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REGULAR PAPER

Two closely-related species of *Cocconeis* (Bacillariophyta): comparative study and typification

Oscar E. Romero[1,*] & Catherine Riaux-Gobin[2,3]

[1]MARUM, Center for Marine Environmental Sciences, University Bremen, Leobener Str., 28359 Bremen, Germany
[2]Centre de Recherche Insulaire et Observatoire de l'Environnement (CRIOBE) URS 3278–CNRS/EPHE/Université de Perpignan, Via Domitia, 52 Avenue Paul Alduy, 66860 Perpignan cedex, France
[3]Laboratoire d'Excellence 'CORAIL', Perpignan University, 58 Avenue Paul Alduy, 66860 Perpignan cedex, France
*Author for correspondence: oromero@uni-bremen.de

Background and aims – After its original description, *Cocconeis grata* A.W.F.Schmidt and *Cocconeis pseudograta* Hust. have been rarely quoted and illustrated. Due to the morphological resemblance between both taxa and because of their rare quotations, a certain degree of taxonomic confusion existed between both diatoms. In order to clarify the taxonomic history of these two taxa, we studied their type materials kept at the Hustedt Collection (Bremerhaven, Germany), and specimens of *C. pseudograta* recently collected in coastal waters of the Tahiti Island (Society Archipelago, southern tropical Pacific Ocean).
Methods – Light and electron microscopy observations as well as morphometric data of frustules are presented, and an amended description for each taxon is provided.
Key results – In addition to differences in stria and areola density, following frustule features allow to distinguish between both taxa: sternum valves (SV) areolae outline and occurrence of external depressions on the SV of *C. grata* and granules on the external valve face and marginal row of small processes on the SV mantle of *C. pseudograta*.
Conclusions – Based on the frustule morphology and morphometric data, we conclude that *C. grata* and *C. pseudograta* are independent species. While the former can still be considered 'American' in its distribution, the occurrence of *C. pseudograta* in coastal waters of the Tahiti Island invalids its previous designation as 'European form'.

Key words – Benthic marine diatoms, biogeography, *Cocconeis*, typification, tropical Pacific.

INTRODUCTION

The most common approach to the identification of taxa of *Cocconeis* Ehrenb. follows the current concepts available in the literature, with scarce reference to the corresponding type material. Although numerous peer-reviewed articles have described *Cocconeis* taxa from different world regions in the past 20 years (e.g. Romero & Rivera 1996, Suzuki et al. 2001, Riaux-Gobin & Romero 2003, De Stefano & Romero 2005, De Stefano et al. 2008, Romero & López-Fuerte 2013), the study of type material has started recently (Jahn et al. 2009, Romero 2011, Romero & Jahn 2013, Riaux-Gobin et al. 2014). Notably, the scarcity of type material revisions concerns species and varieties of *Cocconeis* described in the late 19th and the earliest 20th centuries.

As in many other diatom genera, the morphological variability of frustules of *Cocconeis* from geographically distant populations can lead to misidentifications. This, in turn, can lead to confusions in the taxonomic history of morphologically similar taxa. Such a situation applies to *Cocconeis grata* A.W.F.Schmidt and *Cocconeis pseudograta* Hust. The resemblance between both taxa (Hustedt 1933, 1939) and the scarcity of findings since their original description has led to a certain degree of taxonomic confusion.

Taxonomic history of *Cocconeis grata* and *Cocconeis pseudograta*

Based on materials collected in the Bay of Campeche (Mexican Caribbean Sea), *Cocconeis grata* was first illustrated by A.W.F. Schmidt (in Schmidt et al. 1874–1959 [1894]: plate 192, fig. 65; here reproduced in fig. 1A & B). Shortly afterward, Cleve (1895: 172, plate 2, figs 30–31; here reproduced in fig. 1C & D) published the first description of *C. grata*: valves with elliptic-lanceolate outline, measuring between 40 and 60 µm length and 30 and 44 µm width. The sternum valve (hereon SV, 'upper valve' according to Cleve 1895) has a broad lanceolate axial area and between 10 and 11 ra-

dial transapical striae in 10 µm, build by large areolae. The raphe-sternum valve (hereon RSV, 'lower valve' according to Cleve 1895) has an 'annulus of rudimentary loculi' and 15 striae in 10 µm, finely punctuated and radiate, which become fainter towards the raphe ('median line'). In their short description of *C. grata*, Peragallo & Peragallo (1897–1908: plate 3, figs 6–7, here reproduced in fig. 1E & F) gave identical morphometric data as in Cleve (1895), also mentioning the possible presence of rudimentary loculi. The drawings of *C. grata* by Cleve and by Peragallo & Peragallo show some resemblance with each other (fig. 1C & D, and 1E & F), but in turn differ from those by Schmidt (fig. 1A & B), mainly considering the SV.

Found in the same material with *C. grata*, Schmidt also illustrated *Cocconeis campechiana* P.T.Cleve (in Schmidt et al. 1874–1959 [1894]: plate 190, fig. 36). Cleve (1895) synonymized *C. campechiana* with *C. grata*, without any further discussion. Peragallo & Peragallo (1897–1908) followed Cleve's species concept and considered *C. campechiana* to be a synonym of *C. grata* as well.

In his account of diatoms from Northern Europe, Hustedt (1933: 342, fig. 795) characterized *C. grata* (here reproduced in fig. 1I–K). Hustedt (1933) adopted the description of *C. grata* given by Cleve (1895) and Peragallo & Peragallo (1897–1908) for valves found in samples collected in the Mediterranean Sea and the North Atlantic. His decision based mainly on (1) the resemblance of the existing drawings of *C. grata* and (2) his unsuccessful search of *C. grata* in samples from the Bay of Campeche. The drawings of *C. grata* by Hustedt (1933) were more elaborate than previous ones and his description offered more detailed information of the morphology and structure of the valves, this including the presence of a 'lanceolate central field (in the SV), which occupies almost 1/3 of the valve width'. The RSV of Hustedt's *C. grata* has a straight raphe with closely-located proximal raphe endings and an oblong central area (fig. 1I). Hustedt also provided striae density for both valves (SV = 9–11 in 10 µm; RSV = 20–22 in 10 µm).

A few years later, Hustedt found one unique frustule of *C. grata* in the Mexican Caribbean material and presented a detailed diagnosis and drawings of both valves (Hustedt 1939: 605, figs 21–22; here reproduced in fig. 1G & H). He corrected his identification of *C. grata* (as in Hustedt 1933), which he renamed as *Cocconeis pseudograta* nom. nov. and called it afterward the 'European form' (in opposition to *C. grata*, the 'European form', Hustedt 1939). As properly noted by Simonsen (1987), *C. pseudograta* was a new species and not a 'nom. nov.'. Hustedt (1939) critically commented on the incomplete characterization of *C. grata* by Schmidt (Schmidt et al. 1874–1959) and also questioned the drawings presented by Cleve (1895; fig. 1C & D). He argued that two main valve features allow to distinguish *C. grata* from *C. pseudograta*: (1) the structure of the SV, and (2) a more delicate structure and a very small central area of the RSV of *C. grata* (Hustedt 1939). Hustedt's (1939) figures of the Bay of Campeche specimens (fig. 1G & H) differ, in turn, from Schmidt's drawings (fig. 1A & B).

In this study, we present light and electron microscopy observations of the type material of *Cocconeis grata* and *Cocconeis pseudograta*. In addition to the study of the type materials, observations on the valve morphology of *C. pseudograta* populations collected in the tropical Pacific Ocean are also presented.

MATERIAL AND METHODS

In this work, raw materials and permanent slides housed at the Hustedt Collection (Alfred-Wegener-Institute, Bremerhaven, Germany) were investigated (table 1). In addition to the type material, several marine to brackish sites of Arué District (Tahiti Island, Society Archipelago, tropical southern Pacific Ocean) were sampled in October 23rd–24th 2010 by C. R.-G. In particular, intertidal sediments from a coral reef lagoon and scrapes of one large specimen of *Holothuria atra* Jaeger (Black Sea cucumber) were collected (sample 'Papeete 4', 17°31.431'S 149°31.233'W; marine environment of Pointe Honu, near 'Pomare Tomb', collection C. R.-G., USR 3278, CRIOBE-Perpignan University, Perpignan, France.) The Tahiti samples were preserved in formalin (10% final dilution.) Temperature and salinity were measured *in situ* using a SP 536 portable conductivity meter. Salinity ranged between 32.2 and 33.8 psu. Mean surface temperature was 27.3°C.

For this study, samples from Tahiti Island were repeatedly rinsed with distilled water, then treated with concentrated H_2O_2 to remove organic matter, and rinsed repeatedly with distilled water, alcohol-desiccated and mounted in Naphrax® to make permanent slides. The slides were observed using a Zeiss Axiophot 200 microscope with differential interference contrast (DIC) and photographed with a Canon PowerShot G6 digital camera (CRIOBE-Perpignan University, France.)

For scanning electron microscopy (SEM) examination, preserved field samples were collected on 1 µm Nuclepore filters and rinsed twice with deionized water (milliQ) to remove salt. The filters were air-dried and mounted onto aluminum stubs before coating with gold palladium alloy (EMSCOP SC 500 apparatus) and examined with a SEM Hitachi S-4500 operated at 5 kV (Perpignan University Via Domitia, France), and SEM FEi Quanta Feg 200 (AWI, Bremerhaven, Germany).

Table 1 – Materials of *Cocconeis grata* A.W.F.Schmidt and *C. pseudograta* Hust.; kept in the Hustedt Collection (Bremerhaven, Germany) and studied in this work.

taxon	sample	slide	sampling location
Cocconeis grata	AM63A/C	N12/85	Bay of Campeche, Caribbean, Sea, Mexico
	AM791	ZT1/85	Beach, Beaufort, North Carolina, U.S.A.
Cocconeis pseudograta	E255	ZT4/62	Grip, Norway
	none	M2/99	Adria, San Pietro di Nembi

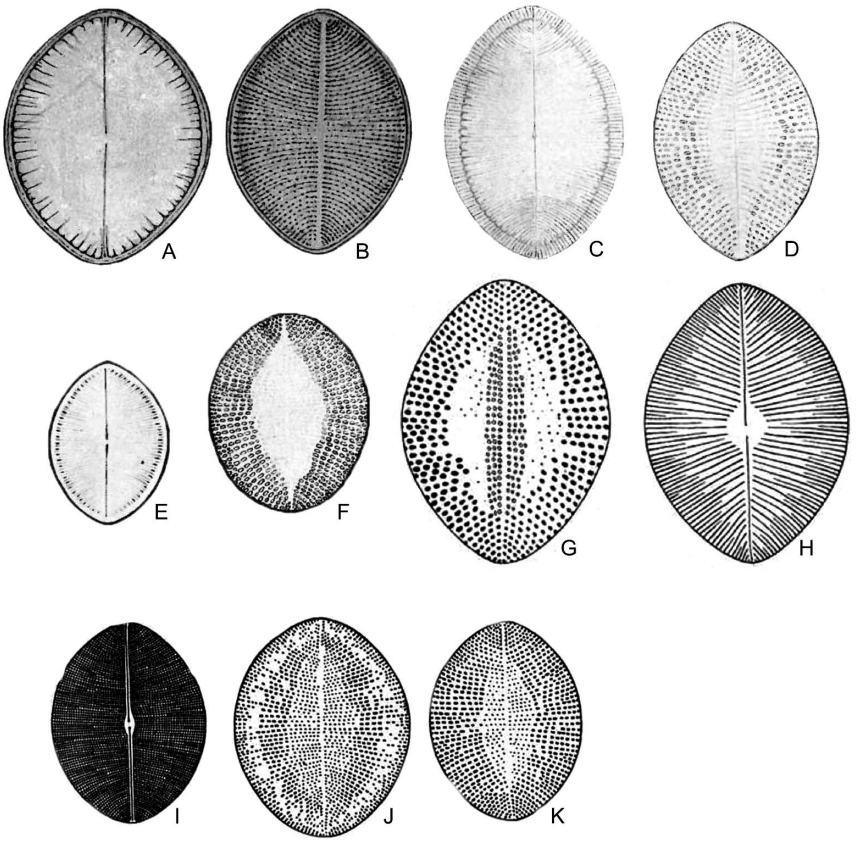

Figure 1 – Drawings of *Cocconeis grata* and *Cocconeis pseudograta*. A & B, *Cocconeis grata*, Schmidt in Schmidt et al. (1874–1959 [1894], plate 192, fig. 65); C & D, *Cocconeis grata* (Cleve 1895); E & F, *Cocconeis grata* (Peragallo & Peragallo 1897–1908); G & H, *Cocconeis grata* (Hustedt 1939); I–K, *Cocconeis pseudograta* (as *Cocconeis grata*, Hustedt 1933).

The terminology recommended in Anonymous (1975), Ross et al. (1979) and Round et al. (1990) is used for the frustule description. For suprageneric taxa, the classification proposed by Round et al. (1990) was followed. Throughout the text, the sternum and raphe–sternum valves are named SV and RSV, respectively (De Stefano & Romero 2005, De Stefano et al. 2008, Romero & Jahn 2013, Romero & López-Fuerte 2013). In the species description, AA is used for the apical axis and TA for the transapical axis. The striae density was counted at the center of the valve face along the axial axis and also at the margin opposite to the valve center.

OBSERVATIONS

Order: Achnanthales P.C.Silva
Family: Cocconeidaceae Kütz.
Genus: *Cocconeis* Ehrenb.

Cocconeis grata A.W.F.Schmidt
Figs 1A–H & 2–3, table 2

Protologue – Schmidt et al. 1874–1959 [1894]: plate 190, fig. 36; plate 192, fig. 65.

Synonyms – *Cocconeis grata* var. *nummularia* (Grev.) Perag. & H.Perag. (Peragallo & Peragallo 1897–1908: 16); non *Cocconeis nummularia* (Grev.) Perag. & H.Perag. (Peragallo & Peragallo 1897–1908: 3/8–9).

Type: figures numbered 2A, 2E–F. – Type locality: Bay of Campeche, Caribbean Sea, Mexico (no particular sampling location known, see below 'Occurrence and Biogeography of *C. grata* and *C. pseudograta*').

Description of *Cocconeis grata* (type material) – Dimension: AA, 34.8–44 µm; TA, 28.2–33.3 µm. Morphology: elliptic valve outline (figs 2, 3A, C, G & H). Externally, the SV is concave along the sternum and the closest neighboring area, and convex mid-way between the sternum and the *crista marginalis* (fig. 3C). The linear sternum does not cross the junction between the valve face and the *crista marginalis* at the apices (fig. 3C). The radiate striae (9–10 in 10 µm on the margin and 7–8 in 10 µm along the axial area) are throughout uniseriate, and they are separated by wide interstriae (fig. 2A & B, fig. 3A & F). Some shorter (between one and four areolae), intercalary striae occur every second to sixth longer striae (fig. 3B & G arrowed). The areolae are poroids (fig. 3D & F), mostly transapically elongated on the external valve face (fig. 3B, E & G). Close to the sternum, some depressions on the external valve face resemble –on its outline– regular areolae; however, these depressions do not go through the valve face (fig. 3C & G). Under the light microscope, they can be distinguished as being smaller and less reflective than regular areolae (fig. 2B). The *crista marginalis* separates the valve face from a very steep margin (fig. 3C & D). On the internal side, an elliptic solid plate of variable width is observed, and

whose outline gradually decreases from the valve middle toward the apices (fig. 3E & G). Valvocopula possibly closed; long filiform fimbriae present (fig. 2H & I).

Externally, the RSV face seems convex along the sternum, while gradually concave halfway between the sternum and the valve margin (fig. 3H). The straight raphe has slightly expanded external proximal endings, which converge in a small, slightly asymmetrical, rounded central area. The distal raphe endings are externally straight (fig. 3I). The striae (18–22 in 10 µm along the axial area and 20–24 in 10 µm at the valve margin) are uniseriate (fig. 3H–J). No observations on the internal valve side. Valvocopula possibly with long fimbriae (fig. 2H–I).

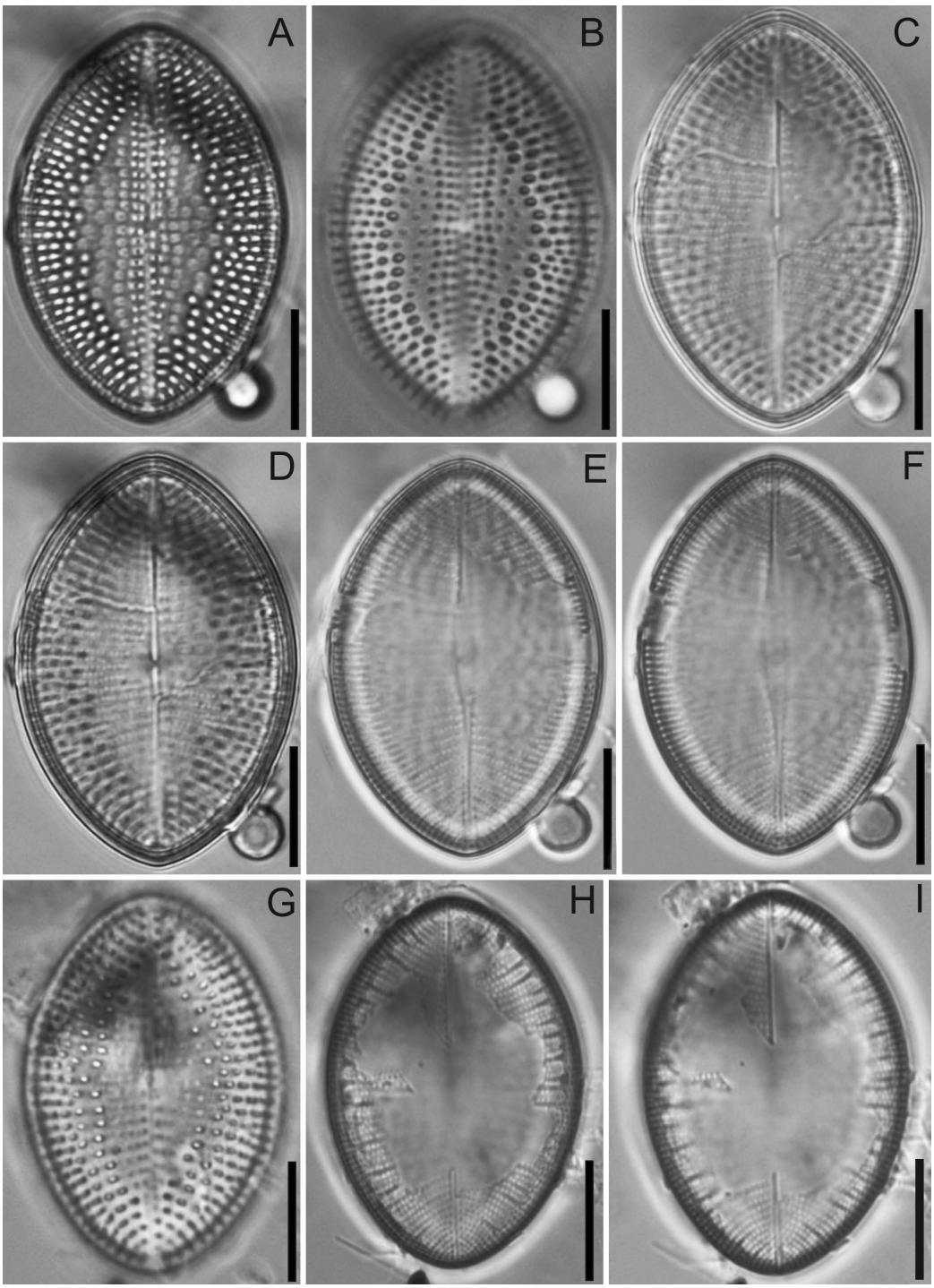

Figure 2 – *Cocconeis grata* from type slides (see table 1), light microscopy: A–F, different foci on one frustule; A & B, focus on SV (A: margin; B: focus on sternum and adjacent area); C & D, different foci on SV and RSV; E & F, different foci on the same RSV (E, raphe area; F: marginal area of RSV); G–I, different foci on the only found frustule. G, focus on SV; H, J: focus on RSV. Scale bars = 10 µm.

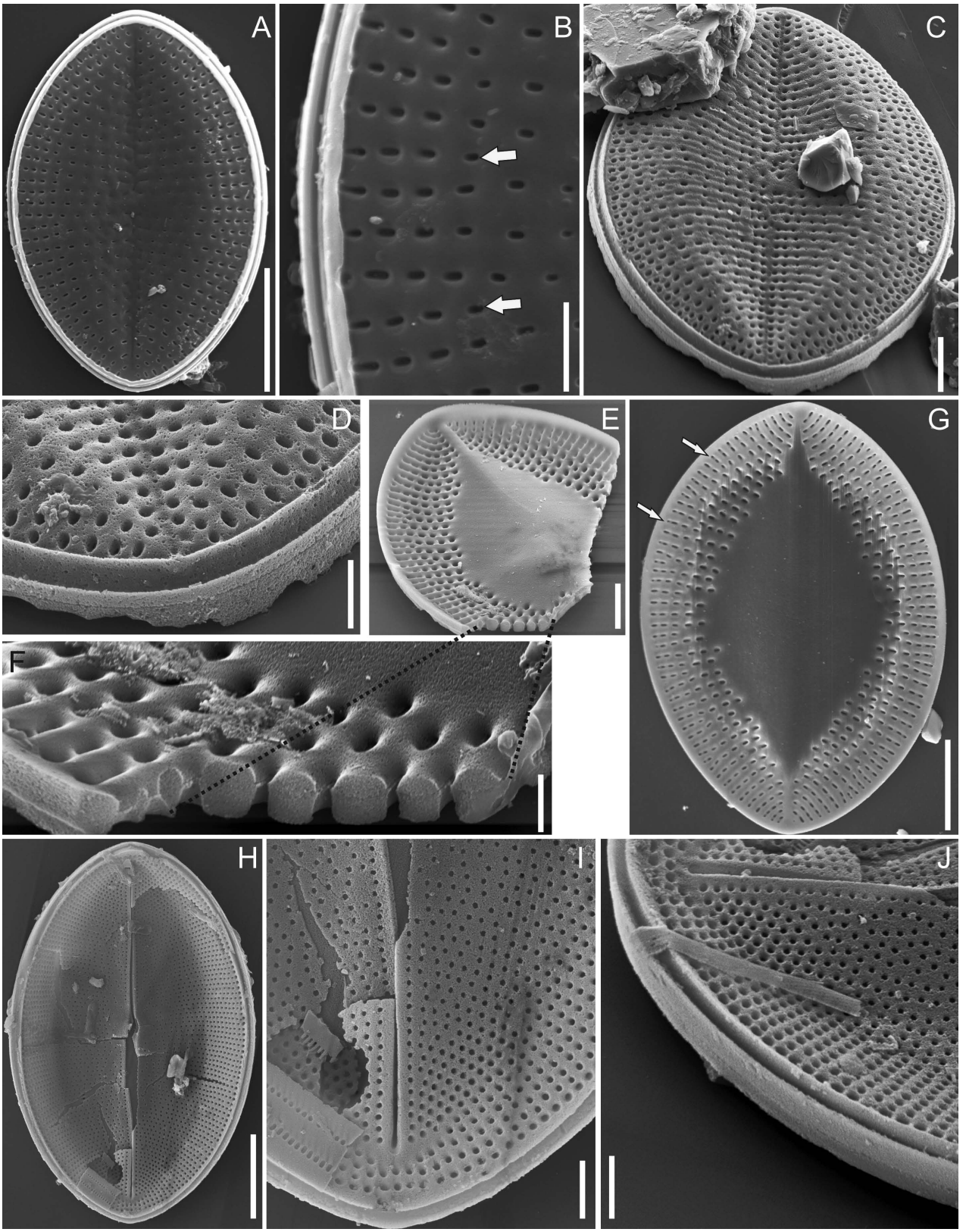

Figure 3 – *Cocconeis grata* from type materials (see table 1), scanning electron microscopy: A–G, SV; A, SV, external view; B, detail of A, left-hand side of SV. Arrows indicate short, intercalary striae; C, SV, external view of 29.2° tilted SV; D, SV, detail of C, valve end; E, SV, interval view, broken valve; F, detail of E; G, SV, internal view. Arrows indicate short, intercalary striae; H–J, RSV, external view; H, entire valve; I, detail of lower end; J, detail of upper end and margin in H. Scale bars: A, G & H = 10 µm; C & E = 5 µm; B, D & I = 2 µm; F & J = 1 µm.

Table 2 – Comparison of morphometry and morphological features of *Cocconeis grata* and *C. pseudograta* from the literature and from our own observations.
References: no obs.: no observations; (1): Schmidt et al. (1874–1959); (2): Cleve (1895); (3): Peragallo & Peragallo (1897–1908); (4) Hustedt (1939); (5): this study; (6) Hustedt (1933).
*: originally described as *Cocconeis grata* A.Schmidt (Hustedt 1933).

Valve features	*Cocconeis grata* A.W.F.Schmidt (original description)	*Cocconeis grata* A.W.F.Schmidt	*Cocconeis grata* A.W.F.Schmidt	*Cocconeis grata* A.W.F.Schmidt	*Cocconeis pseudograta** Hust.	*Cocconeis pseudograta* Hust. (type material)	*Cocconeis pseudograta* Hust. (Tahiti specimens)
References	(1)	(2, 3)	(4)	(5)	(6)	(5)	(5)
apical axis (µm)		40–60	35–60	34.8–44	40–60	25.6–37.9	14.5–33.9
transapical axis (µm)		30–44	27–47	28.2–33.3	30–44	19.6–27.8	9.5–25.2
valve outline	elliptic to subrhombical	elliptic or elliptic-lanceolate	elliptic	elliptic	elliptic-lanceolate	oblong-elliptic	oblong-elliptic
sternum valve							
sternum	linear	narrow linear	narrow linear, broad in the valve centre	linear	narrow	linear-lanceolate, widens towards the central part	narrow
central area		large, irregularly lanceolate	large rhombic-elliptic	large rhombic-elliptic	rhombic-elliptic	rhombic-elliptic	rhombic-elliptic
striae density (in 10 µm)		10–11 (margin)	10 (margin)	7–8 (axial area) 9–10 (margin)	~ 9–11	14–16 (margin)	15–18 (margin)
striae areolation	uniseriate	uniseriate	uni-triseriate	uniseriate	uniseriate	uniseriate	uniseriate
areola occlusion	no obs.	no obs.	no obs.	no obs.	no obs.	external	hymenes with linear perforations
valvocopula type	no obs.	no obs.	no obs.	possibly closed	no obs.	open in one end	open
fimbriae	no obs.	no obs.	no obs.	long filiform	no obs.	short undulations	absent
raphe-sternum valve							
raphe profile	linear	straight	linear	linear	linear	linear or slightly sigmoid	linear
central area	small, oval	small	small, oval	small, asymmetrical, oval	small	wide, elliptic to subcircular	wide, elliptic to subcircular
striae density (in 10 µm)	no obs.	15 (margin)	14–16 (margin)	20–24 (margin) 18–22 (axial area)	20–22 (margin)	16–18 (sternum) 20–24 (margin)	45–50 (sternum) 28–32 (margin)
striae areolation	uniseriate	uniseriate	uniseriate	uniseriate	uniseriate	uniseriate	uniseriate
areola outline	no obs.	no obs.	no obs.	circular	circular	circular to subrectangular	circular to subrectangular
type of occlusion	no obs.	no obs.	centric array hymenes	no obs.	no obs.	centric array hymenes	no obs.
valvocopula type	no obs.	no obs.	no obs.	no obs.	no obs.	open in one end	no obs.
fimbriae	no obs.	no obs.	no obs.	possibly long	no obs.	no obs.	absent?

***Cocconeis pseudograta* Hust.**
Figs 1I–K & 4–7, table 2

Protologue – Hustedt 1933: 341–342, fig. 795 under the name *C. grata*.

Neotype: Simonsen (1987; plate 375, fig. 9–14) proposed slide Zt4/62, Grip, Norway 1, as the neotype (also studied here, table 1). Specimens from slide Zt4/62 are shown in fig. 4A–C. – Neotype locality: Grip, Norway (no particular location was listed nor coordinates were given) (see discussion below in 'Occurrence and Biogeography of *C. grata* and *C. pseudograta*').

Description of *Cocconeis pseudograta* (type material) – Dimension: AA, 25.6–37.9 µm; TA, 19.6–27.8 µm. Morphology: oblong-elliptic valve outline (figs 4, 5A, B & I).

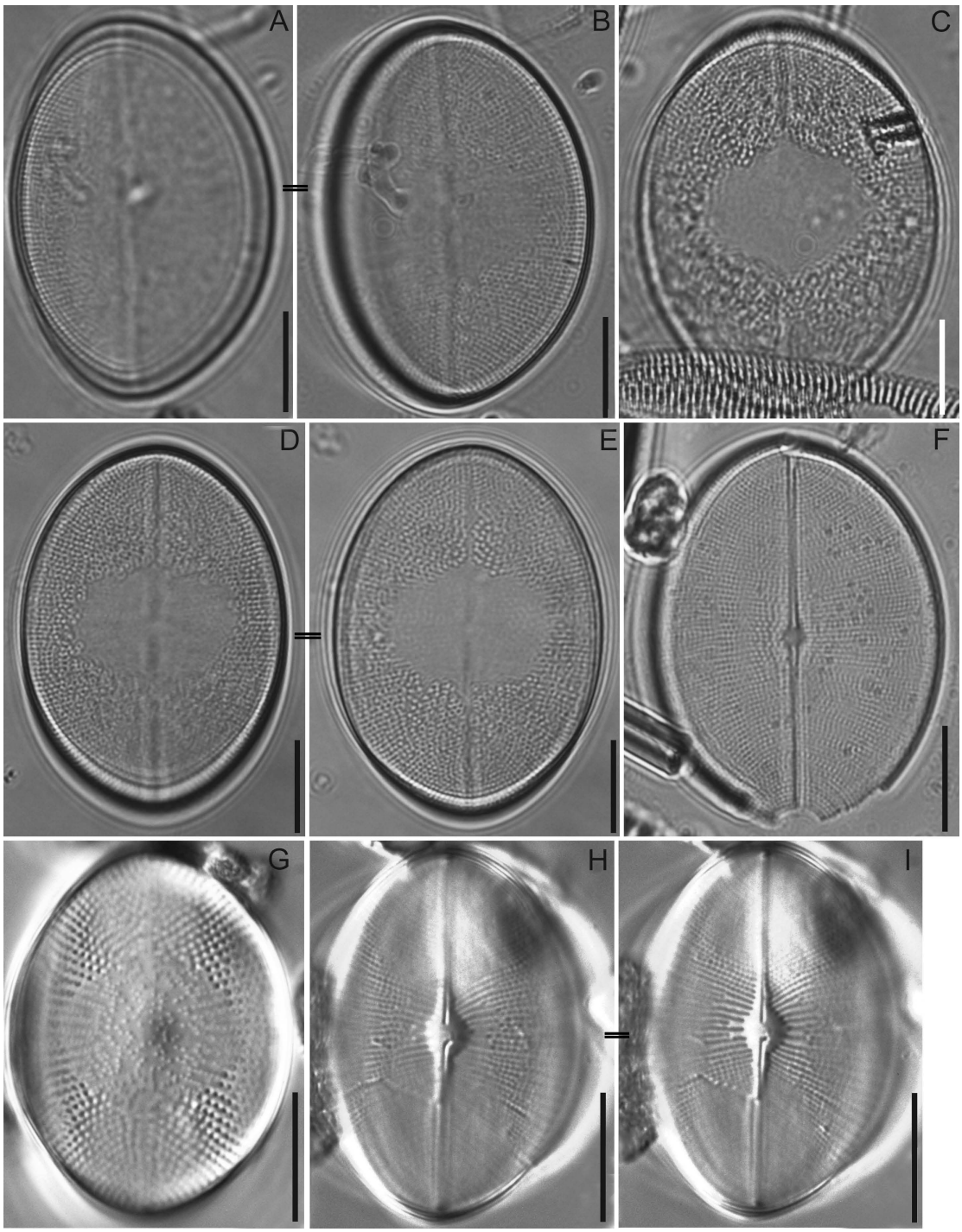

Figure 4 – *Cocconeis pseudograta* from neotype slides (A–F; see table 1) and Tahiti Island specimens (G–I), light microscopy: A & B, different foci on the same SV; C, SV; D & E, different focus on the same SV; F, RSV; G–I, different foci on the same frustule; G, focus on SV; H & I, focus on RSV. Scale bars = 10 µm.

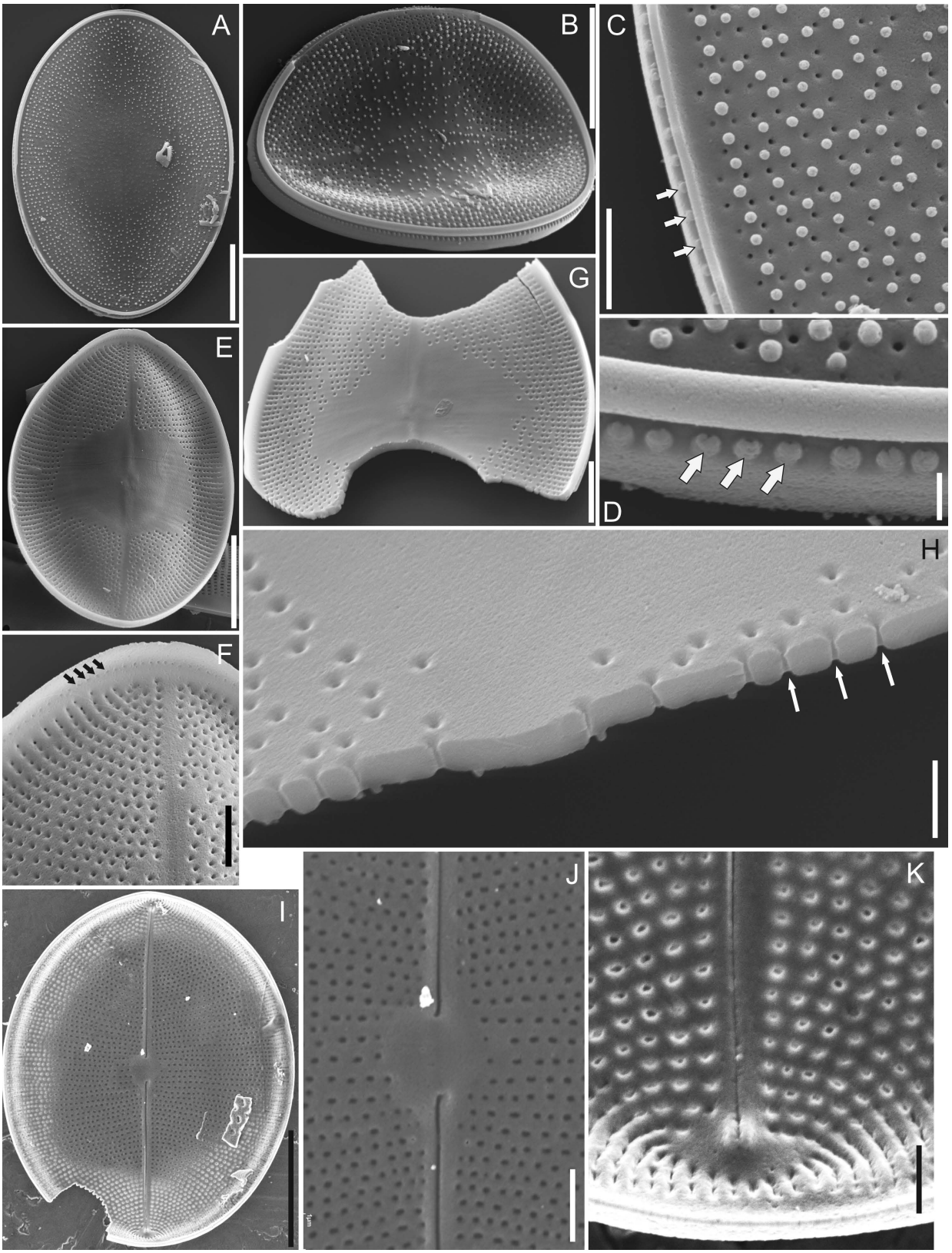

Figure 5 – *Cocconeis pseudograta* from type material (table 1), scanning electron microscopy: A–H, SV; A, external view; B, external view of a 29.3°-tilted valve; C, detail of A. Arrows indicate processes; D, detail of B. Arrows indicate processes; E, internal view; F, detail of E. Arrows indicate the internal openings of processes; G, internal view; H, detail of G. Arrows indicate poroids; I–K, RSV; I, internal view; J, detail of central area; K, detail of distal area and helictoglossa. Scale bars: A, B, E & I = 10 µm; G = 5 µm; C, F, I & J = 2 µm; H & K = 1 µm; D = 0.5 µm.

The SV is externally concave along the sternum and the adjacent area (fig. 5B), rising abruptly toward the low mantle, which is delimited by a *crista marginalis* (fig. 5C & D). The sternum is linear-lanceolate on the two-thirds of the valve close to each end and widens abruptly in the central third (easily recognizable on the internal valve side) (figs 4C–E, 5E). The delicate radiate striae (14–16 in 10 µm at the margin) are uniseriate (fig. 4C–E), slightly curved, consist of a tiny, rounded poroids, and separated by narrow interstriae (fig. 5C, D & H). Close to the *crista marginalis*, some shorter

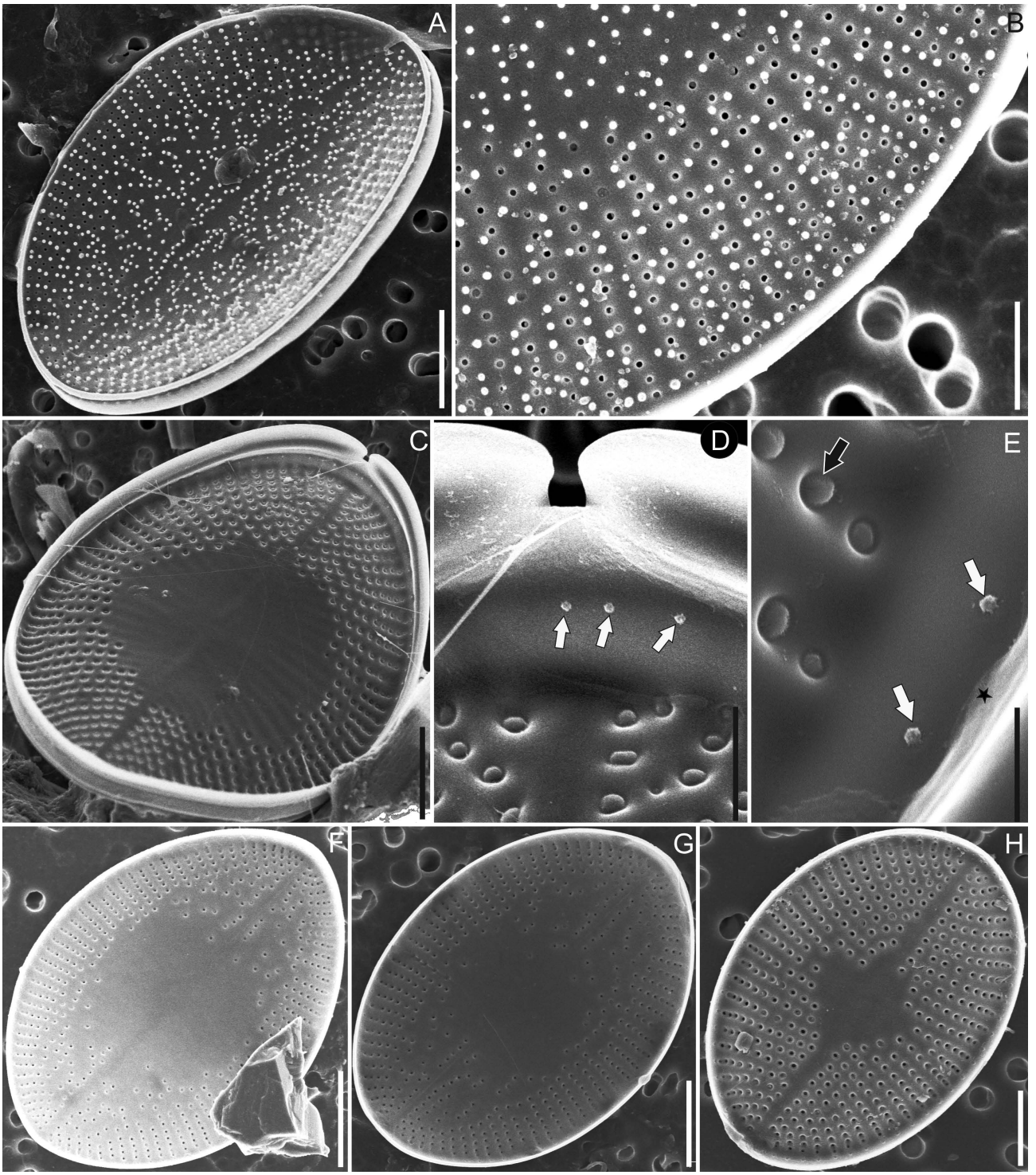

Figure 6 – *Cocconeis pseudograta* (SV, from Tahiti Island), scanning electron microscopy: A, external view; B, detail of A; C, internal view of SV with open valvocopula; D, detail of valvocopula aperture and processes (arrows); E, detail of the internal domed hymenes with marginal slits (black arrow), processes (white arrows) and the undulations of the valvocopula (asterisk); F–H, internal view of SV showing different widths of the central area. Scale bars: A, C, F & G = 5 µm; B & H = 2 µm; D & E = 1 µm.

striae (between one and four areolae) are observed. Numerous small granules are widespread throughout the external valve face, and become less dense (fig. 5B) or are absent in the central area (fig. 5A). On the mantle, small processes are externally topped by an embossed design pierced by a small aperture, while a velum covers the internal side (fig. 5C & F, arrows). Internally, the SV is a solid plate along the sternum and the central, wide hyaline area (fig. 5F & G). No observations on the valvocopula and/or fimbriae.

Only one RSV was found, lying on its internal face. The RSV has a slightly bent raphe and an oblong, asymmetrical central area (fig. 5I & J); distal raphe endings slightly deflect in opposite directions, and terminate in a delicate helictoglossa, which is separated from the valve margin by small hyaline area and a few areolae (fig. 5K). Striae, 16–18 in 10 µm along the axial area and 20–24 in 10 µm at the valve margin, are uniseriate and composed of rounded to subrounded areolae (fig. 5J & K). A few shorter, intercalary striae are observed (fig. 5I). Areolae occlusion not observed. Valvocopula not found.

Tahiti Island specimens – Dimensions: AA, 14.5–33.9 µm; TA, 9.5–25.2 µm. Morphology: oblong-elliptic valve outline (figs 4G–I, 6A & F–H). Externally, the slightly silicified, strongly concave SV resembles (fig. 6A) the neotype SV (fig. 5A & B). The narrow sternum is linear-lanceolate in each uppermost two-thirds of the valve and widens abruptly in the central one-third (fig. 6C). Radiate, uniseriate striae (15–18 in 10 µm on the margin), slightly curved, with tiny, rounded poroids (fig. 6B, D & E). As in the type material, shorter striae are present (fig. 6B). Small, more or less equidistant

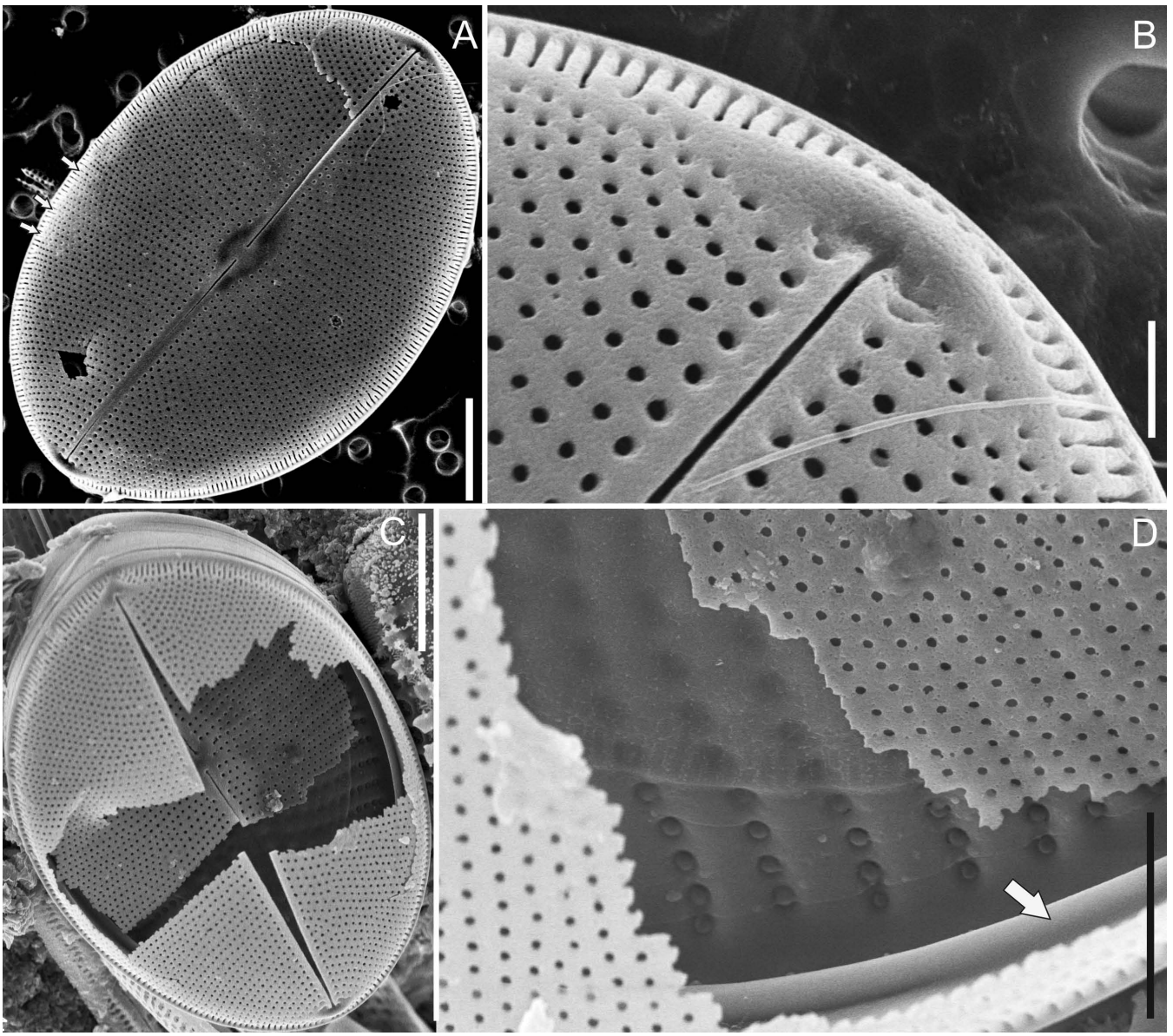

Figure 7 – *Cocconeis pseudograta* (RSV, from Tahiti Island), scanning electron microscopy: A, external view; B, detail of A; C, broken RSV; D, detail of C. Note the internal face of the SV. The arrow indicates the RSV valvocopula (?). Scale bars: A & C = 5 µm; B = 1 µm; D = 2 µm.

granules are arranged in rows following the interstriae, while they are scattered on the central hyaline area (fig. 6A & B). Poroids are internally occluded by domed hymenes with short, marginal slits (fig. 6D & E, black arrow). Processes (fig. 6D & E) as in the type specimens. On the internal side, the valve is a solid plate in the central part (with variable width, fig. 6C, F–H). Robust valvocopula open in one end, with short undulations (fig. 6C, D & E, asterisk).

Externally, the concave RSV has a straight raphe and an oblong, asymmetrical central area (fig. 7A). Distal raphe endings are separated from the valve margin by small semi-lunate hyaline area and one row of elongated areolae (fig. 7B). Striae (28–32 in 10 µm along the margin and 45–50 in 10 µm along the axial area) are uniseriate and composed of rounded areolae (fig. 7A & D). A few shorter (up to four striae), intercalary striae appear every third to sixth longer striae (fig. 7A). Areolae occlusion not observed. Valvocopula possibly with short undulations (fig. 7D, arrow).

DISCUSSION

Our light and electron microscope observations and morphometric data on type material allow confirming that *C. grata* and *C. pseudograta* are two independent species. Below we discuss some taxonomical issues related with each of both taxon, and compare particular frustule features with those of other *Cocconeis* taxa.

Comments on *Cocconeis grata*

In general, our observations conform to the original description and drawings by Hustedt (1939). We exhaustively studied raw samples and permanent slides of the type material and found only scarce valves (mostly SVs). In contrast to Hustedt's morphometric data (table 2), our range of AA and TA of the few found SV and the only observed RSV is lower than originally reported (table 2). A possible explanation for this difference lays in the fact that Hustedt (1939) mainly followed descriptions provided by Cleve (1895) and Peragallo & Peragallo (1897–1908), and possibly adopted the range provided in the late 1890s by Cleve (whose materials we did not study and whose description was consequently followed by Peragallo & Peragallo 1897–1908).

Comments on *Cocconeis pseudograta*

We have studied two raw samples and corresponding permanent slides kept at the Hustedt Collection: samples collected in Norway (Grip) and Croatia (San Pietro dei Nembi, Northern Adriatic Sea) (table 1). Hustedt listed the later location with an Italian name; nowadays, however, it is a Croatian locality in Ilovik (44º28'N 14º33'E). As already noted by Simonsen (1987), valves of *C. pseudograta* in slides from samples collected in Norway and the Adriatic Sea are scarce. Mostly SV are found, while only one RSV has been found in all studied slides kept at the Hustedt Collection. Samples containing *C. pseudograta* collected in the Tahiti Island contained several SV, the RSV being more scarce and difficult to find.

Our own measurements on the striae density of the SV of *C. pseudograta* are slightly higher than those provided by Hustedt (1933). In turn, the RSV of Tahiti specimens has a higher density of striae than the neotype (table 2). Except for this difference, all valve features of both neotype and Tahiti specimens match well (figs 4–7.) Because of this and due to the scarcity of RSV occurring in both materials, we prefer to assign the RSV of specimens from Tahiti to *C. pseudograta*. We hope that current research on the diatom community from the Society and other archipelagos from the tropical Pacific Ocean (e.g. Riaux-Gobin et al. 2014) will help to solve this morphometric issue.

Frustule features of *Cocconeis grata* and *Cocconeis pseudograta* compared with morphologically related species

Crista marginalis – A common feature to both *C. grata* and *C. pseudograta* SV is the occurrence of a *crista marginalis* (figs 3C & D, 5C & D, 6A). Located on the external face of SV, it marks the beginning of the short and steep SV mantle of the two studied species, and separates the marginal row of processes of the SV of *C. pseudograta* (fig. 5C & D). So far only a few *Cocconeis* taxa are known to possess a *crista marginalis*. It surrounds the entire valve margin in the SV of *Cocconeis peltoides* Hustedt (Riaux-Gobin et al. 2011b), while it is submarginal in its location in the SV of *Cocconeis coronatoides* Riaux-Gobin and Romero and marks the limit where the monoseriate striae become bi- to triseriate (Riaux-Gobin et al. 2010, 2011b). The high *crista marginalis* of the RSV of *Cocconeis costata* var. *hexagona* Grunow limits the valve surface from a steep mantle (Romero & Rivera 1996).

Areolae and depressions of the SV – The outline and density of areolae are features that allow distinguishing between both taxa. Poroids of *C. grata* SV are mostly transapically elongated on the external valve face (fig. 3C & D), while those of *C. pseudograta* SV are roundish and smaller (figs 5C & F, 6B & D).

The outline of the depressions on the external SV of *C. grata* resembles regular areolae (fig. 3A). On the internal SV side (fig. 3B & E), these depressions do not show any aperture (fig. 3E). The depressions of *C. grata* are easily distinguishable with light microscopy as smaller and less reflective than regular SV areolae (fig. 2A & B). The external face of the SV of *C. pseudograta* has no depressions (granules are present, see discussion below).

Marginal row of inwardly occluded processes – A marginal row of inwardly occluded processes is present in the narrow SV mantle of *C. pseudograta* (figs 5C, D & F, 6D & E), but absent in the SV of *C. grata* (fig. 3A–D). Rimoportulae frequently occur in centric and non-raphid diatoms, while they are less common in raphid genera (e.g. in *Eunophora* Vyverman, Sabbe & D.G.Mann, Vyverman et al. 1998, and *Peronia* Bréb. & Arnott *ex* Kitton, Round et al. 1990). The tiny rimoportulae of *C. pseudograta* SV are arranged in a marginal row along the valve margin and are denser than the striae (55–60 processes/10 µm vs. 20–24 striae/10 µm in the neotype; 45–50 striae/10 µm in specimens from Tahiti). On the external SV face, a pierce embossed structure covers the process and a tiny, circular aperture, facing the marginal

ridge, is present (fig. 5D). A star-like domed velum in seen on the internal side (fig. 6D & E).

Morphologically, *C. pseudograta* rimoportulae resemble those already reported in other *Cocconeis* species. The internal openings of the rimoportulae in *Cocconeis peltoides* var. *archaeana* Riaux-Gobin & Compère are either closed by an helicoidal star-like plug – resembling those of *C. pseudograta* – or an open crater-like structure as in *Cocconeis peltoides* Hust. (Riaux-Gobin et al. 2011b). In the SV of *Cocconeis germainii* Riaux-Gobin, Witkowski & O.E.Romero, the processes are very small and appear as raised small domes (Riaux-Gobin et al. 2007). A common feature to all cited species is the regular arrangement over the SV margin and the external aperture by a minute pore. As observed in *C. pseudograta*, common features of the processes of *C. peltoides*, *C. peltoides* var. *archaeana* and *C. germainii* are the small size (diameter = 0.10–0.20 µm) and the internal opening is not lipped.

Granules on the external face of *C. pseudograta* SV – The occurrence of small, almost conical, circular-outlined granules is observed on the external side of the SV of *C. pseudograta*, while they are absent in *C. grata*. Present in both type and Tahiti Island specimens, granules of *C. pseudograta* are widespread throughout the external SV, while they become less dense in the central valve and along the sternum (figs 5A & B, 6A & B).

The location and arrangement of granules on the SV of *Cocconeis* seem to be species-specific. On the SV of *Cocconeis contermina* A.W.F.Schmidt, they are irregularly scattered among the alveoli, while are only present on the lanceolate axial area of the SV of *Cocconeis materoxanae* De Stefano (De Stefano & Romero 2005). A particular sort of granules are observed on the SV of *Cocconeis distans* Gregory, described as repeated protuberances opening on the top of transapically elongated poroids (De Stefano et al. 2006). The SV of *C. pseudomarginata* Gregory show irregularly arranged granules scattered over the external valve surface (Romero & Navarro 1999). A few granules, irregularly arranged along the interestriae are seen on the SV of *Cocconeis pinnata* Gregory (Riaux-Gobin et al. 2014). The SV of the tropical *Cocconeis coronatoides* Riaux-Gobin & Romero has granules surrounding groups of areolae on the valve face, and smaller granules on the mantle (Riaux-Gobin et al. 2010, 2011a).

Occurrence and biogeography of *C. grata* and *C. pseudograta*

Schmidt (in Schmidt et al. 1874–1959) provided no accurate information on a particular sampling location in the Bay of Campeche (Mexico, the type locality of *C. grata*), nor if a particular kind of substratum was sampled. This Bay locates in waters of the southern Gulf of Mexico (Caribbean Sea) and is bordered on its right side by the Peninsula of Yucatan (c. 19–21°N 89°W). The location of the first record of *C. grata* allows assuming that this diatom is tropical/subtropical in its temperature preferences. Unfortunately, we did not have the opportunity to study material left by Cleve and/or Peragallo & Peragallo. Therefore, we are not able to confirm whether these researchers actually studied both taxa and whether its distribution is as geographically wide as quoted by Hustedt (1933, 1939).

In his description of *C. pseudograta* (as *C. grata* A.W.F.Schmidt), Hustedt (1933) included neither the sample localities nor the corresponding slides and adopted the criterion of species distribution given by Cleve (1895). For this reason Simonsen chose the slide Zt4/62 (Hustedt Collection) with material collected in Grip (Norway) as the neotype. As properly commented by Simonsen (1987), Hustedt (1933) failed to give a type locality for *C. pseudograta*. Grip, the neotype locality proposed by Simonsen (1987), is an archipelago consisting of tenths of islets and skerries about 15 kilometers into the Norwegian Sea northwest off Kristiansund (c. 63°N 7°W). Hustedt (1933) characterized *C. pseudograta* as a littoral form with a wide range of latitudinal distribution between the Mediterranean Sea and Island. He also stated *C. pseudograta* to be quite abundant in the Mediterranean Sea. Interestingly, we found only one valve in the permanent slide labeled 'San Pietro dei Nembi' (Croatian coast, Northern Adriatic Sea). Our finding of *C. pseudograta* in a coral reef lagoon and on a scrape of *Holothuria atra* (Black Sea cucumber) from Tahiti allows expanding its distribution into the tropical Pacific Ocean.

Only a few published records of *C. pseudograta* are known. In their account of the diatom community thriving in coral sand from the Mascarenes (Western Indian Ocean), Riaux-Gobin et al. (2011c) identified *C.* cf. *pseudograta*. The recent record of *C. pseudograta* associated with the monocotyledon *Posidonia oceanica* Delile in the Eastern Mediterranean (Majewska et al. 2014) matches the low-latitude distribution of this taxon. Hustedt gave no detailed information on which kind of substratum the valves of *C. pseudograta* occurred in northwestern German coastal waters. Considering that Hustedt (1939) studied shallow mudlands, we speculate *C. pseudograta* to be epylithic and/or epipsammic.

Hustedt (1939) argued that *C. grata* and *C. pseudograta* differed in their main geographic distribution. He considered *C. grata* as '(North)American' in its occurrence while *C. pseudograta* was 'European'. Therefore, they were also separated on the basis of their distribution. Based on the occurrence of *C. pseudograta* in coastal waters of the Tahiti Island, we argue that its designation as 'European form' is no longer valid.

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