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## *Navicula dermochelycola* sp. nov., presumably an exclusively epizoic diatom on sea turtles *Dermochelys coriacea* and *Lepidochelys olivacea* from French Guiana

by

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### Abstract

Epizoic diatom communities were studied on four turtle species (*Chelonia mydas* L., *Eretmochelys imbricata* L., *Lepidochelys olivacea* Eschscholtz and *Dermochelys coriacea* Vandelli) from the Eastern Caribbean, the Equatorial West Atlantic and the South Pacific. In the present study, we focused on one taxon of *Navicula* Bory de Saint-Vincent, described here as a new taxon, which was found on seven individuals of *Dermochelys coriacea* and one individual of *Lepidochelys olivacea* in French Guiana. The new *Navicula* taxon was found neither on *Chelonia mydas* (83 specimens examined) nor *Eretmochelys imbricata* (13 specimens examined). Furthermore, the new taxon appears to have a restricted biogeography, as so far it has only been reported from French Guiana. A species of *Navicula* is reported for the first time as a supposedly exclusive epizoic taxon. *Navicula dermochelycola* sp. nov. is characterized in detail; its ultrastructure is described and compared with other members of *Navicula*.

**Key words:** Rotifera, eutrophic lake, effects of Dreissena's, fish, Crustacea



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## Introduction

Epizoic diatoms have been described from diverse hosts (from invertebrates to vertebrates) and diverse environments (from freshwater to marine; see i.e. Wuchter et al. 2003; Wetzel et al. 2010; Riaux-Gobin & Witkowski 2012; Romagnoli et al. 2014; Frankovich et al. 2016; Riaux-Gobin et al. 2017 and references therein). Several genera have been described as presumably obligately epizoic (or potentially commensal, even if no specific study on real relationships between the host and the diatom has ever been published), including *Epipellis* R.W. Holmes, *Bennettella* R.W. Holmes (Holmes 1985; Holmes & Nagasawa 1995; Denys & Van Bonn 2001; Denys & De Smet 2010), *Epiphialina* R.W. Holmes, Nagasawa & Takano, *Plumosigma* T. Nemoto, *Tursiocola* R.W. Holmes, Nagasawa & Takano (Nemoto 1956; Holmes et al. 1993; Denys 1997), *Chelonicola* Majewska, De Stefano & Van de Vijver and *Poulinea* Majewska, De Stefano & Van de Vijver (Majewska et al. 2015) and *Medlinella* Frankovich, Ashworth & M.J. Sullivan (Frankovich et al. 2016). At the species level, several diatoms have also been described as possibly obligate epizoic, including *Tripterion kalamensis* R.W. Holmes, S. Nagasawa & Takano (1993) and *Tripterion philoderma* R.W. Holmes, S. Nagasawa & Takano (1993), *Pseudofalcula hyalina* (Takano) F. Gómez, L. Wang & S. Lin (Takano 1983; Gomez et al. 2018), *Pseudohimantidium pacificum* Hustedt & Krasske in Krasske (Krasske 1941; Fernandes & Calixto-Feres 2012), *Luticola deniseae* Wetzel, Van de Vijver & Ector (Wetzel et al. 2010), and *Mastogloia sterijovskii* A. Pavlov, Jovanovska, C.E. Wetzel, Ector & Levkov (Pavlov et al. 2016).

In 2014–2018, a total of 142 sea turtles from the Equatorial West Atlantic, the Caribbean Basin and the South Pacific were sampled. In the course of these studies, presumably an exclusively epizoic species of *Navicula* was encountered. The purpose of the presented report is to describe this new taxon based on light and scanning electron microscopy observations.

## Materials and methods

### Projects and surveyed zones

The study is part of FEDER Martinique (Fonds Européen de Développement Régional), ANTIDOT (Association of News Tools to Improve the understanding of the Dynamic Of Threatened marine turtles, Mission pour l'Interdisciplinarité), DEAL Martinique (Conventions 2012/DEAL/0010/4-4/31882

and 2014/DEAL/0008/4-4/32947), the ODE Martinique (Convention 014-03-2015) and CNRS/IPHC (Centre National de la Recherche Scientifique/Institut Pluridisciplinaire Hubert Curien) programs for Martinique (Caribbean Basin) and French Guiana (Equatorial West Atlantic), and part of several projects concerning turtles in the Society Archipelago (South Pacific) coordinated by Te Mana O Te Moana (Observatoire des Tortues marines en Polynésie française). Details of sampling sites are presented in Table 1.

### Ethic statements

The protocols applied in Martinique and French Guiana were approved by Conseil National de la Protection de la Nature (CNP, <http://www.conservation-nature.fr/acteurs2.php?id=11>). The protocol applied in the South Pacific was approved by French Polynesia (Permit number 2157 of Ministère de la promotion des Langues, de la culture, de la Communication et de l'Environnement granted to "Te Mana O Te Moana").

### Field and Laboratory Methods

Epizoic taxa associated to the turtle carapace or soft shell were superficially scraped (sampling period 2014–2018) from small surfaces with a blade. One hundred and forty-two samples from four turtle species [*Chelonia mydas* L. (CM), *Eretmochelys imbricata* Linnaeus (EI), *Lepidochelys olivacea* Eschscholtz (LO) and *Dermochelys coriacea* Vandelli (DC)] were examined (Table 1). This material was kept in Eppendorf tubes® and preserved with ethanol. Scanning electron microscopy (SEM) stubs were prepared with some drops of this material, filtered on a Whatman® Nuclepore filter (1 µm pore size, 13 mm ø) and rinsed twice with deionized water (Milli-Q®) to remove salts. Filters were air-dried, mounted onto aluminum stubs and then coated with gold-palladium alloy (EMSCOP SC 500 sputter coater) and examined under a Hitachi S–4500 SEM operated at 5 kV, calibrated with Silicon grating TGX01 (C2M, Perpignan, France). For light microscopy (LM), the material was washed with distilled water to remove salts, treated with 30% H<sub>2</sub>O<sub>2</sub> for 2 h at 70°C to remove organic matter, rinsed several times in distilled water, alcohol-desiccated and mounted on glass slides using Naphrax®. Diatom slides were examined using Zeiss Axiophot 200 with differential interference contrast (DIC) optics and photographed with a Canon PowerShot EOS1000D digital camera (CRIOBE–USR 3278, Perpignan, France) and Zeiss Axio

Table 1

## Sampled turtles and geographical location

oceanic basin	location	species	nesting or subadult	juvenile	latitude	longitude	
Caribbean Sea (Martinique)	Grande Anse d'Arlet	CM		8	14°30'10.95"N	61°05'13.01"W	
	Anse du Bourg d'Arlet	CM		22	14°29'13.43"N	61°04'58.89"W	
		EI		1			
	Macabou	DC	1		14°29'55.06"N	60°49'25.42"W	
	Les Sâlines	EI	1		14°23'45.55"N	60°52'14.74"W	
	Anse Noire	EI			1	14°31'42.47"N	61°5'20.36"W
		CM			1		
	Prêcheur	EI			1	14°49'60"N	61°12'0"W
		CM			1		
	Carbet	CM			4	14°42'0"N	61°10'60"W
Saint Pierre	CM			2	14°43'60"N	61°10'60"W	
	EI			1			
Petite Anse d'Arlet	CM			1	nd	nd	
Equatorial Atlantic Ocean (French Guiana)	Yalimapo	DC	16		5°44'47.96"N	53°56'37.36"W	
		CM	13				
	Aztèque	CM	4		5°41'12.57"N	53°43'49.07"W	
		DC	1				
	Cayenne	DC	3		4°55'10.54"N	52°16'5.31"W	
LO		6					
South Pacific Ocean (French Polynesia)	Te Mana O Te Moana (Moorea)	CM		2	17°29'39.639"S	149°52'13.527"W	
	Tiaraunu (Tetiaroa)	CM	7		16°59'21.5"S	149°34'48.1"W	
	Oroatera (Tetiaroa)	CM	1		16°59'37.4"S	149°32'24.0"W	
	Onetahi (Tetiaroa)	CM	9		17°01'17.9"S	149°35'45.2"W	
	Tahiti	EI		1	nd	nd	
	Afareaitu (Moorea)	EI		2	17°33'S	149°48'W	
	Nu'uroa (Moorea)	CM		1	17°32'5.793"S	149°54'10.325"W	
	Haapiti (Moorea)	CM		1	17°33'56.2"S	149°52'09.9"W	
	Atiha (Moorea)	CM		1	17°35'09.7"S	149°50'26.8"W	
	Tetiaroa	EI			2	nd	nd
		LO	1			nd	nd
	Tikehau	CM		1	15°S	148° 10'W	
	Moorea Lagoon	EI		1	nd	nd	
	Maatea (Moorea)	EI		1	17°35'13.92"S	149°48'20.7036"W	
	Opunohu (Moorea)	CM		1	17°30'28.7352"S	149°51'23.9328"W	
	Temae (Moorea)	CM	1		nd	nd	
	Moorea	EI	1		nd	nd	
	Papetoai (Moorea)	CM		1	nd	nd	
Horoatera (Tetiaroa)	CM	1		nd	Nd		

CM = *Chelonia mydas*; EI = *Eretmochelys imbricata*; LO = *Lepidochelys olivacea*; DC = *Dermochelys coriacea*

Imager M2 (Carl Zeiss, Jena, Germany) with a 100 × Plan Apochromat oil immersion objective (N.A. = 1.46) equipped with DIC (University of Szczecin, Institute of Marine and Environmental Sciences, Szczecin, Poland).

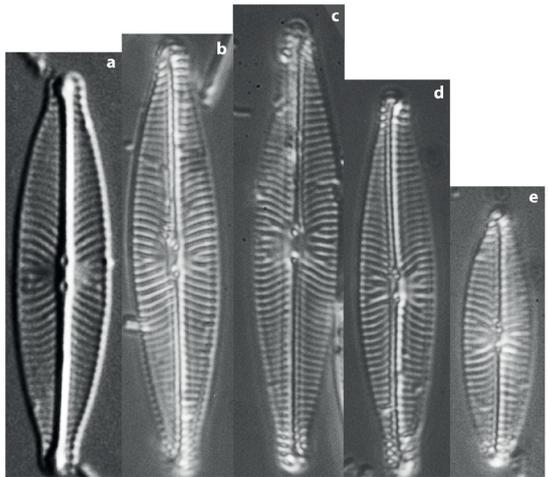
## Results

***Navicula dermochelycola*** Witkowski, Kociolek & Riaux-Gobin sp. nov. LM Fig. 1a–e, SEM Figs 2–4, Table 2.

**Description:** Valves small (15–25 µm long, 4–5 µm wide), lanceolate to linear-lanceolate, with subrostrate

to capitate apices (with varied shapes of apices in smaller specimens; Fig. 1a–e, Fig. 2a–e). In LM, a distinct terminal hyaline area is visible at each apex (Fig. 1a–e), corresponding to the helictoglossa and adjoining structure detailed with SEM (Fig. 3b).

**External view (SEM):** Raphe filiform, axial area very narrow and linear. Central area moderately expanded, round to irregular, more or less symmetrical, delineated by alternately longer and shorter striae on both sides of the raphe (Fig. 2a–e). Proximal raphe endings distant, slightly expanded and bent toward the valve primary side (opposite to distal raphe endings; Fig. 3a–c). Distal raphe ends hooked, typical



**Figure 1a–e (LM)**

*Navicula dermochelycola* sp. nov. Note the alternating longer and shorter striae around the central area. Scale bar = 10  $\mu\text{m}$  for all images

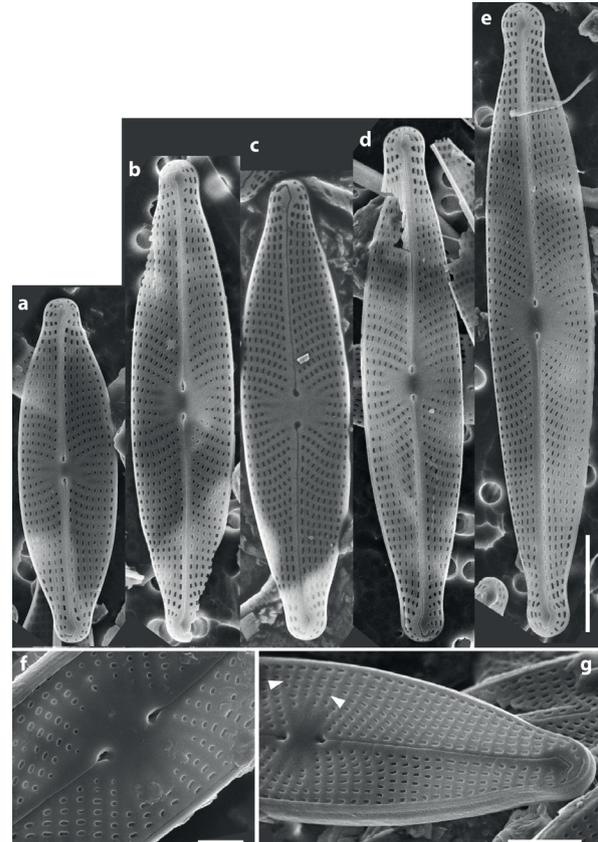
for *Navicula* sensu stricto (Fig. 2c). Transapical striae radiate, in the middle strongly arched around the central area, becoming slightly convergent toward the apices. Striae equidistant, 24–30 in 10  $\mu\text{m}$ . Transapical striae composed of apically elongate areolae, aligned along a rhombic pattern, ca. 50 in 10  $\mu\text{m}$ . Valve surface flat with oblong striae forming areolae positioned between slightly raised virgae (Fig. 2g).

**Internal view (SEM):** Raphe slit opening laterally along an elevated rib (Fig. 4a). Internal proximal raphe endings close to each other and simple (Fig. 4c). Raphe terminating distally as an obliquely-positioned helictoglossa surrounded by an elevation or thickened terminal nodule (Fig. 4b), corresponding to the hyaline apical area observed in LM (see above). Girdle narrow, composed of several open and plain copulae. The margin of the valvocopula externally ornamented with one row of short transapical and elongate puncta or slits (7 in 500 nm, Fig. 3d), absent on the head pole (= closed pole) of the copula.

**Holotype:** LM slide BM 101 943 (National History Museum, London, U.K.); illustrated in Figure 1.

**Isotypes (here designated):** Slide SZCZ 25795 in collection A. Witkowski (Institute of Marine and Environmental Sciences, Szczecin, Poland).

**Type locality:** Nesting *Dermochelys coriacea* '103' from the Equatorial West Atlantic (French Guiana, locality Yalimapo); 5°44'47.96"N; 53°56'37.36"W. Collector: Damien Chevallier.



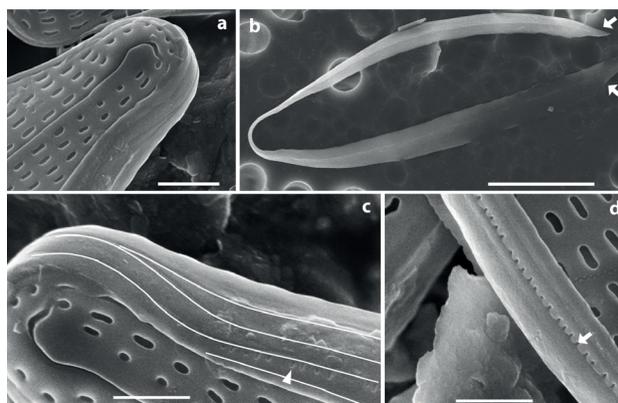
**Figure 2a–g (SEM)**

*Navicula dermochelycola* sp. nov.: (a–e) External views of complete valves in valve view, with capitate apices; (f, g) Large and irregular central area with alternating longer and shorter striae (g arrowheads). Note the decussate pattern of areolae. Scale bars 4  $\mu\text{m}$  (a–e); 1  $\mu\text{m}$  (a, f); 2  $\mu\text{m}$  (g)

**Etymology:** The epithet *dermochelycola* was given referring to the turtle host, *Dermochelys coriacea* ("dermochely" from the turtle host *Dermochelys*, and "cola" from colere, meaning living in Latin).

**Distribution:** Found on seven nesting DC from French Guiana (localities: Yalimapo and Aztèque) out of 20 specimens analyzed. Found also on one nesting LO (locality: Cayenne, French Guiana) out of six specimens analyzed. Apparently absent on other turtle species (83 CM, 13 EI) and observed only in French Guiana.

**Taxonomic remarks:** This diatom is unique among many species of *Navicula* due to the areolae of striae, which are more ellipsoidal in this new taxon. Our new species resembles *Navicula rostellata* Kützing (Table 2), but the central striae are alternately longer and shorter (Fig. 2g), and the new species is distinguished



**Figure 3a–d (SEM)**

*Navicula dermochelycola* sp. nov. External views: (a) Capitate apex with visible convergent striae. (b) Free open copula without ornamentation. (c) Apex with open copulae and one row of transapically-elongate and irregularly-spaced puncta (c arrowhead). Note that puncta are not present on copulae at the apex. (d) Copula (probably the valvocopula) with a row of irregular short puncta (d arrow). Scale bars: 3  $\mu\text{m}$  (b); 1  $\mu\text{m}$  (a); 700 nm (c); 600 nm (d)

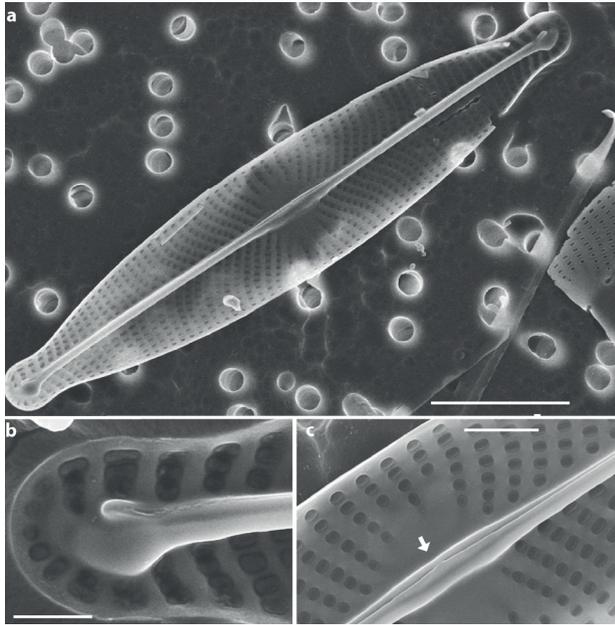
by a higher stria density compared to *N. rostellata* (28 in 10  $\mu\text{m}$ , versus 12–14 in *N. rostellata*). *N. rostellata* is a freshwater taxon (Kützing 1844; Lange-Bertalot 2001; Potapova & Kociolek 2011), even if also reported from brackish and marine habitats. The new species also shares some similarities with *Navicula salinarum* Grunow, but is distinguished by a higher stria density compared to *N. salinarum* (13–17 in 10  $\mu\text{m}$  in *N. salinarum*) and a different valve shape (roughly elliptical-lanceolate in *N. salinarum*; Krammer & Lange-Bertalot 1986; Patrick & Reimer 1966; Kociolek 2011). Widespread *Navicula capitatoradiata* Germain (Germain 1981; Rushforth & Spaulding 2010) also shares some similarities with our new species, but is lanceolate in shape, has a lower stria density and a smaller central area compared to our taxon (Germain 1981; Gasse 1986; Lange-Bertalot 2001). *Navicula fucicola* Taasen (Taasen 1975), which is endophytic in “the mucilage of the apical slit of *Fucus vesiculosus* L.” (op. cit.: p. 5), also shares some similarities with the new taxon in terms of size and striae densities (12–22  $\mu\text{m}$  in length, 5–7  $\mu\text{m}$  in width, 20–24 striae in mid-valve, 24–28 near apices), but the latter is broadly elliptic-lanceolate in contrast to *N. dermochelycola* sp. nov., which is lanceolate to linear-lanceolate.

**Table 2**

Features of *Navicula dermochelycola* sp. nov. (French Guiana, Yalimapo, DC). Comparison with several other *Navicula* taxa (see text)

	<i>Navicula dermochelycola</i> sp. nov.	<i>Navicula rostellata</i>	<i>Navicula capitatoradiata</i>	<i>Navicula salinarum</i>	<i>Navicula fucicola</i>
n (SEM observations)	21	nd	nd	nd	nd
Valve shape	linear elliptic with capitate apices	linear to linear-lanceolate, strongly capitate, narrow ends	lanceolate, protracted and rostrate apices	elliptical-lanceolate, rostrate apices	lanceolate, rostrate-capitate
Length	15–25 (19 $\pm$ 3.0)	32–50	35–40	30–40	12–22
Width	3.7–5.0 (4 $\pm$ 0.3)	8–10	7–8	9–10	5–7
L/W	4 $\pm$ 0.8	nd	nd	nd	nd
Stria density	24.5–30 (28 $\pm$ 1.6)	11–14 (15)	12–13	14–16	20–24; 24–28 on apices
Areolae on the valve face	radiate to arched striae, apically elongate areolae	striae bent and radiate around the center, apically elongate areolae	radiate to arched striae, apically elongate areolae	striae arched, parallel near apices	striae radiate to arched
Striae around the central area	alternatively short and long	all of the same length	alternatively short and long	alternatively short and long	of irregular length
Central area	distinct, bow-tie/butterfly like, expanded, not really asymmetrical	expanded and asymmetrical	relatively restricted	relatively large, symmetrical	small
Ethology	epizoic, marine	freshwater, cosmopolitan	freshwater	brackish	marine, in the mucilage of the apical slit of <i>Fucus vesiculosus</i>
Reference	present study	Kützing 1844 Lange-Bertalot 2001 Potapova & Kociolek 2011	Germain 1981 Rushforth & Spaulding 2010	Grunow in Cleve & Grunow 1880 Patrick & Reimer 1966 Germain 1981 Kociolek 2011	Taassen 1975

nd - no data; Morphometrics expressed as indicated in the original description or in later compilations. Morphometrics of *N. dermochelycola* sp. nov. expressed as min.–max and mean  $\pm$  SD ( $\mu\text{m}$ , stria density in 10  $\mu\text{m}$ ). SD = standard deviation. n = specimens observed in SEM.



**Figure 4a-c (SEM)**

*Navicula dermochelycola* sp. nov. Internal valve views: (a) Entire valve with strongly arched striae near the central area. (b) Helictoglossa laterally-positioned on the central rib. (c) Asymmetrical position of the proximal raphe endings on the elevated rib (c arrow). Note the ellipsoidal areolae. Scale bars: 5  $\mu\text{m}$  (a); 3  $\mu\text{m}$  (c); 600 nm (b)

The central area in *N. fucicola* is also less expanded than in our new taxon (Taasen 1975). It should be noted that the ornamentation on the valvocopula of *N. dermochelycola* sp. nov. (one row of short slits; Fig. 3c–d) seems unusual for this genus.

## Discussion and conclusion

Behavioral studies of marine turtles have not reported any deaths or diseases due to macro- and micro-flora associated with their carapace and skin, while deaths due to accidental fishing (Finkbeiner et al. 2011 and references therein) or following the ingestion of plastics (Nelms et al. 2015; Wilcox et al. 2018; Yaghmour et al. 2018) are confirmed and listed. Furthermore, studies describing cleaning stations for sea turtles (Sazima et al. 2010) tend to prove that turtles come there to restrict the proliferation of algal epibiont material on their bodies and are not that much inconvenienced by them. We can therefore hypothesize that diatoms (such as *Navicula dermochelycola* sp. nov.) live in a close relationship with

their host and that the term commensalism (no vital damage to one with respect to the other, but shelter for diatoms and perhaps a mode of dissemination) can be used. However, we have used the term “presumably exclusively epizoic” throughout the text.

On the other hand, diatoms as well as macroalgae need light to grow, even if epizoic *Tursiocola* from Manatees was demonstrated to be apochlorotic (Frankovich et al. 2018). Thus, an excessive sediment discharge, such as the one by the Amazon River to the Guiana coast, may affect the acclimation of diatoms on turtles, possibly explaining the very poor colonization on turtles in that region (Riaux-Gobin et al. in revision). Moreover, DC is considered to be the most deep-diving sea turtle (Bjorndal 1996; Dodge et al. 2011), which probably prevents effective growth of diatoms.

Nevertheless, 33% of the DC individuals examined in this study host a specific diatom (*Navicula dermochelycola* sp. nov.) that has never been found elsewhere, except for one LO individual from the same environment (as mentioned above). The acclimation of this taxon may also be due to the specific structure of the leather-like carapace of DC.

As a result of the present survey of sea turtles from tropical sectors of the Atlantic and Pacific oceans, a new benthic diatom taxon is reported, which so far has never been found in another environment. *Navicula dermochelycola* sp. nov. was only present in French Guiana and mainly hosted by DC.

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