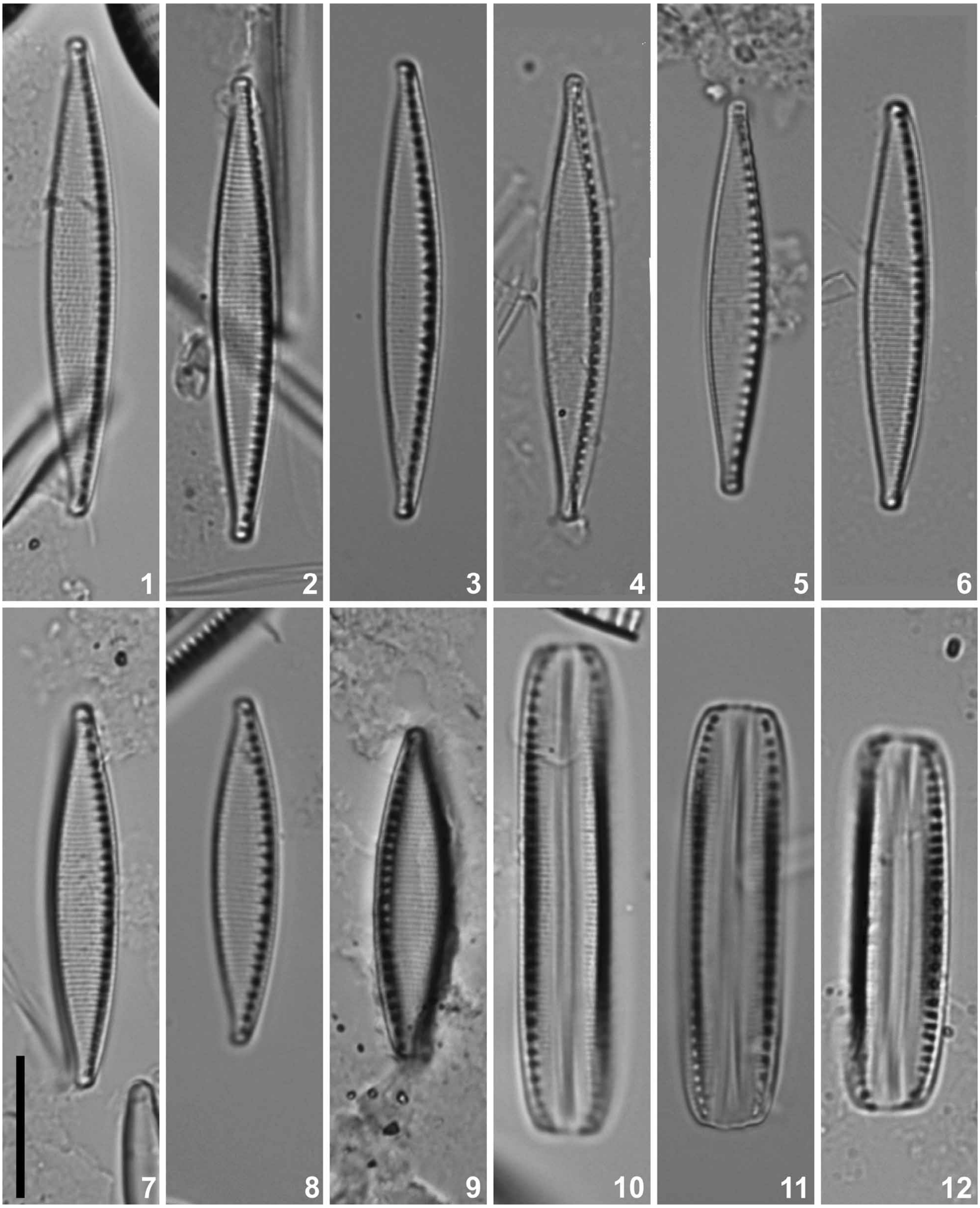
Abbreviations: LM=Light microscope; SEM=Scanning electron microscope.

CESH = Centre for Ecological Science Herbarium Diatom Collection, Indian Institute of Science, Bangalore, India. Other herbarium abbreviations follow Index herbariorum (http://sweetgum.nybg.org/ih/).

New Species Descriptions

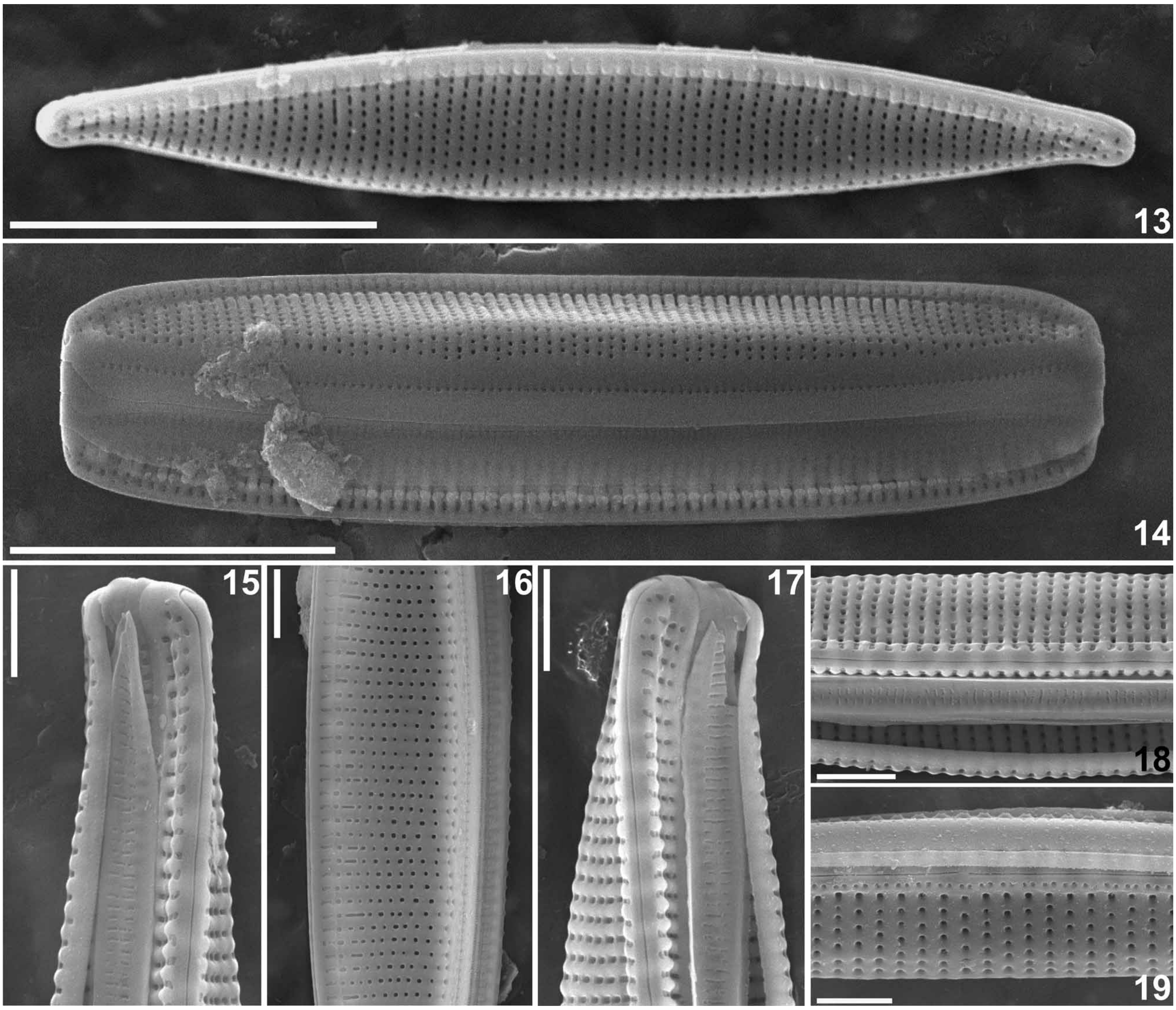
Nitzschia taylorii Alakananda, P.B.Hamilton & Karthick, sp. nov. (Figs 1–25)

Valves lanceolate to linear lanceolate with protracted round to capitate apices. Valve mantle wider on keel side with siliceous nodules present immediately below keel. Length 22–42 µm, width 5–7.5 µm with 21–25 striae

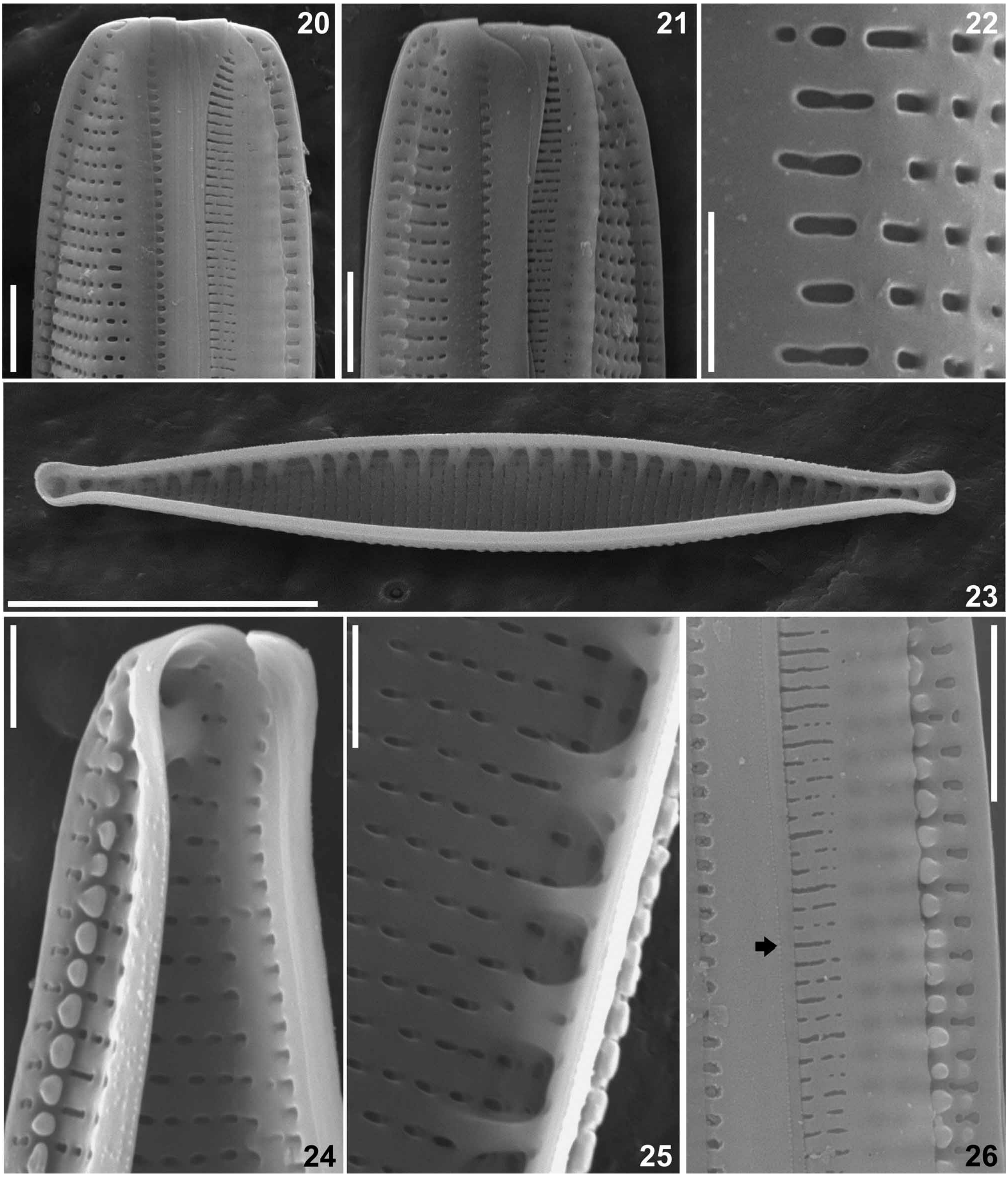


FIGURES 1–12. Light microscopy of *Nitzschia taylorii* sp. nov. Figs. 1–9. LM of valve view showing the size diminution series. Figs. 10–12: LM of girdle view. Fig. 7 = holotype. Scale bars = 10 µm. (Specimens from holotype slide CESH-5-1881)

in 10 µm and 10–14 fibulae in 10 µm. Keel marginal, rounded, elevated from valve face and mantle (Figs 13, 14). Raphe continuous from apex to apex without central area (Fig. 18) and with terminal apices deflected towards valve face as a continuous loop across apex mantle (Figs 15, 17). Striae uniseriate across valve face, extending onto keel (Fig. 16). Mantle on opposite side of keel, with 2–3 elongated areolae and a broad hyaline basal margin (Fig. 18). On keel side, mantle with 2–4 elongated areolae comprising each stria (Figs 20, 21) with a solid surface at basal margin scattered with small papillae (Fig. 26). Areolae on valve face round to elongated depressions, not occluded (Fig. 22). Internally, each stria covered by hymen, and fibulae round to rectangular in shape throughout valve (Figs 23, 25). Cingulum composed of numerous open copulae. Epicingulum of four bands, all with different surface structure. Valvocopula with single row of large elliptical pores on pars exterior and a row of small fine papillae along bottom of band (Figs. 20, 26). Second and third bands with narrow external exposure, with no visible pores, but with fine papillae along bottom of band (Fig. 26). Fourth band broad with a series of narrow elongated pores along pars exterior and a wide area devoid of structure at base of band (Fig. 21).



FIGURES 13–19. SEM micrographs of *Nitzschia taylorii* sp. nov. Fig. 13. SEM of external view of the entire valve. Fig. 14. SEM of girdle view showing valve bands. Fig. 15. SEM view of valve apex with deflected raphe terminal. Figs 16–17. SEM external view of raphe, valve mantle and areolae structure. Fig. 18. SEM of external view of central area of *N. taylorii* showing uninterrupted raphe. Fig.19. SEM of external view of central area of *N. frustulum* showing interrupted raphe. Scale bar in Figs 13, 14 = 10 µm; Figs 15–19 = 2 µm. (Specimens from sample CANA 85055)



FIGURES 20–26. SEM micrographs of *Nitzschia taylorii* sp. nov. Figs 20, 21. SEM of external girdle view showing valve bands. Fig. 22. SEM external view of areolae structure. Fig. 23. SEM internal view of entire valve. Fig 24. SEM internal view of valve apices. Fig. 25. SEM internal view showing round to rectangular shape fibulae. Fig 26. SEM external valve view showing basal margin scattered with small papillae (note arrow mark). Scale bars in Figs 20, 21, 26 = 2 µm; Figs 22, 24, 25 = 1 µm; Fig. 23 = 10 µm. (Specimens from sample CANA 85055)

Type:—INDIA. Bangalore: Begur wetland situated at Bangalore, 12° 52’ 20" N, 77° 37’ 58" E, elevation 900 m, March 2009. *B. Alakananda & G. Supriya*, s.n. (holotype CESH-5-1881! (circled specimen on slide); isotype CANA 85055! (circled specimen on slide)).

TABLE 1. Comparison of characters for selected members of linear-lanceolate species of *Nitzschia* to *N. taylorii* sp.nov. (CA = central area, N-CA = no central area)

Taxon	Valve: Shape	Length (μm)	Width (μm)	Fibulae (in 10 μm)	Fibula shape	Central area	Striae (in 10 μm)	Areolae	Reference
N. taylorii sp. nov.	Lanceolate, linear-lanceolate with protracted round to capitate apices	22–42	2.5–5	12–14	Round to rectangular	N-CA	~50	Rounded; covered by the hymen	Current study
N. costei Tudesque, Rimet & Ector	Linear (longest valve); lanceolate to elliptical (Small valves); Protracted towards the subcapitate ends	8–45	2.5–4.5	9–12	Not equidistance, Irregularly distributed	CA	23–27	--	Tudesque *et al.* (2008)
N. fonticola Grunow	Lanceolate with rostrate to subcapitate ends	7–46	2.5–5.5	10–12	Wider separation between 2 central fibulae	CA	26–30	--	Tudesque *et al.* (2008)
N. macedonica Hust.	Linear-lanceolate to linear with protracted rostrate to subcapitate ends	10.3–48.6	2.8–4	12–17	Irregularly distributed and connected with one or two transapical costae	CA	27–35	Rounded to slightly elongated	Tudesque *et al.* (2008)
N. tropica Hust.	Narrowly lanceolate with weakly protracted ends	14.5–44.6	3–3.7	8–10	Irregularly distributed along the keel	CA	23–25	--	Tudesque *et al.* (2008)
N. frustulum (Kütz.) Grunow	Valves elliptical, lanceolate, linear-lanceolate to linear	3–60	2–4.5	8–16	Block shaped	CA	19–32	--	Taylor *et al.* (2007b)
N. liebethruthii Rabenh.	Valves lanceolate, linear lanceolate to linear	5–40	2–4.5	7–10	Block shaped	N-CA	20	--	Taylor *et al.* (2007b)

Ecology:—*Nitzschia taylorii* was found in three wetlands viz., Begur, Hulimavu and Vaderahalli, characterized by basic pH (8.6 ± 0.6), alkalinity of 293.3 ± 80.8 mgL^{-1} and conductivity of 735.7 ± 322.5 μScm^{-1}. Nitrate and phosphate values of these wetlands were 0.56 ± 0.95 mgL^{-1} and 0.33 ± 0.39 mgL^{-1} respectively. BOD and COD of these wetlands were recorded as 13.5 ± 7.9 mgL^{-1} and 37.3 ± 11.7 mgL^{-1} respectively.

Etymology:—The species epithet is named for our colleague and friend Dr. Jonathan Charles Taylor (North West University, South Africa) whose support for diatom studies in India is hereby acknowledged and who has been an inspiration to both Karthick and Alakananda.

Observations:—*Nitzschia taylorii* is distinguished by the separation of the keel from the valve face, the continuous raphe, large uniseriate areolae on the keel, row of nodules on the mantle below the keel, and the distinct morphology of the epicingulum bands. This taxon can be compared to *N. solita* Hustedt (1953: 152) in its general outline, although *N. taylorii* is more lanceolate (not constricted linear-lanceolate) and with distinct capitate apices. Both *N. taylorii* and *N. solita* have the same areolate morphology on the valve face and keel. *Nitzschia taylorii* is distinguished from *N. steynii* Cholnoky (1966: 207) by its lanceolate shape, the broader fibulae and finer striae. *Nitzschia taylorii* is less similar to *N. frustulum* (Kützing 1844: 63) Grunow (in Cleve & Grunow1880: 98) and distinguished by its lanceolate shape with protracted capitate apices, uniseriate striae (not biseriate) on the keel. Further *N. frustulum* is presented with an interrupted raphe (Fig. 19), where as *N.*

taylorii is characterized by an uninterrupted raphe (Fig. 18). Specimens of *N. frustulum* sensu lato, identified from brackish-like waters, have also been identified from the type locality of *N. taylorii*.

Four other taxa, *N. costei* Tudesque, Rimet & Ector (2008: 485), *N. macedonica* Hustedt (1945: 946), *N. tropica* Hustedt (1949: 147) and *N. liebethruthii* Rabenhorst (1864: 157) have similar valve outlines, approximate stria densities and fibula structure. In LM observations these taxa could be confused. In SEM, the differences between taxa were distinguished by valve outline (*N. tropica*), areolae formation on the keel (*N. macedonica*, *N. tropica*, *N. costei*), valve surface relief (*N. macedonica*), silica nodules on the mantle side of the keel (*N. taylorii*) and cingulum structure (*N. tropica*, *N. costei*) (compare Table 1).

Nitzschia williamsii Alakananda, P.B.Hamilton & Karthick, *sp. nov.* (Figs 27–53)

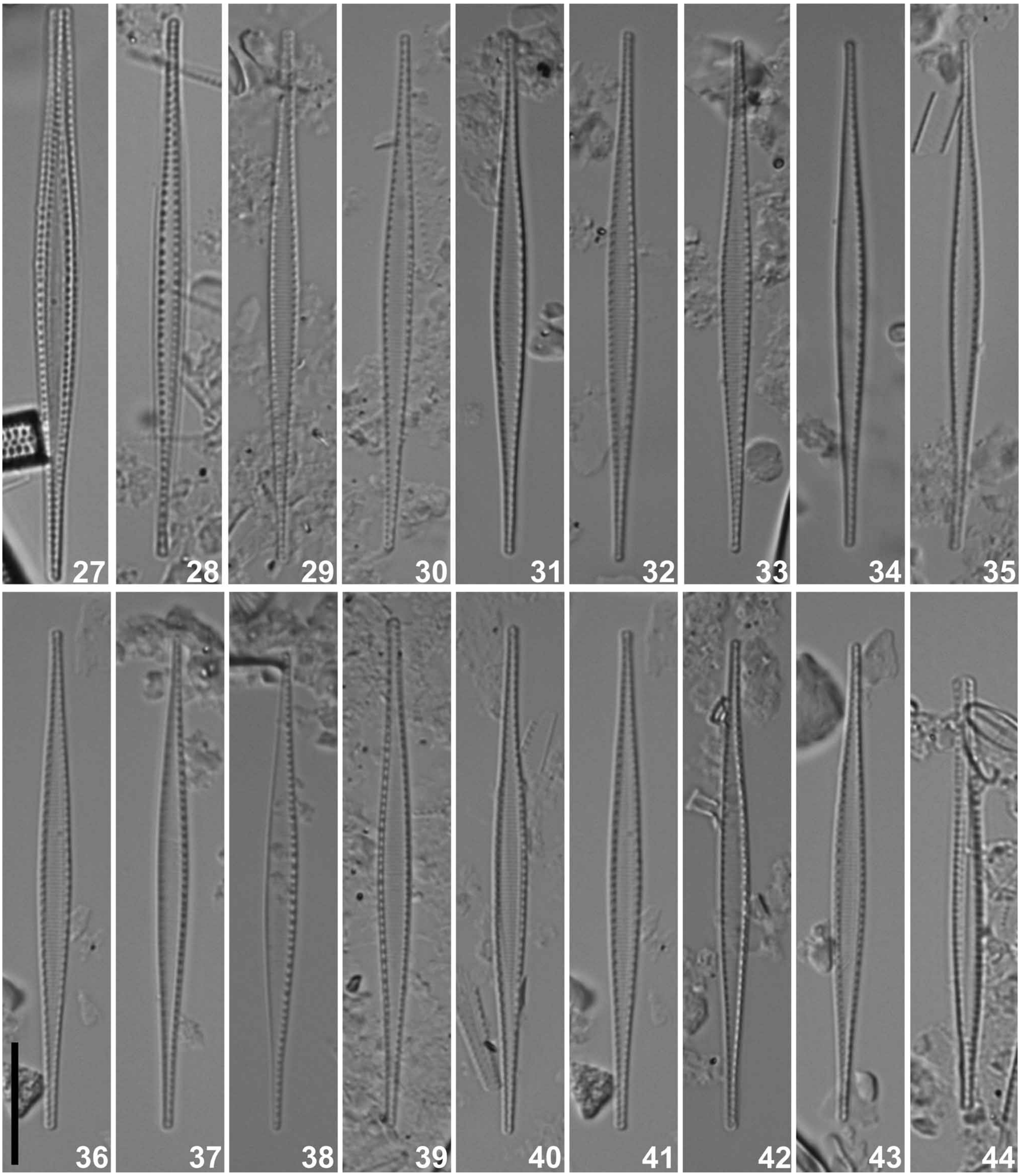
Valves spindle-formed with flatly-rounded apices. Frustules twisted typically, showing valves tilted, with both valve and mantle exposed (Figs 45–48). Length 42–62 µm, width 2.5–5 µm with 12–14 fibulae, 28–35 striae and ~50 areolae in 10 µm. Keel elevated and marginal from valve face in centre (Fig. 45). Raphe continuous, positioned next to a thickened ridge atop keel. Terminal raphe fissures form a small hook along each apex mantle (Figs 47, 48). Striae uniseriate and recessed between elevated ridges on valve face (Fig. 49). Keel with single series of areolae on either side of raphe. Keel and valve face areolae round, occluded by recessed hymen (poroid plate) (Fig. 50). Internally, areolae are irregularly spaced and not apically aligned (Fig. 52). Areolae rounded and covered by hymen. Fibulae round to rectangular, weakly expanding to form attachments on internal valve face and mantle (Figs 51, 52). The two central fibulae are separated by a space equivalent to one to two times their respective widths which corresponds to the presence of a central area; close to apices fibulae broader and more flattened, with small interfibular spaces which are less than width of a single fibula (Fig. 53). Copulae narrow open bands with a single row of pores on pars exterior.

Type:—INDIA. Bangalore:Thalghattapura Wetland at Bangalore, 12° 52' 1" N, 77° 31' 52.66" E, elevation 900 m, March 2009. *B.Alakananda & G.Supriya s.n.* (holotype CESH-05-1882! (circled specimen on slide) isotype CANA 85056! (circled specimen on slide))

Ecology:—*Nitzschia williamsii* was found to be abundant in wetlands with alkaline waters, basic pH (8.7 ± 0.3), alkalinity of 228 ± 91.9 mgL^{-1}, 0.05 ± 0.01 mgL^{-1} nitrates and 0.05 ± 0.003 mgL^{-1} phosphates. Moderate specific conductance (789.5 ± 1.4 μScm^{-1}), and oxygen demand (BOD, 13 ± 0.4 mgL^{-1}; COD, 32 ± 2.8 mgL^{-1}) concentrations were recorded. This wetland catchment was characterized by more open spaces and less built-up areas compared to other wetlands in the region. The autecology of associated taxa needs to be further defined.

Etymology:—The species epithet is named for our colleague and friend Dr. David M. Williams (Natural History Museum, London) whose support for diatom studies in India is hereby acknowledged.

Observations:—There are many taxa in the subgenus *Lanceolatae* which have a linear-lanceolate attenuated to needle-like (aciculate) shaped valve (Lange-Bertalot & Simonsen 1978). Differences between these are often subtle and mostly without SEM illustrations of the characters. Hustedt described a number of these taxa and in LM they are typically distinguished by a combination of characters from valve-shape to stria density (Table 2). *Nitzschia williamsii* is distinguished by the linear attenuated (not strictly acicularis) shape, the rounded (not capitate) apices and stria density. *Nitzschia williamsii* is most similar to *N. acicularioides* Hustedt (1959: 415), although the valves are more lanceolate, the striae are visible in LM and the fibulae are broader. *Nitzschia subacicularis* Hustedt (in Schmidt *et al.* 1922: pl. 348, fig. 76), *N. suchlandti* Hustedt (1943b: 233) and *N. kuetzingioides* Hustedt (1959: 417) are smaller with a more lanceolate valve outline and different stria densities, whereas *N. rostellata* Hustedt is strictly aciculoid with coarser striae. *Nitzschia williamsii* is easily distinguished from *N. gracilis* Hantzsch (in Cramer 1860(7): 40) sensu lato by the much lower stria density. *Nitzschia williamsii* is often observed contorted especially towards the apices.



FIGURES 27–44. *Nitzschia williamsii* sp. nov. LM. Valves showing the size diminution series. Fig. 33 = holotype. Scale bar = 10 µm. (Specimens from holotype slide CESH-5-1881)

FIGURES 45–53. SEM micrographs of *Nitzschia williamsii* sp. nov. Fig. 45. SEM of external view of the entire valve. Fig. 46. SEM of internal view of the entire valve. Figs 47, 48. SEM of external view of the valve apex showing terminal raphe fissures form a small hook along each apex mantle. Figs 49, 50. SEM external view of uniseriate striae with areolae recessed between elevated ridges. Fig. 51. SEM of internal view of valve centre showing the uniseriate striae. Fig. 52. SEM of internal view of the valve showing the round to rectangular fibulae and its spacing. Fig. 53. SEM of internal view of the valve apex showing the internal terminal nodule. Scale bars in Figs 45, 46 = 10 µm; Figs 47, 48, 50, 53 = 2 µm; Fig. 49 = 0.5 µm; Figs 51, 52 = 1 µm. (Specimens from sample CANA 85056)

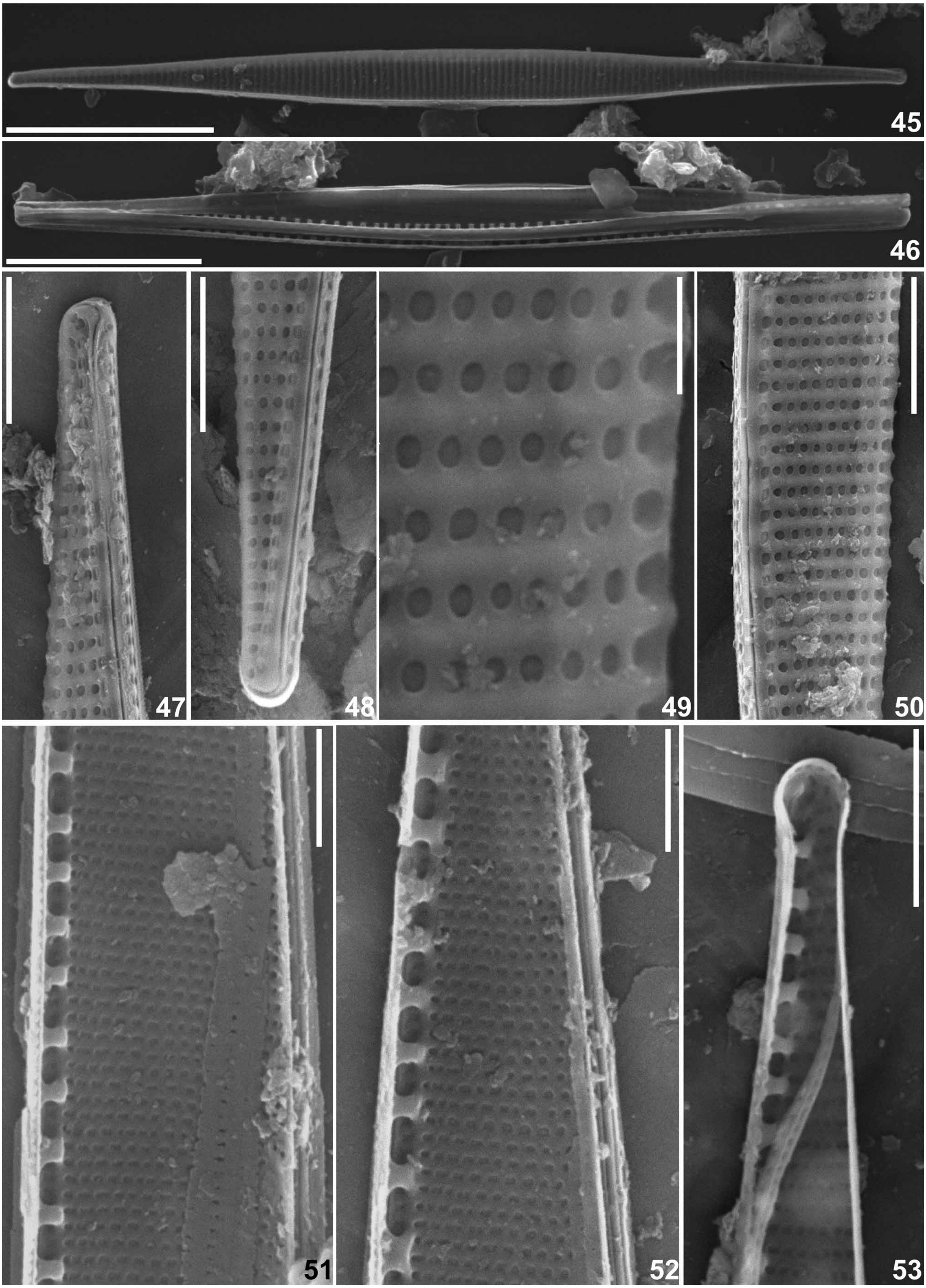


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TABLE 2. Comparison of characters for selected members of linear-lanceolate to aciculoid taxa within the subgenus Lanceolatae species of *Nitzschia* to *N. williamsii* sp. nov. (CA = central area, N-CA = no central area)

Taxon	Valve Shape	Length (μm)	Width (μm)	Fibulae (in 10 μm)	Fibula shape	Central area	Striae (in 10 μm)	Areolae	Reference
N. williamsii sp. nov	Linear-lanceolate, weakly attenuated with rounded apices	42–62	2.5–5	14	Round to rectan-gular	N-CA	28–35	Rounded and covered by a hymen	Current study
N. acicularis Freng.	Aciculoid with capitate apices	30–150	2.5–5	15–22	--	N-CA	60–72	--	Krammer & Lange-Bertalot 1988
N. gracilis Hantzsch	Linear-lanceolate; attenuated, round to capitate apices	40–110	2–4	(12) 16–18	Small	N-CA	Barely visible	--	Lange-Bertalot and Simonsen, 1978
N. pumila Hust.	Aciculoid with capitate apices	30–37	2.5–3	14–18	Small	N-CA	Indis-tinct	--	Lange-Bertalot and Simonsen, 1978
N. rostellata Hust.	Aciculoid with rounded to subcapitate apices	50–100	2.5–5	11–14	--	N-CA	26–30	--	Lange-Bertalot and Simonsen, 1978
N. subacicularis Hust.	Linear-lanceolate with rounded apices	30–42	2–2.5	13–17	Small	CA	22–33	--	Lange-Bertalot and Simonsen, 1978
N. suchlandtii Hust.	Lanceolate, with subcapitate apices	25–50	2–3	13–19	--	N-CA	34–36	--	Hustedt 1943a
N. acicularoides Hust.	Linear to mildly aciculoid with capitate apices	40–60	2.5–3	13–17	--	N-CA	36–40	--	Lange-Bertalot and Simonsen, 1978
N. acicularioides Arch.	Spindle shaped in the middle and vary in different individuals	51–59	2–2.5	--	--	--	Barely visible	--	Archibald, 1966

Discussion

Wetlands in urban landscapes are often neglected for biodiversity exploration and conservation priorities, even though they are biologically rich habitats. The current study reports *Nitzschia taylorii* and *N. williamsii* abundantly inhabiting Begur and Thalghattapura wetlands respectively. These wetlands are with moderate to high trophic conditions and thus these two taxa can be considered indicators of elevated trophic state. Traditional taxa, such as *Nitzschia palea* (Kützing 1844: 63) Smith (1856: 89), *N. intermedia* Hantzsch ex Cleve & Grunow (1880: 95), *N. dissipata* (Kützing 1844: 64) Grunow (1862: 561) and *N. acicularis* (Kützing 1844: 63) Smith (1853: 43) are widely recognized as pollution tolerant taxa and often considered as key species in water quality indices (Prygiel *et al.* 1999, Saros & Fritz 2000, Lavoie *et al.* 2009, Wang *et al.* 2005, Mayama *et al.* 2011). Studies on pollution tolerant genera such as *Nitzschia* will aid further in autecological and biogeographical studies apart from developing diatom based water quality indices for wetlands of Peninsular India.

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