Acta Bot. Croat. 68 (2), 339–349, 2009

CODEN: ABCRA 25
ISSN 0365–0588

New diatom taxa from the world's first marine Bioblitz held in New Zealand: *Skeletomastus* a new genus, *Skeletomastus coelatus* nov. comb. and *Pleurosigma inscriptura* a new species

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Diatoms were investigated as part of the world's first marine Bioblitz held on the south coast of Wellington, New Zealand, in October 2007. Two unusual diatoms were associated with the red alga *Herposiphonia ceratoclada.* They were examined by light and scanning electron microscopy. The first, previously described as »*Cocconeis coelata*« Arnott ex Greville, is a thick-ribbed biraphid diatom that appears to have an undular raphe and marginal septum like some *Mastogloia* species, but it does not have partecta in the valvocopula. Instead it has short marginal ribs that support a pseudoseptum. Its solid lyre and multiseriate striae with cibrate pores could be modifications of similar structures in *Aneumastus aksaraiensis* Spaulding et al. It typifies the new genus *Skeletomastus* which appears to be close to both *Mastogloia* and *Aneumastus,* hence the name. The second is a proposed new *Pleurosigma* species, *P. inscriptura* with naviculoid symmetry and a markedly sigmoid raphe system with deflected ends. It appears bright blue in darkfield illumination which helped us locate it in other samples from the Wellington region. We compare it with some other *Pleurosigma* species and consider their diffraction properties.

Keywords: Diatom, Mastogloiaceae, pseudoseptum, costae, lyre, sigmoid, multiseriate, ultrastructure, *Cocconeis*, *Aneumastus, Skeletomastus, Pleurosigma*

Introduction

Bioblitzes aim to document biodiversity in a limited space and time. Experts collect samples and identify species while an invited public watches. In October 2007 a bioblitz was held in the Taputeranga Marine Reserve situated near Wellington City on the coast of Cook Strait, New Zealand. The reserve covers 840 hectares from the foreshore south to 41°22.00' S and from 174°44.67' E to 174° 47.37' E. High energy tidal flows of alternating warmer and cooler water passing through Cook Strait have formed a rocky shore with little shingle or sand. The diatom flora of the reserve is rich, with at least 164 taxa, the commonest genus being *Cocconeis* (19 taxa).

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The diatoms described in this paper were found on subtidal seaweed. One diatom resembled an unusual *Mastogloia* and we propose a new genus for it. The other is a new species of *Pleurosigma* unknown to two experts on *Pleurosigma*: Stidolph from New Zealand (STIDOLPH 1992, 2002) and Sterrenberg from the Netherlands (STERRENBURG 1990; 1991a, b; 2001; 2003).

Material and methods

Seaweeds and other organisms growing in the subtidal zone were collected by divers. Macroalgae were lightly sonicated in a tank (50 kHz, 10 min) to free any epiphytic diatoms. The loose material was partially oxidised with warm hydrogen peroxide and disaggregated with hydrochloric acid before washing. Some of the resulting suspension was mounted in Naphrax (RI 1.73) and examined on a light microscope (LM) (Leitz diaplan) under direct light and differential interference contrast. Diatoms were photographed with a Leica camera and photographs improved in Picasa (cropped, converted to greyscale and sharpened).

Standard darkfield (STERRENBURG 1991a) was obtained using an Olympus CHB student grade microscope with an Abbé condenser and tungsten bulb with the addition of a black disc to obscure the cone of direct lighting at 40X and 100X magnification. This revealed *Pleurosigma* valves on the slides and allowed their darkfield colour to be recorded. Their bright blue colour was used to locate further valves. Part of the suspension was subjected to stronger sonication with an immersed probe (20 kHz, 5 min) to break apart the valves of »*Cocconeis coelata*«. The suspensions were dried down onto aluminium stubs. They were then plasma-ashed for about 1 hour at 5 watts in air at a pressure of about 0.5 millibars. The sample was positioned about 50 mm downstream of the discharge zone to avoid RF heating of the aluminium stub. The procedure effectively removes any organic material without altering the silicate structure. The stub was gold-coated and first examined with a JEOL 733 microprobe in scanning electron mode to see whether the diatom originally called »*Cocconeis coelata*« had obvious partecta. It and a specimen of the *Pleurosigma* were then photographed using a JEOL 6500F scanning electron microscope (SEM).

Theoretical ratios for angles between oblique striae versus oblique pore-spacing are as follows:

If p pores per 10 µm were counted in an oblique line then $a = 10/p$ µm, and if q (transverse) lines of pores per 10 µm were counted crossing a transapical (longitudinal) line then $c = 10/q$ µm. In this case

$$\cos(\theta/2) = c/a = p/q \text{ so } q/p = \sec(\theta/2)$$

For $\theta < 132°$ changing θ by a given percentage changes q/p or p/q by a smaller percentage: $p/q = \cos(\theta/2)$; therefore $\frac{d(p/q)}{d\theta} = ½ \sin(\theta/2)$; so $\frac{q}{p}\frac{d(p/q)}{d\theta} = -1/2 \tan(\theta/2)$; $1/2 \tan\theta/2 < 1$ if $\tan\theta/2 < 2$; therefore $½\, \theta < 66°$ and $\theta < 134°$.

So q/p is obtained more accurately from a measurement of θ than θ is by a measurement of p/q.

Results

Skeletomastus

***Cocconeis coelata* Arnott ex Greville** is a species of neither *Cocconeis* nor *Mastogloia* so we erect a new genus for it.

Skeletomastus (M. A. Harper gen.nov.) monotypic genus

Latin diagnosis: Valvae costis radialibus munitae inter se longitudinaliter connexis ita in sterna laterales duo (lyrae similia) coalescentes. Costae breves marginales (costulae) pseudoseptum fulcrant, quod est (in frustulo) a valvocopula tectum. Striae multiseriatae ex poris circularibus cribratis constantes. Rami raphis extra circa centrum opposite undulati, intra recti.

Valves fortified by radial ribs connected to each other longitudinally thus coalescing into two lateral sterna (similar to a lyre). Short marginal ribs (riblets) support a pseudoseptum, with this being covered (in the frustule) by the valvocopula. Striae multiseriate consisting of circular cribrate pores. Branches of the raphe externally, with respect to the centre oppositely undulate, internally straight.

Etymology: Greek, Skeletos – skeleton and mastus – nipple, the first part based on the comment on its skeleton-like structure in Greville (1862), the second on the similarities in its structure to *Aneumastus* (particularly *A. aksaraiensis*).

Skeletomastus coelatus (Arnott ex Greville) M. A. Harper, comb. nov.

Basionym: *Cocconeis coelata* G. A. Walker Arnott ex Greville, R. K. (GREVILLE 1862: page 234, plate 10, figures 5 and 6).

Lectotype: On British Museum slide, G. A. W. Arnott 56, (S 631) BM 1492 labelled *Eupleuria ocellata* on *Ballia.* This slide contains syntypes and I concur with David William's choice of a lectotype (WILLIAMS 1988: 103, Fig. 4). *Eupleuria ocellata* is a synonym of *Entopyla ocellata* (Arnott) Greville also present on the slides.

Synonyms: *Skeletomastus coelatus* (Arnott ex Greville) M. A. Harper (this paper), *Cocconeis coelata* Arnott ex Greville 1862, *Campyloneis coelata* (Arnott ex Greville) Chase (HABIRSHAW 1885: 79), *Navicula coelata* (Greville) Cleve 1894 (SCHMIDT 1894, Pl.193, Figs. 5, 3). *Cocconeis coelata* erroneously attributed to Gregory (GRUNOW 1867: 9). Some records give »*caelata*« an orthographical variant of the epithet and others »*coelatus*« (VAN LANDINGHAM 1968).

Skeletomastus coelatus has raphes on both valves so it does not fit the genera *Cocconeis* and *Campyloneis*. It was synonymised with a query by CLEVE (1894) with *Diploneis campylodiscus* (Grunow in Schmidt) Cleve (VAN LANDINGHAM 1968). We disagree with this placement because this diatom does not have solid lyres or undular raphes, (see *Navicula campylodiscus* Grunow (SCHMIDT 1875, Pl. 8, Figs. 9, 12). The figures of the original *Navicula campylodiscus* show that the areas each side of the axial rib differ from the multiseriate areas near the margin, giving the appearance of longitudinal canals that have large pores where they meet the outer multiseriate areas. These features indicate *Navicula campylodiscus* really does belongs to the genus *Diploneis*.

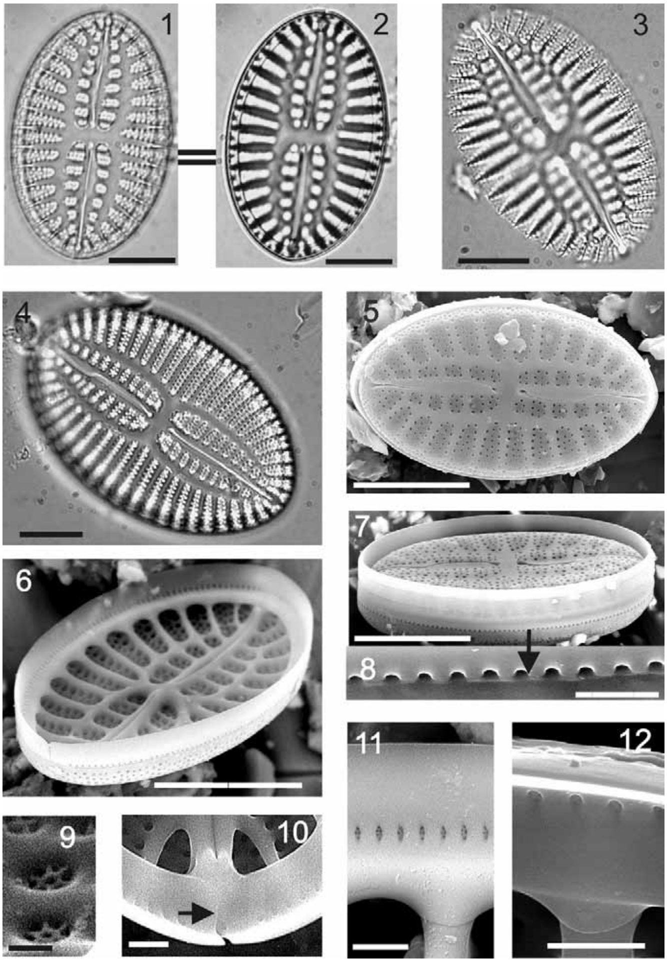

Plate 1. *Skeletomastus coelatus* [*Cocconeis coelata*] in LM (Figs. 1–4) and SEM (Figs. 6–12). Fig.1 – high focus, undulate raphe and multiseriate striae; Fig. 2 – same view low focus, marginal pseudoseptum with short ribs; Fig. 3 – new valve without pseudoseptum showing that this forms later; Fig. 4 – larger valve; Fig. 5 – external view of pores and raphe; Fig. 6 – oblique apical view of split valvocopula and internal costae, Fig. 7 – oblique side view of hypovalve sunk inside epicingulum; Fig. 8 – external pores on edge of valvocopula; Fig. 9 – internal view cribrate pores; Fig. 10 – internal apical view of valvocopula overlapping pseudoseptum; Fig. 11 – internal cibrate valvocopula pores; Fig. 12 – internal view, valvocopula over pseudoseptum. Scale bars: 10 μm (Figs. 1–7); 1 μm (Figs. 10–12), 0.25 μm (Fig 9).

Illustrations: Originals in Greville (1862: Pl. 10, Figs. 5 and 6) include an aberrant rapheless valve. Multiseriate striae are shown in Schmidt (1894: Pl. 193 Fig. 53) and on in the lectotype photograph (Williams 1988: Fig. 4, p. 103).

Exsiccata: Van Heurck (H.V.H.) slides issued as #244 »*Cocconeis coelata* Gregory« (erroneous authority). We have examined one in the Healey Collection (slide # 2682) held at NIWA, Wellington, New Zealand.

The elliptical valves of *Skeletomastus coelatus* have multiseriate striae (Figs. 1, 3, 4), lyrate areas (Figs. 1–4) and a marginal »septum« (Fig. 2) visible under LM. All our valves (>108, including 16 frustules) were fully raphid so this diatom is not a *Cocconeis*. Their length in the original description is 13 to 46 µm (GREVILLE 1862), some of the Island Bay valves were longer (range 22 to 53 µm). Our valves were 11.5 to 34 µm wide. Unlike most diatoms larger valves are wider in proportion, though their lyres are not (Figs. 1, 4). The undular external raphe slits (Fig. 1), become straight inside (Fig. 2). Under the marginal »septum« are projections between the costae (Fig. 2), these appear too short to be partecta walls. Valves without »septa« (Fig. 3) indicate that these form later.

In SEM the flat valves have round pores, wide virgae and undulate raphe slits with expanded ends but no terminal fissures (Fig. 5). Internal views of the valve (Figs. 6, 16) show that thick costae underlie the hyaline areas and pores have cibra (Figs. 9, 14). The raphe slit is in a thick axial ridge (Figs. 6, 13) and a fascia connects this ridge to the solid (Fig. 13) lateral costae of the lyrate area (Figs. 6, 13). The valve mantles have scattered pores (Figs. 5, 6), and some have plaques at their edge (Fig 13). Part of the split valvocopula (Fig 6) overlaps the pseudoseptum (Figs. 10–12), and it has simple pores outside (Fig. 8) and cibrate pores inside (Fig. 11). There are at least two more plain copulae (Fig. 7). Removal of the valvocopula shows the pseudoseptum forms a shelf round the valve edge (Figs. 13 –15). This shelf is supported by short inward-branching ribs (Figs. 14, 15) that appear to be outgrowths of the pseudoseptum. When the pseudoseptum is broken off, the ribs come off with it tearing the valve (Fig. 16). Costae have »shoulders« where they meet the pseudoseptum and only a narrow strap-like neck continues to the edge (Fig. 17); this could well form a connected space under the pseudoseptum. Evidence for this is in Figure 14, where the floor of the right-hand intercostal space (plus arrow) has more highlights than its neighbour to the left. Lighting in the latter is obstructed by a particle under its left rib.

Habitats

Skeletomastus coelatus has been collected from Cook Strait and Stewart Island. The original material on *Ballia* sent to G. A. W. Arnott by W. H. Harvey (ARNOTT 1858) came from the HMS Acheron collections from one of these areas or from Fiordland. It was recorded on coralline algae from Lyall's Bay (now called Lyall Bay), Cook Strait by PETIT (1877). Our material was on *Herposiphonia ceratoclada* from a neighbouring bay (Island Bay). It has been found at Traills Bay, Stewart Island (STIDOLPH pers. comm..) in a mixed sample RM 135. This is preserved on the S.R.S. slides 996–1000 at NIWA (Wellington) and also on British Museum Slide 100875. It is epiphytic on finely-branched red algae.

Pleurosigma

This *Pleurosigma* species is unlike other *Pleurosigma* species with naviculoid symmetry so we erect a new species for it.

Pleurosigma inscriptura M. A. Harper, sp. nov.

Latin diagnosis: *Valvae lanceolatae polis rotundatis, ambito respectu axem apicalem transapicalemve symmetrico, 70–110 µm longae, 16–21 µm latae. Striae decussatae, striis obliquis ad angulum c. 70° intersecantis et areolis 16–20 in 10 µm praeditis, striis transapicalibus 20–23 in 10 µm. Raphe sigmoidea, ad centrum respectu lineam inter nodulos polares angulo c. 3° inclinata.*

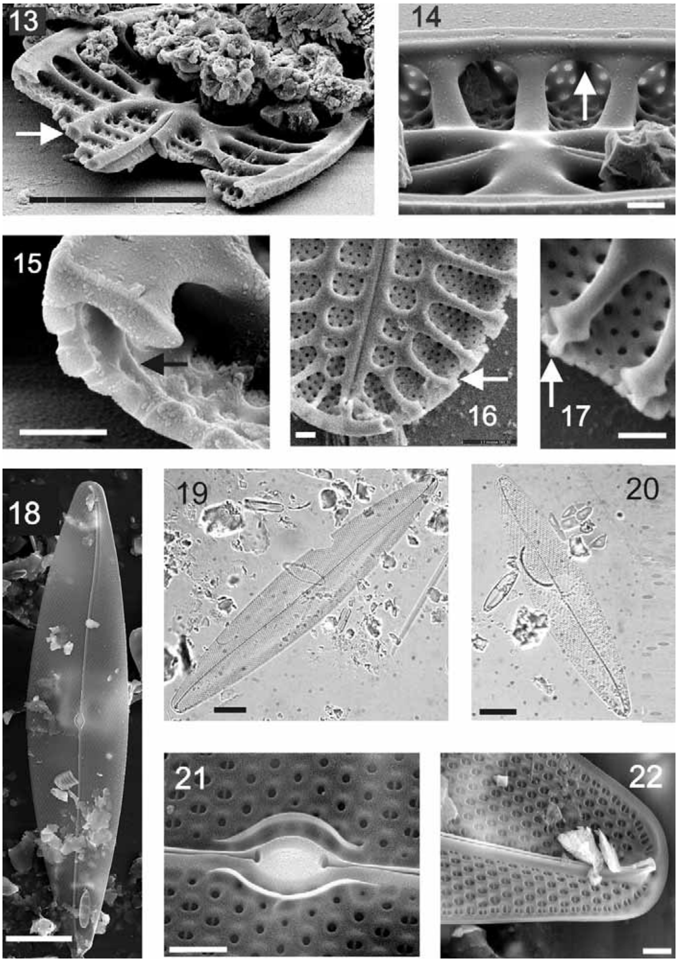

Plate 2. Skeletomastus coelatus [*Cocconeis coelata*] in SEM (Figs. 13–17), *Pleurosigma inscriptura* in LM (Figs. 19–20), in SEM (Figs. 18, 20–21). Fig. 13 – oblique view of broken valve (arrow indicates solid lyrae); Fig.14 – view across centre (arrow indicates short rib); Fig. 15 – detail of figure 13 arrow points to branched marginal rib; Fig. 16 – broken pseudoseptum (arrow shows tear in valve made by lost rib); Fig.17 – detail of broken valve (arrow indicates end of costa); Fig. 18 – SEM internal view of *P. inscriptura* valve; Fig. 19 – large specimen; Fig. 20 – smaller specimen; Fig. 21 – internal view, central nodule; Fig. 22 – internal view of apex. Scale bars: 10 µm (Figs. 13, 18–20), 1 µm (Figs. 14–17, 20, 21).

Valves lanceolate with rounded poles, symmetrical in outline with respect to the apical and transapical axes, 70–110 µm long, 16–21 µm wide. Striae decussate, with the oblique striae intersecting at an angle of c. 70° and provided with 16–20 areolae in 10 µm, 20–23 transapical striae in 10 µm. Raphe sigmoid, inclined at the centre by an angle of c. 3° with respect to a line drawn between the polar nodules.

Locus typicus: Island Bay, Wellington, New Zealand.

Holotype: *Pleurosigma inscriptura* slide CHR 593684 Allan Herbarium, Lincoln, New Zealand.

Isotypes: Slide 939 – *Pleurosigma inscriptura* NIWA Herbarium, National Institute of Water and Atmospheric Research, Riccarton, New Zealand.

Etymology: Named for the appearance of its sigmoid raphe, as if handwritten.

The lanceolate valves (Figs. 18–20) are so symmetrical that the outline of flat-lying valves can be flipped about either the apical or transapical axis and will still coincide with the original, unlike sigmoid valves (which have one pervalvular axis of symmetry). Most naviculoid *Pleurosigma* species are slightly sigmoid in outline (ROUND et al. 1990); some examples are listed in table 1**.** *Pleurosigma inscriptura* valves range from 70–110 µm (based on 22 valves) with a length to width ratio of c. 5 (range 4.2 to 6.3). The oblique striae have c. 18.5 areolae in 10 µm (range 16 to 20). In *Pleurosigma* the areolar centres alternate between lying on the apical axis and to one side of it; both sets form part of transverse lines of striae and all lines are included in estimates of striae frequency. Transverse striae have a mean frequency of 22 striae in 10 µm (range 19.5 to 23). The ratios of counts on transverse striae to oblique striae have a mean of 1.18 (range 1.16 to 1.22). The mean angle of intersection for oblique striae is 69° (range 66° to 73°). The raphe system is clearly sigmoid near its centre as well as at its ends, unlike many other naviculoid *Pleurosigma* species. This results in a larger raphe angle (ca 3° to the line joining its polar ends) and its ends are more deflected. In LM the central raphe endings appear joined and deflected in the same direction. The polar raphe ends appear like small triangles (STERRENBURG 1991a).

Details of the valve interior in SEM (Figs. 21, 22) show that most areolae open internally by double poroids and are »obex« (STIDOLPH 1992) without rims, but a few are single near the centre (Fig. 21). The centre has no hyaline areas outside a small oval central nodule

Tab. 1. Characteristics of *P. inscriptura* compared with some other symmetrical *Pleurosigma* species with clearly sigmoid raphes.

Characteristic	*P. Inscriptura* nov sp.	*P. strigosum* W. Smith	*P. Rigidum* W. Smith	*P. Williamsii* Reid 2002	*P. Obtusum* Mann
Darkfield	bright teal	bright blue	indigo	unknown	none
Oblique angle	65–73°	64–70°	52–57°	c.50°	68°
"striae / 10 µm	17–20	16–18	17–21	18–25	14, 20
Transverse " / "	19.5–23	17–20	16–20	18–25	22
Raphe angle	2.5–4°	6–10°	4–8°	c.5°	c. 12°
" central ends	close	small	close	close	deflected
Terminal areas	triangle	small	triangle	funnel	(obscured)
Shape	lanceolate	lanceolate	lanceolate	lanceolate	rhombic
Symmetry	3 axes	3 axes	< 3 axes	3 axes	< 3 axes
Length µm	70–132	120–200	200–360	90–320	126, 131
Width µm	16–21	28–37	38–45	15–38	29, 30
Reference	this paper	STERRENBURG 2003	STERRENBURG 2001	REID 2002	STIDOLPH 2002

and its ridges (Fig. 21). Raphe slits lie between two equally thickened ribs, their internal central ends are rounded with slight deflections to the same side. The apical raphe ends have helictoglossae that are turned to opposite sides (Fig. 22).

Habitats

Most valves of the new *P. inscriptura* were collected with red algae from Wellington's South Coast. Some came from *Herposiphonia ceratoclada* growing subtidally on the brown alga *Halopteris,* but they also occurred occasionally on *Gigartina circumcincta* from Island Bay, and on a coralline alga (cf. *Arthocardia* sp.) collected at low spring tide from Owhiro Bay (next to Island Bay). *Pleurosigma inscriptura* also occurred among epiphytic diatoms collected from Titahi Bay (COCHRAN 2002: slide MTB-1EP). *Pleurosigma stidolphii* was also present in some samples.

Tab. 2. Comparison of spectral colour wavelengths with mean spacing of oblique striae and mean angles in three main colour groups and with *P. inscriptura*.

Colour	Wavelength nm	Oblique striae per 10 µm. mean (range)	Spacing nm (difference)	Oblique angle mean (range means)
Indigo (5 spp.)	425–450	19.6 (18–21.3)	510 (70)	57.5° (55–63)
Blues (11 spp.)	450–500	19.3 (16–25)	518 (40)	67.6° (60–81)
P. inscriptura		18.3 (17–20)	546 (36)	68.6° (65–73)
Greens (0 spp.)	500–570			
Gold (5 spp.)	570–590	12.6 (10–18)	790 (200)	90.8° (80–100)

Discussion

Skeletomastus

Early workers (SCHMIDT 1894) realised *»Cocconeis coelata«* was biraphid, so not a *Cocconeis*. Initially we interpreted our LM views of *Skeletomastus coelatus* as being those of a species of *Mastogloia*. These have raphe slits that undulate but in the same direction (PADDOCK and KEMP 1990), can be multiseriate and may have a solid lyre; but always have a loculate valvocopula (COX 2006). SEM views show *Skeletomastus coelatus* has no valvocopular chambers, rather a pseudoseptum supported by short marginal ribs. Comparison with other genera and species show its cibra are like those of *Craspedostauros britannicus* (COX 1999), but positioned as in *M. danseii*; its valvocopula split overlaps the pseudoseptum as in *Mastogloia danseii* (COX 2006) and its pseudoseptum is continuous as in *Achnanthes coarctata.* These are all members of the natural group Mastogloiales (COX 2006, COX and WILLIAMS 2006), which is largely defined by the presence of the two fore and aft H-shaped plastids such taxa possess. We have not seen the plastids of *S. coelatus.* Lyrate genera outside this group differ from *S. coelatus*: *Fallacia* has a hollow lyre, *Diploneis* has channels next to the raphe and *Lyrella* does not have costae (thickened interstriae) and has internal siliceous flaps on its pores. Although *S. coelatus* appears to fit in the Mastogloiales, it does not fit easily in any genus: unlike *Aneumastus* it has no flaps on external pores*,* nor does it have a stauros-like *Craspedostauros,* nor is it monoraphid like *Achnanthes* (COX 2006). Therefore we have erected a new genus *Skeletomastus* for it. It has most in common with *A. aksaraiensis* Spaulding, Akbulut and Kociolek which has biseriate striae with round pores except next to the raphe, connections between costae forming a zig-zag lyre and terminal pseudosepta (Spaulding et al. 2003). Spaulding et al. (2003) suggest *A. akaraiensis* is either basal to *Mastogloia* or derived from it – this could well be true of *Skeletomastus*.

Pleurosigma inscriptura

This species differs from most *Pleurosigma* species in combining a naviculoid valve with a clearly sigmoid raphe; otherwise its features are typical of *Pleurosigma* in general (ROUND et al. 1990, REID 2002). Its raphe and oblique angle, terminal areas and darkfield colour are the characteristics (STERRENBURG 1991a), rather than dimensions and stria frequency, that distinguish it from the other species (Tab. 1). The bright greenish-blue colour seen in standard darkfield is typical of *Pleurosigma* species with striae of similar frequency and oblique striae at an angle of 60–70° (Tab. 2). This angle is a more sensitive parameter (STERRENBURG 1991a) than the ratios. Theoretical ratios for angles between oblique striae versus oblique pore-spacing (see Methods) give a more sensitive measure of differences between species than the ratio of frequency of transverse and oblique striae**.**

The external pores of *Pleurosigma* species are slits and their darkfield colours are mainly due to diffraction through these. Mean oblique stria spacing is approximately related to observed colour (Tab. 2). Other factors that could affect this are whether slits overlap, type of internal pores, variable spacing in different parts of the valve and other properties of pores and walls.

Acknowledgements

The authors thank David Mann of the Royal Botanic Garden, Edinburgh, for useful comments and the Latin diagnoses, Ursula Cochran of GNS Science, Wellington, for editing the manuscript, David Flynn of the Electron Microscopy Unit, Science Faculty, Victoria University of Wellington, for scanning electron micrographs, Stuart Stidolph for information and Frithjof Sterrenburg, Sijbekarspel, Netherlands for help with the taxonomy of *Pleurosigma* and Phil Garnock-Jones of the School of Biological Sciences, Victoria University of Wellington for help with the synonymy of *Skeletomastus coelatus*.

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