

New freshwater diatom genus, *Edtheriotia* gen. nov. of the Stephanodiscaceae (Bacillariophyta) from south-central China

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SUMMARY

We describe a new genus and species of the diatom family Stephanodiscaceae with light and scanning electron microscopy from Libo Small Hole, Libo County, Guizhou Province, China. *Edtheriotia guizhoiana* gen. & sp. nov. has striae across the valve face of varying lengths, and are composed of fine striae towards the margin and onto the mantle. Many round to stellate siliceous nodules cover the exterior of the valve. External fultoportulae opening are short tubes; the opening of the rimportula lacks a tube. Internally a hyaline rim is positioned near the margin. Marginal fultoportulae possess two arcuate opercles. Areolae have domed cribra on the valve face. Those near the margin do not open to the interior. One to three sessile to slightly-raised rimportulae are placed on the valve face towards the margin; they do not terminate shortened striae towards the center of the valve. This suite of features distinguishes this group from other known genera in the Stephanodiscaceae. *Cyclotella shanxiensis* is transferred to *Edtheriotia*, making the new combination *E. shanxiensis* (Xie & Qi) Kociolek et al. comb. nov. It differs from *E. guizhoiana* by the presence of open pores near the margin of the valve. Features of the new genus are compared and contrasted with others in the Family Stephanodiscaceae. The two species of the genus, unlike most in the family, are known only from rivers and ponds from China and Japan.

Key words: Bacillariophyta, China, diatom, new species, Stephanodiscaceae, *Edtheriotia*.

INTRODUCTION

China is a large (nearly 10 million km²) country of diverse ecosystems, from northern temperate and wet in the north east, to mountainous and dry in the northwest, to subtropical in the south-central portion of the country to tropical on the island of Hainan (Editorial Committee for China's Physical Geography 1985). Elevation extends from sea level in the east to peaks over 8800 m in Tibet. Reflecting this diversity of habitats, the flora and fauna of China are known to have many endemic families, genera and species (e.g. Wu 1980; Chen & Bi 2007; Hong & Blackmore 2015), with several areas being recognized as being biodiversity hotspots, with high numbers of unique and threatened species (Tang et al. 2006).

In contrast, reports on genera and species of freshwater diatoms in China suggest this group of organisms is similar to

the flora of Europe and North America (see lists of genera reported in the freshwater diatom flora of China, Chin 1951; Qi 1995). Early attempts to describe new genera of freshwater diatoms from China have proven difficult to verify (*Porosularia* of Skvortzow 1976) or to be teratologies of previously-known genera (*Amphiraphia* Chen & Zhu 1983, representing initial valves of *Caloneis*; see Mann 1989). More recently, descriptions of endemic species from freshwater environments of China have grown dramatically (e.g. Li et al. 2009; Liu et al. 1990, 2007, 2015; You et al. 2013, 2015; Peng et al. 2014; ; among many others), and genera that are found only in the region (e.g. *Oricymba* Zhang et al. 2015) or as disjuncts with other regions (*Gomphosinica* Kociolek et al. 2015) have also been detailed. From freshwaters, two new genera have been described as endemic to China, the naviculoid genus *Sichuania* Li et al., described from karst regions of Sichuan Province and *Tibetiella* Li et al. (2010), a member of the Fragilariaceae described from mountainous regions of Tibet.

The purpose of the present report is to describe a new genus of the Stephanodiscaceae collected in natural areas south of Guiyang, in south-central China. We present light microscope (LM) and scanning electron microscope (SEM) observations of the new genus, and compare it with similar genera. We discuss the ecological setting in which the genus is found, and its placement among similar genera of the family.

MATERIALS AND METHODS

A composite sample of benthic algae and sediments was collected in a pond at the roadside in the Libo Small Hole, Guizhou Province, China on 2 October 2015. Geographic coordinates of collection site are: N25°27'480" E107°69'192". The water in the pond contained an abundance of *Spirogyra* spp. floating on the surface of water as well as bottom sediments and several species of emergent higher plants. Water temperature (18.5°C) and pH (7.5) were measured in situ with a YSI Pro Plus (YSI Instrument, Yellow Springs, OH, USA).

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In the laboratory, samples were treated with concentrated nitric acid using the Microwave Accelerated Reaction System (Model MARS, CEM Corporation, Mathews, NC, USA), following the procedures of Parr *et al.* (2004) and modified as described in You *et al.* (2015b).

For LM observations, cleaned materials were air dried onto glass coverslips and permanently mounted onto slides with Naphrax mounting medium (Brunell Microscopes, Chippenham, UK). LM observations were conducted using an Olympus BX-51 light microscope (Olympus America Inc., Center Valley, PA, USA) and an Olympus DP-71 digital camera.

For SEM observations, cleaned materials were air dried onto glass coverslips and mounted on aluminum stubs. Stubs were coated with approximately 1 nm of gold using a Cressington 108 sputter coater (Cressington Scientific Instruments Ltd., Watford, UK). SEM images were taken using a JSM 7401 field emission SEM (JEOL, Tokyo, Japan) at an acceleration voltage of 3 kV.

We examined type material of *Perithyra denaria* Ehrenberg, the type species of a genus suggested by Xie and Qi (1984) to possibly be related to the Chinese taxa. We examined mica slides from the Ehrenberg Collection at the Museum für Naturkunde, Berlin, from the type locality of this taxon identified by Ehrenberg (1854). Observations were made on the micas with a Olympus BX-51 light microscope. The mica used for observations is illustrated in Figure 1.

Terminology to describe valve ultrastructure follows that of Theriot and Serieyssol (1994) and Khursevich and Kociolek (2012).

RESULTS

Order Stephanodiscales Nikolaev & Harwood

Family Stephanodisceae Glezer & Makarova

Edtheriotia Kociolek, You, Stepanek, R.L. Lowe et Q-X. Wang, gen. nov.

Description. Valves circular, with valve face flat. A narrow unornamented area is located near the valve margin internally. Valve face externally has rows of siliceous granules aligned with the striae. Near the margins externally there are fine poroids, and star-shaped to rounded granules. Areolae locular, open externally with round foramina, they are occluded internally by individual domed cribra on the valve face. Individual areolae around the valve margin are without occlusions. Externally areolae arranged in radial rows of unequal length, separated by hyaline, non-thickened strips. These hyaline areas are wider in the center of the valve, narrower at the margin and onto the mantle. Marginal fuloportulae distinct, internally each with two arcuate opercles, located on a costa (which may be bifurcated) or between two costae. External fuloportula openings are distinct, as a thickened short tube, with 1–3, sessile to slightly raised rimportulae are present on the valve face, not located at the end of a shortened stria. External fuloportula opening wider than areolae, but external tube is wanting.

Type species of the genus. *Edtheriotia guizhoiana* Kociolek *et al.*

Etymology. Named in honor of Dr. Edward C. Theriot, Director of the Texas Natural Science Center and Professor, Department of Integrative Biology, University of Texas, Austin, for his many and varied contributions in diatom taxonomy, systematics and evolution.

Comments. Marginal fuloportulae were demonstrated in '*Cyclotella shanxiensis*' by Tanaka 2007. Plate 35. Figure 6; Houk *et al.* 2010. Tab. 295 Figure 6 or between two costae (see Xie & Qi 1984, plate 2, fig. 8a,b; Tanaka 2007. plate 35, fig. 2; Houk *et al.* 2010, tab. 295, fig. 4). Variation in the number and structure of the rimoportulae was documented by Xie and Qi (1984), Tanaka (2007) and Houk *et al.* (2010).

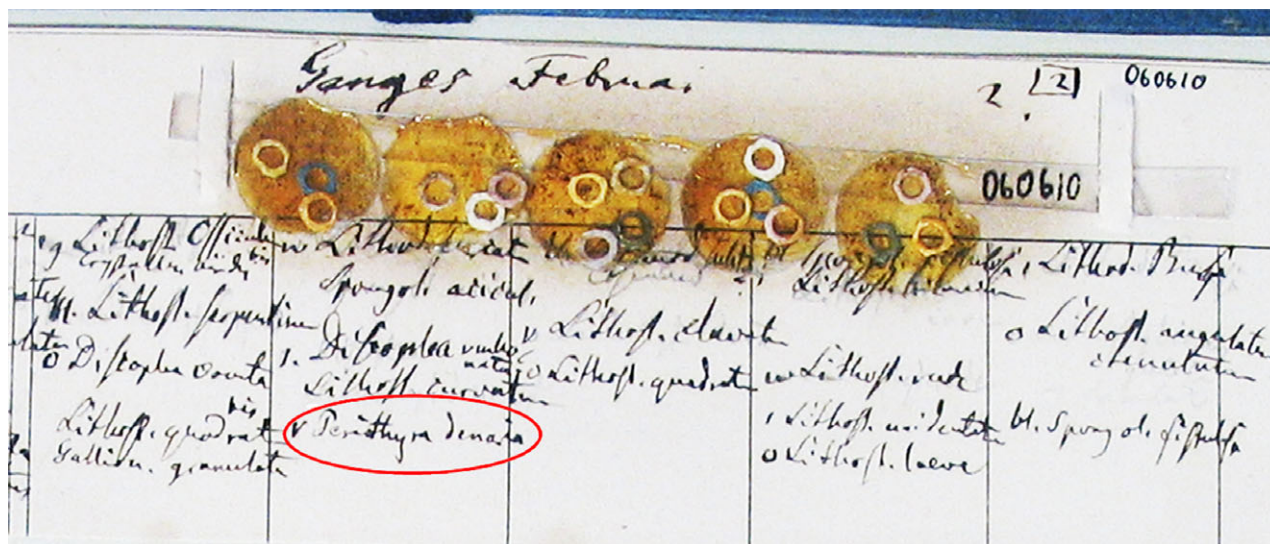
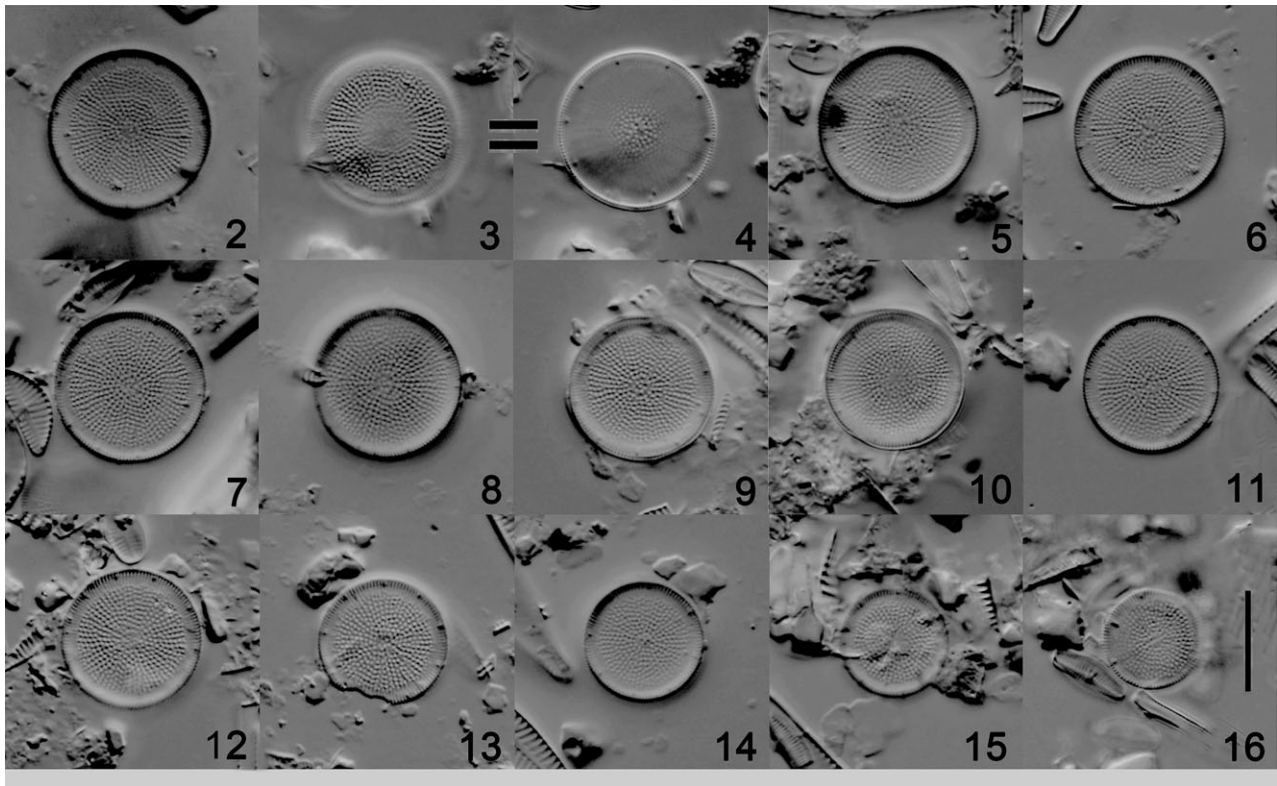


Fig. 1. Image of the mica strip EC 060610 used to study the type of the name of the genus of *Perithyra*, *P. denaria*, from the Ehrenberg collection.



Figs 2–16. *Edtheriotia guizhoiana* sp. nov. Light microscopy, valve view showing size diminution series. Scale bar = 10 μ m. Figure 6 is of the holotype. Images from holotype slide JPK 10 443 at COLO.

Edtheriotia guizhoiana Kociolek, You, Stepanek, R.L. Lowe et Q-X. Wang, sp. nov. (Figs 23–25)

Holotype. Slide and material 10 443, Kociolek Collection, University of Colorado, Museum of Natural History Diatom Herbarium, Boulder, USA. Holotype specimen illustrated in Figure 6.

Isotype. Slide and material GZ-1510041, Shanghai Normal University, Biology Department Diatom Herbarium, Shanghai, China.

Type locality. In the lentic pond, Libo Small Hole, Libo County, Guizhou, China. Collected by J.P. Kociolek & Q-X Wang, 2 October 2015.

Etymology. The species is named for the province in which it was discovered.

Description

Valves circular, valve face flat, 10.0–16.0 μ m in diameter (Figs 1–16). A narrow unornamented area is located near the valve margin internally. Valve face externally has rows of siliceous granules aligned with the striae (Figs 17–21). Striae 16–20/10 μ m at the margin of the valve, areolae loculate, opening externally by round foramina, central areolae occluded internally by domed cribra (Figs 22–24,26), marginal openings occluded by hyaline area around the margin of

the valve (Figs 22–25). Areolae arranged in radial rows of unequal length, 29–36/10 μ m within a stria near the center of the valve, 44–60/10 μ m towards the margin of the valve, striae separated by hyaline strips. Marginal fultoportulae, 6–11 per valve, distinct, internally each with two arcuate opercules (Figs 23–25). External fultoportula openings are distinct, as a thickened short tube. 1–2 sessile to slightly raised rimportula is present on the valve face, not located at the end of a shortened stria (Fig. 17), and the external fultoportula opening wider than areolae, but external tube is wanting (Figs 19,21).

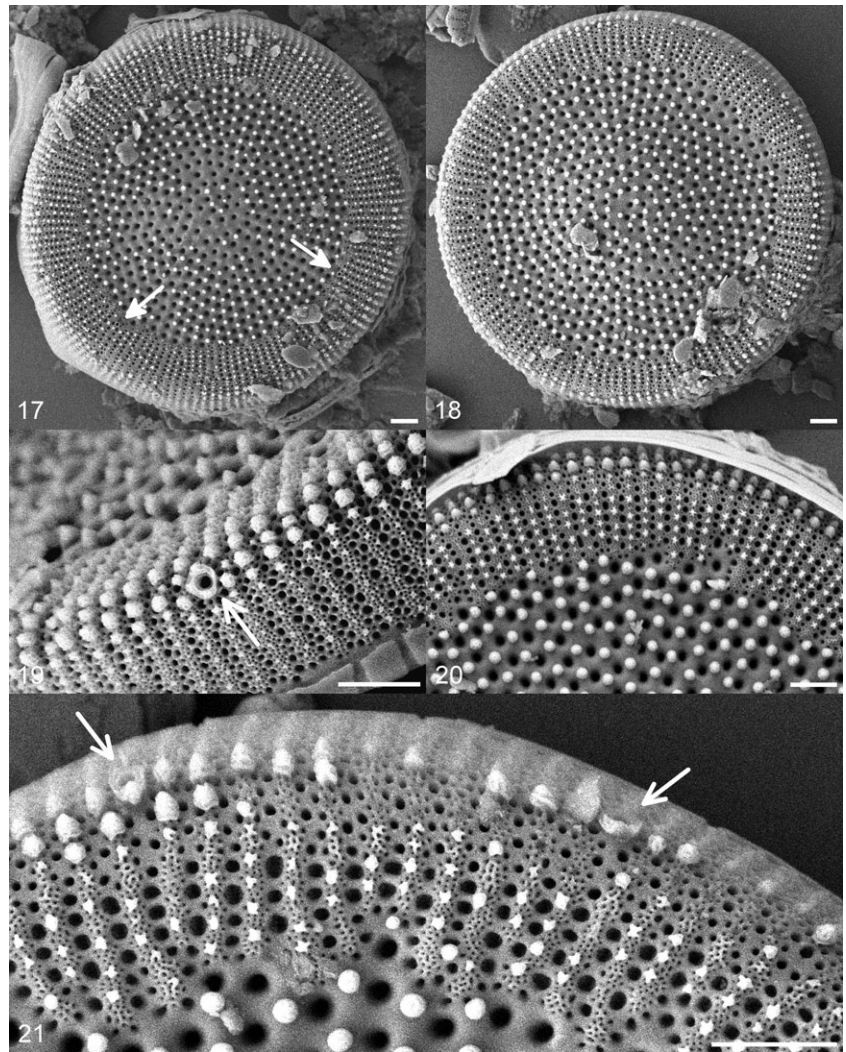
Edtheriotia shanxiensis (Xie & Qi) Kociolek, You, Stepanek, R.L. Lowe et Q-X. Wang, comb.nov.

Basionym: *Cyclotella shanxiensis* (Xie & Qi, 1984), Light, scanning and transmission electron microscopic studies on the morphology and taxonomy of *Cyclotella shanxiensis* sp. nov. In: D.G. Mann (ed.), Proceedings of the Seventh International Diatom Symposium; 22–27 Aug 1982, Philadelphia, PA. Koeltz Science Publishers, Koenigstein. P. 188, plates 1–4, 17 figures.

DISCUSSION

A detailed discussion of the systematics of thalassiosiracean and stephanodiscoid diatoms has been presented by Kociolek *et al.* (2014), based on the phylogeny of Alverson *et al.* (2007). From that discussion we will present ideas about our

Figs 17–21. Scanning electron micrographs (SEM) of *E. guizhoiana* sp. nov. (valve exterior). 17,18. Complete valve view, showing striae with areolae of different sizes from the margin to the center. Striae in the center may extend to the middle or be of a more length. Valve face is covered with a variety of different sized siliceous nodules. External openings of rimportulae are indicated by the white arrows. Distinct fascicles are wanting. 19. Valve mantle showing the striae extend completely across the mantle. Rounded and star-shaped siliceous granules are evident. A small tubular expression of the marginal fuloportula is evident. 20,21. Marginal area of valve face showing pores of a variety of sizes, as well as rounded and stellate granules. Arrows indicate external fuloportulae openings. Spines are lacking. Scale bar 1 μm .

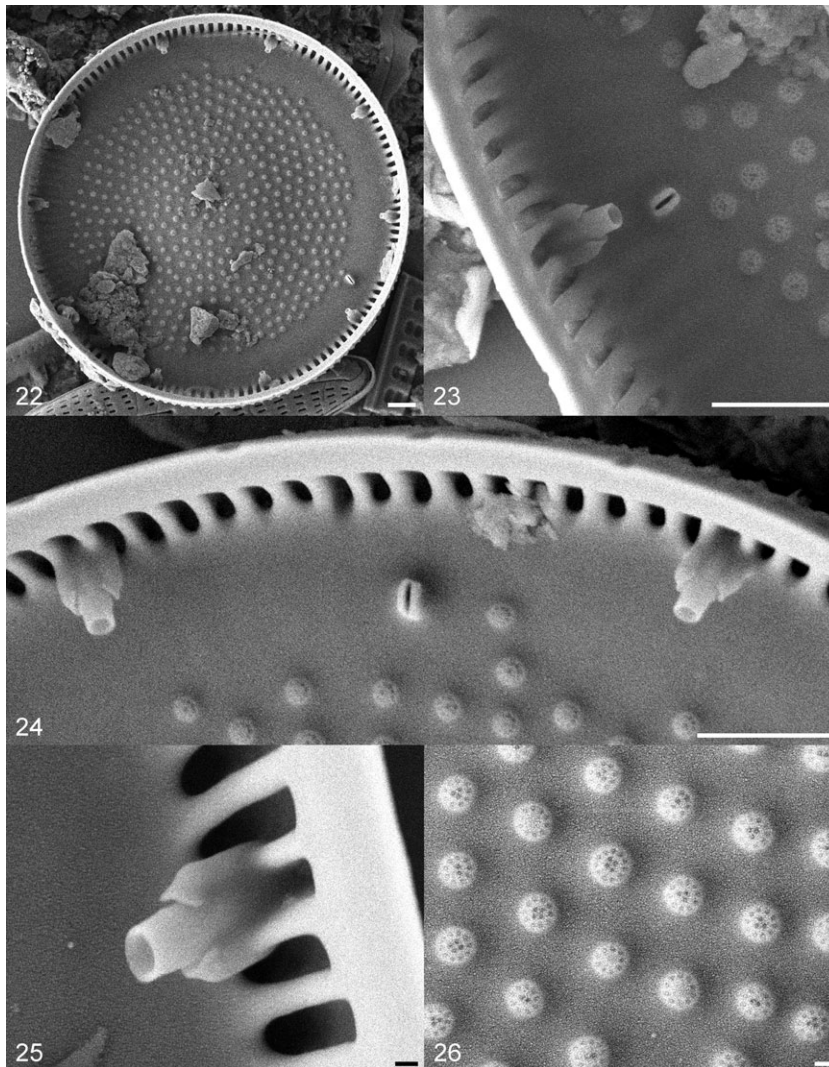


conclusion of these diatoms from China being recognized as a distinct genus.

Given the two different patterns of ornamentation on the valve (Xie & Qi 1984), these species might be assigned to the genus *Cyclotella sensu stricto*. *Cyclotella* as typified by *C. tecta* Håkansson & Ross (= *C. distinguenda* Hustedt) has a solid central area, and slab-like stalks associated with the rimoportulae (Khursevich & Kociolek 2012; Kociolek *et al.* 2015b). Houk *et al.* (2010) continue to treat this species as a member of *Cyclotella*. *Edtheriotia* species have a perforated center and sessile rimoportulae; its morphology suggests a placement outside the Thalassiosiraceae.

Among the Thalassiosiales, *Edtheriotia* is best considered as a member of the Stephanodiscaceae, by virtue of its distinct fuloportula morphology, having three or fewer satellite pores with lunate or arcuate opercles (Kociolek *et al.* 2014). Our new genus resembles *Pliocaenicus* Round & Håkansson emend Stachura-Suchoples & Khursevich by lacking fasciculate striae and being without external extensions of the rimoportulae (Khursevich & Stachura-Suchoples 2008), but differs by lacking the 'centrifugal roofing over' of the marginal costae and central fuloportulae. The fuloportulae occur on or between costae, a feature it shares with *Lindavia*

(Khursevich & Kociolek 2012). Within this family, internal domed cribra align these species from China with *Lindavia* (*Puncticulata* in Alverson *et al.* 2007), *Cyclostephanos*, *Stephanodiscus* and *Strelnikoviella* (Kociolek *et al.* 2014). A recent publication describing the genus *Pantocsekiella* Kiss & Ács in Ács *et al.* (2016), suggests this genus, which lacks internal domed cribra, is a highly derived member of the family. Khursevich (1989) transferred *Cyclotella shanxiensis* to *Cyclostephanos*; however, it lacks fascicles, marginal spines and valve face fuloportulae; all of these characteristics distinguish it from *Cyclotstephanos* (Round *et al.* 1990; Khursevich & Kociolek 2012) and *Stephanodiscus* (Round 1982; Theriot *et al.* 1987; Kociolek *et al.* 2015b). According to Houk *et al.* (2010, p. 44), Nagumo transferred Xie and Qi's species to *Puncticulata* (current name = *Lindavia*). Lack of the rimoportulae at the end of shortened striae and the complex valve face fuloportulae distinguishes these species from *Lindavia* (see Khursevich & Kociolek 2012 for features of this genus, listed as *Handmannia*; see also Budzynska & Wojtal 2011), and, as stated previously, the significant differences of these two Chinese taxa from the type species of *Cyclotella* (and their distant phylogenetic position, outside the Stephanodiscaceae, Alverson *et al.* 2007), suggest it cannot be



Figs 22–26. SEM of *E. guizhoiana* sp. nov. (valve interior). 22. Entire valve view, showing the marginal hyaline area, distinct marginal fuloportulae and the rimoportula positioned on the valve face. Central fuloportulae are absent. 23,24. Valve face: mantle junction showing a sessile rimoportule located near a fuloportula (Fig. 23) and slightly raised rimoportule between fuloportulae (Fig. 24) located on the face. It appears as though areolae do not extend to the margin or to the mantle internally. 25. Marginal fuloportula with two opercles. Marginal costae are thin, and primary only. 26. Valve face areolae with domed cribra. Perforations of the cribra are coarse. Scale bar 1 μm in Figures 22–24; 100 nm in Figures 25–26. Images taken from type gathering, JPK sample 10 443 at COLO.

placed in that genus. It has its own distinguishing features such as fine marginal poroids, presence of an unornamented area near the margin (instead of near the center), and coarsely-perforated domed cribra.

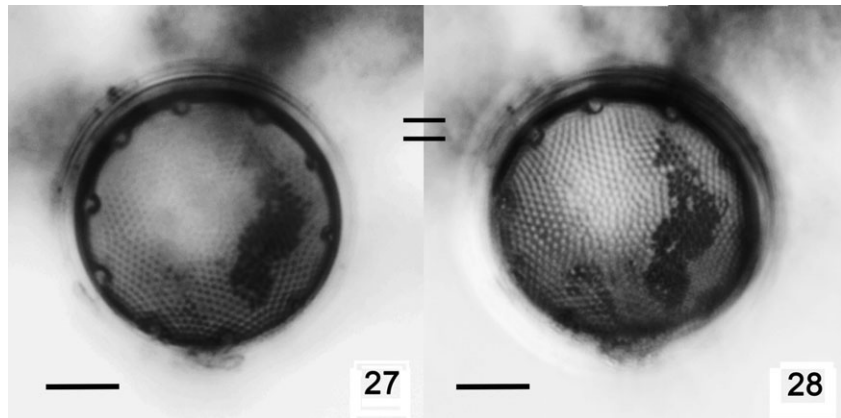
One species outside of China that should be investigated for consideration in *Edtheriotia* is *Lindavia thienemannii* (Jurilj) Nakov *et al.*, described from the spring region of Lake Ohrid, Macedonia, which is alkaline (Levkov *et al.* 2007). Two SEM images of this taxon show similar structure of the external valve face and mantle (as *Puncticulata thienemannii* (Jurilj) Levkov, Levkov *et al.* 2007, plate 10, figs 1,2). Internally, there are several rimoportulae. These are positioned near the margin, as in *Edtheriotia*, versus at the ends of shortened striae as in *Lindavia*.

Outside the genus, *Edtheriotia* species share some superficial similarities with *Cyclotella andina* Theriot *et al.* (1985). Like *Theriotia* species, *C. andina* has indistinct fascicles, and striae are of varying lengths that are composed of much smaller areolae near the margin and onto the mantle. Internally, both *Edtheriotia* species and *C. andina* possess two opercles around the fuloportulae and domed cribra. *Cyclotella andina* possesses long spines and a stalked labiate processes occurring on a costa. This species also has complex alveoli, a

feature it shares with another similar species, *C. omarensis* (Kuptsova) Loseva & Makarova, a tangentially undulate species (Theriot *et al.* 1985). However, Stachura-Suchoples and Khursevich (2007) transferred this species to *Pliocaenicus* Round & Håkansson based upon several highly variable features including the presence of tangentially undulate valves, presence of alveolae and lack of external tubes of the fuloportulae (see also Khursevich & Kociolek 2012), features all lacking in *Edtheriotia* species. This genus is primarily fossil (7 of the 9 species assigned to it are known only as fossils; Khursevich & Kociolek 2012), and known from Europe and Asia (Round & Håkansson 1992; Wang 1999; Tanaka & Kobayasi 1999). The large number of highly variable features used by Stachura-Suchoples and Khursevich (2007) to diagnose *Pliocaenicus* suggests a reinvestigation of features in the species of that genus. It is also possible the placement of *C. andina* might be best accommodated there.

A brief comment is offered here on character evolution in the Stephanodiscaeae as prompted by the description of *Pantocsekiella* as a member of this family. All members of this family were described as having domed internal cribra (Alverson *et al.* 2007), which would help to diagnose the family, but not help distinguish members of the family from one

Figs 27–28. Light micrographs of a single specimen *P. denaria* from type collection, mica EC 060610b, violet ring, of the Ehrenberg Collection. The '=' indicates the figures represent two different focal planes of the same specimen. Scale bar 10 μm .



another. So, relative to other diatom groups the feature of internal, domed cribra is apomorphic, but pleisiomorphic relative to the group itself. The feature could be considered pleisiomorphic for the family and, given the phylogenetic position of *Pantocsekiella*, a secondary loss of domed cribra would have occurred in this genus (See Ács *et al.* 2016, fig. 5).

The two species of *Edtheriotia* share similarities of external valve morphology, including organization of striae, radiating rows of siliceous nodules, the fine structure of fine pores and siliceous bumps and stellate sculpting near the margins. Internally, the two also share the prominent marginal fultoportulae with two lunate opercles and a single, sessile labiate process. The two species differ in that *E. shanxiensis* is larger (up to 27 μm in diameter, versus up to 16 μm in diameter in *E. guizhoiana*), and has areolae that are open nearly all the way to the margin (see Xie & Qi, 1984, fig. 15a,b). That is, its unornamented area at the margin is quite narrow, while in *E. guizhoiana*, the unornamented area is much wider. The characteristic bumps and star-shaped nodules can be variable, as documented by Tanaka (2007, plate 34, figs 1,2) and Houk *et al.* (2010, Table 294, figs 13,14), suggesting this feature may not be diagnostic for the genus or even for a species.

Although these species are included in the Stephanodiscaceae, whose members are mostly planktonic, both *Edtheriotia shanxiensis* and *E. guizhoiana* have only been found in rivers (including in the Inabe River, Japan; Houk *et al.* 2010) or small ponds. Both species appear to occur only in relatively pristine areas. *Edtheriotia guizhoiana* is known from waters high in calcium, from a karst region in south-central China, while *E. shanxiensis* is known from a more eastern region towards Beijing and into Japan.

Xie and Qi (1984) suggested their species may be related to *Perithyra* Ehrenberg ex. Van Heurck, a genus described from the Ganges River (Ehrenberg 1854). The genus was described but not seen directly by Van Heurck (1896). In Ehrenberg's drawings (1854, plate 35A/9, figs 5,6), the unequal and non-fasciculate nature of the striae are evident, suggesting a resemblance with *Edtheriotia*, but the internal 'tubercles' (Van Heurck 1896, p. 492), interpreted here as the marginal rimoportulae, are as wide as 4–5 striae at the margin (seen in both Ehrenberg's and Van Heurck's drawings, and in the figures of the type specimens; see also Figs 27–28.). These structures are much larger than what is seen in either species from China, and are evident in light

micrographs of the type species, *P. denaria*. This species was described from dredgings from the mouth of the Ganges River (Ehrenberg 1854), and the unique internal structures and striae pattern (see Figs 27,28) distinguish it from either species of *Edtheriotia*.

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